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Atopic Dermatitis

Research and Clinical Updates and Perspectives

Edited by
Stamatis Gregoriou

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Editor

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About the Editor

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Stamatios Gregoriou is an associate professor of Dermatology in the National and Kapodistrian University of Athens. He has published more than 200 papers in peer review indexed journals and more than 20 book chapters. His academic interests include atopic dermatitis, contact dermatitis, chronic spontaneous urticaria, hyperhidrosis and nail disorders. He is a co-author of the EADV/EDF 2022 living guidelines for atopic dermatitis

Preface

Our understanding of Atopic dermatitis is evolving. Available literature information is increasing geometrically each year. The quality of data available, allows us to understand pathogenesis better than any time before. In addition, an arsenal of novel treatments that utilize the amendment of the pathogenic pathways of inflammation and epidermal barrier restoration have been available in the last years. This special issue addresses both pathogenetic and treatment recent knowledge for students and healthcare professionals interested in atopic dermatitis

Stamatis Gregoriou
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Editorial

Atopic Dermatitis: Sailing beyond the Sunset with a Multitude of Novel Treatments

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Atopic eczema or dermatitis (AD) is a chronic pruritic inflammatory cutaneous disorder with an incidence up to 20% in children and 10% in adults depending on region and ethnicity. Due to severe pruritus, the chronicity of skin lesions, AD has been shown to have a vast psychosocial burden for patients. The AD patients report heavily decreased quality of life, raised stigmatization level and frequently develop secondary psychiatric comorbidities, such as depression and anxiety. The risk of suicidal thoughts and attempts is significantly increased [1,2]. Moreover, it is clear that not only the sick individual suffers but the disease affects also the family members [3]. Taking the above into consideration we are dealing with a common and important clinical problem. Understanding the AD pathogenesis in depth, developing new treatment options will definitely help holistic approaches to AD patients.

Atopic is a Greek adjective meaning literally "out of place" and teleologically "unexplained". Eczema is a Greek noun meaning "due to boiling". Dermatitis is also Greek, meaning cutaneous inflammation. Although we describe "Greek" only in the definition of Isocrates, who considered Greeks to be all those sharing in Greek education, Edward Perry was particularly insightful when asked by Coca and Cooke to help denominate the hypersensitivity to environmental allergens. A lot about the pathogenesis of atopic dermatitis still remains poorly illuminated. However, in this new era, rapid advances in basic science, dermatology and allergology research, along with several new therapeutic agents, set out a more optimistic approach for both clinicians and patients.

Epidermal components of AD pathogenesis include loss of function filaggrin gene polymorphisms; however, filaggrin variants are not the sole etiology for filaggrin downregulation in AD skin, and 40% of carriers of filaggrin null alleles never experience eczema. Filaggrin mutations have been reported to be associated with AD severity and persistence in adulthood, suggesting that filaggrin mutations represent a particular endotype [4]. The effect of Th2 inflammatory response on filaggrin downregulation and the normalization of filaggrin expression in patients treated with monoclonal antibodies or JAK-inhibitors is a field under exploration at this time. In addition, the impact of the new agents on other barrier-related proteins, such as involucrin and loricrin, should be part of an additional scope of investigation.

AD epidermis has decreased lipids, particularly ceramides, in both lesional and non-lesional skin as well as increased free fatty acids. IL-4 has been reported to downregulate ceramides synthesis via signaling through the STAT6 pathway. Alterations in the lipid composition of the epidermis can impair innate immunity and result in microbial superinfection in AD skin [5]. The role of the changes in the lipid microenvironment, such as Th2 inflammation via activation of CD1a antigen presenting proteins on Langerhans cells in AD, remains largely under-investigated. Although most moisturizers are beneficial in reducing the number of AD flares, evidence does not support that one moisturizer is better

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than another. In addition, despite early studies on the use of cleansers and moisturizers to prevent AD in infants and children reporting positive trends, more recent large-scale studies have failed to confirm these results.

Genetic and acquired defects of tight junctions have been reported to play a crucial role in barrier dysfunction in AD patients. Decreased expression of claudin 1 has been associated with an increase in serum biomarkers of the Th2 driven response, suggesting cross-talk between the epithelial barrier and immunological inflammation in AD [6]. Areas of current investigation include the correlation between impaired tight junctions and antigen presentation, the interaction between claudin and filaggrin expression, and the effect of claudin expression on the proliferation and differentiation of keratinocytes.

Microbiota dysbiosis is emerging as an important feature of AD pathogenesis. *Staphylococcus aureus* is the principal pathogen that triggers the host immune system-related inflammation in the acute phase. In addition, the chronic persistence of *S. aureus* on eczematous skin lesions has been associated with the identification of staphylococcal biofilm communities on the skin of patients with AD. Recent reports suggest that *Staphylococcus epidermidis* might not be an innocent bystander but in certain conditions might also contribute to the inflammatory reaction in AD pathogenesis. *Cutibacterium acnes* enhances *S. aureus* cytolytic activity and the subsequent production of pro-inflammatory cytokines. Malassezia allergens can trigger a specific IgE response as part of AD pathogenesis. The role of *Candida albicans*, which has been found to often be part of the microbiota of AD, patients is not clear [7]. Novel therapeutic interventions, including probiotic and prebiotic preparations, as well as skin microbiota transplantation, are an emerging intervention to regulate microbiota dysbiosis, but evidence of their efficacy is still being evaluated.

AD is associated with food allergy, asthma, and allergic rhinitis and the gradual transition of one atopic disease into another has been denominated as the atopic march. Defining phenotypes and endotypes within the AD patient population in order to predict the future development of atopic march is an evolving effort. Although dupilumab is the only agent at this time that has an indication regarding AD, asthma, chronic rhinosinusitis with nasal polyposis, and eosinophilic esophagitis, the efficacy of the other novel therapeutic agents for AD, in the subpopulations of AD patients with other atopic comorbidities, is also a field of current clinical research interest.

The plethora of phenotypes and endotypes has raised investigational interest in the overlapping of AD and psoriasis, once considered to be two opposing immunopathogenic Th2 and Th1 disorders. Asian, pediatric, and intrinsic types of AD involve Th17, which is an important inflammatory pathway in psoriasis pathogenesis. As we investigate non-atopic comorbidities of AD, the shared patterns of comorbidities, such as metabolic syndrome or cardiovascular disorders, are considered, although evidence at this time is controversial. Clinical diagnosis, especially in children, might also be difficult as 20% of the cases might lack typical lesions and present a combination of both disease features, the so-called psoriatic eczema. In addition, plaque psoriasis restricted on the limbs might resemble nummular eczema while erythrodermic psoriasis might have an immunologic overlap of both Th17 and Th22 cells.

Animal model data could be helpful in understanding endotypes, phenotypes, and even the pharmacological properties of novel agents in a pre-clinical setting. The translational value of such data is still under debate, as there might be an overestimation of the potential efficacy of novel agents following encouraging results of in vivo studies in animal models. Published data of animal model characteristics might be helpful in selecting the appropriate model for a specific study purpose. The investigational aim should be to increase the predictability and translatability of animal model results to human clinical studies.

New developments of treatment modalities have recently resulted in the approval of first topical JAK-inhibitor—ruxolitinib cream for patients suffering from mild to moderate AD. This is a crucial step in topical AD therapy; for decades we only had the possibility of using topical corticosteroids and topical calcineurin inhibitors. With a good safety profile, ruxolitinib cream will definitely be of patients' benefit, improving both pruritus and skin

lesions [8,9]. Several more topical agents like tapinarof, difamilast, and roflumilast are also in advanced stage of development. Long term efficacy and safety of these formulations and comparison with current topical treatments are areas that are lacking data at this moment.

EADV/EDF guidelines for the treatment of AD in patients' candidates for systemic therapy suggest both classical systemic therapy as well as anti-IL4 and/or IL13 monoclonal antibodies and JAK-inhibitors as the first line treatment for the disorder. Even though the new agents provide long-term control of the disorder with a favorite safety profile, there are still matters under debate concerning their use, particularly from the view of the payers. The intermittent employment of the novel agents, transition from the higher dosage to the lower in JAK inhibitors, as well as class side effects, particularly in the light of the FDA warning, are questions still under discussion.

Studies have investigated the association of serum biomarkers for AD with treatment outcomes. An association does not necessarily allow prediction, and biomarkers have not currently been proven useful for patient stratification for systemic therapy for AD in published literature data. A recent consensus prioritized reliability, clinical validity, a high positive predictive value, prediction of the therapeutic response, and disease progression as potential biomarkers in AD and psoriasis [10]. Striving for ideal biomarkers in inflammatory cutaneous disorders is an ongoing effort, compromised by significant obstacles mainly associated with validity that does not allow the utilization of biomarkers in a way similar to their use in the disease of cancer. However, cost-effectiveness and the subsequent reimbursement of the novel agents could benefit considerably from the establishment of reliable biomarkers and are expected to drive further research in the field. Machine learning might offer significant insights in utilizing the available data of serum biomarkers.

Even as the present has revolutionized our view and treatment of AD, the future holds even more hope for the patients; multiple clinical trials and subpopulations analysis are currently underway. The main challenge at this time is to improve access to the novel therapies for an increased number of patients. Registries, biomarkers, and shared decisions by all stakeholders will be needed to attain this goal and even offer individualized treatment to AD patients.

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References

1. Kaaz, K.; Szepietowski, J.K.; Matusiak, L. Influence of Itch and Pain on Sleep Quality in Atopic Dermatitis and Psoriasis. *Acta Derm. Venereol.* **2019**, *99*, 175–180. [CrossRef] [PubMed]
2. Dalgard, F.J.; Gieler, U.; Tomas-Aragones, L.; Lien, L.; Poot, F.; Jemec, G.; Misery, L.; Szabo, C.; Linder, D.; Sampogna, F.; et al. The Psychological Burden of Skin Diseases: A Cross-Sectional Multicenter Study among Dermatological Out-Patients in 13 European Countries. *J. Investig. Dermatol.* **2015**, *135*, 984–991. [CrossRef] [PubMed]
3. Marciniak, J.; Reich, A.; Szepietowski, J. Quality of Life of Parents of Children with Atopic Dermatitis. *Acta Derm. Venereol.* **2017**, *97*, 711–714. [CrossRef] [PubMed]
4. Furue, M. Regulation of Filaggrin, Loricrin, and Involucrin by IL-4, IL-13, IL-17A, IL-22, AHR, and NRF2: Pathogenic Implications in Atopic Dermatitis. *Int. J. Mol. Sci.* **2020**, *21*, 5382. [CrossRef] [PubMed]
5. Pavel, P.; Blunder, S.; Moosbrugger-Martinz, V.; Elias, P.M.; Dubrac, S. Atopic Dermatitis: The Fate of the Fat. *Int. J. Mol. Sci.* **2022**, *23*, 2121. [CrossRef] [PubMed]
6. Yoshida, T.; Beck, L.A.; De Benedetto, A. Skin barrier defects in atopic dermatitis: From old idea to new opportunity. *Allergol. Int.* **2022**, *71*, 3–13. [CrossRef] [PubMed]
7. Hrestak, D.; Matijašić, M.; Paljetak, H.; Drvar, D.L.; Hadžavdić, S.L.; Perić, M. Skin Microbiota in Atopic Dermatitis. *Int. J. Mol. Sci.* **2022**, *23*, 3503. [CrossRef] [PubMed]
8. Papp, K.; Szepietowski, J.C.; Kircik, L.; Toth, D.; Eichenfield, L.F.; Leung, D.Y.; Forman, S.B.; Venturanza, M.E.; Sun, K.; Kuligowski, M.E.; et al. Efficacy and safety of ruxolitinib cream for the treatment of atopic dermatitis: Results from 2 phase 3, randomized, double-blind studies. *J. Am. Acad. Dermatol.* **2021**, *85*, 863–872. [CrossRef] [PubMed]

9. Smith, P.; Yao, W.; Shepard, S.; Covington, M.; Lee, J.; Lofland, J.; Naim, A.; Sheth, T.; Parikh, B.; Yeleswaram, S. Developing a JAK Inhibitor for Targeted Local Delivery: Ruxolitinib Cream. *Pharmaceutics* **2021**, *13*, 1044. [CrossRef] [PubMed]
10. Ziehfreund, S.; Tizek, L.; Hangel, N.; Fritzsche, M.; Weidinger, S.; Smith, C.; Bryce, P.; Greco, D.; Bogaard, E.; Flohr, C.; et al. Requirements and expectations of high-quality biomarkers for atopic dermatitis and psoriasis in 2021—A two-round Delphi survey among international experts. *J. Eur. Acad. Dermatol. Venereol.* **2022**. *online ahead of print*. [CrossRef] [PubMed]

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Article

Dupilumab Improves Skin Barrier Function in Adults with Atopic Dermatitis: A Prospective Observational Study

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Abstract: Epidermal barrier dysfunction plays an important role in atopic dermatitis (AD). The difficulty of objectively assessing AD severity and the introduction of new biologicals into clinical practice highlight the need to find parameters to monitor clinical outcomes. The aim of this study is to evaluate the impact of dupilumab on skin barrier function and compare it with other treatments in patients with AD. A prospective observational study was conducted in adults with AD treated with topical corticosteroids (TCS), cyclosporine, or dupilumab. The main outcome measures after 16 weeks of treatment were Eczema Area and Severity (EASI)-50 (50% improvement in EASI), and transepidermal water loss (TEWL)-50 (50% improvement in TEWL). Forty-six patients with AD were included in the study. The proportion of patients who achieved EASI-50 at week 16 was significantly higher in patients receiving dupilumab (81.8% vs. 28.6% vs. 40%, $p = 0.004$). In eczematous lesions, TEWL decreased in patients receiving dupilumab (31.02 vs. 12.10 $g \cdot h^{-1} \cdot m^{-2}$, $p < 0.001$) and TCS (25.30 vs. 14.88 $g \cdot h^{-1} \cdot m^{-2}$, $p = 0.047$). The proportion of patients who achieved TEWL-50 at week 16 was higher for dupilumab than for cyclosporine or TCS. Temperature only decreased in the dupilumab group. Stratum corneum hydration increased in eczematous lesions and non-involved skin only in patients with dupilumab. In conclusion, dupilumab improves skin barrier function in patients with AD better than TCS or cyclosporine, both in eczematous lesions and in non-lesioned skin.

Keywords: atopic dermatitis; dupilumab; skin barrier; transepidermal water loss

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1. Introduction

Atopic dermatitis (AD) is a chronic cutaneous inflammatory disease caused by genetic and environmental factors [1,2]. It is one of the most prevalent skin diseases, with a prevalence ranging from 0.96% to 22.6% in children and from 1.2% to 17.1% in adults [3], with higher prevalence in industrialized countries [4]. Clinically, it is characterized by recurrent and itchy eczematous lesions, excoriations, scaling and dry skin [1,5]. It is also a disease that greatly impairs the quality of life of patients and their cohabitants [6].

Epidermal barrier dysfunction, immune dysregulation, and gut dysbiosis may play a role in this disease [7]. Skin barrier dysfunction is considered the first step in the development of AD [8,9]. Filaggrin (FLG) mutations lead to alterations in the differentiation and growth of a normal stratum corneum, increasing transepidermal water loss (TEWL) [10]. Moreover, skin barrier dysfunction increases allergic sensitization to antigens [11], an independent risk factor for developing food sensitization [12]. Skin barrier impairment, reflected in high TEWL and temperatures is also related to more severe disease [13].

Multiple diagnostic tools are used to evaluate severity in patients with AD, most commonly the Eczema Area and Severity Index (EASI) and SCORing Atopic Dermatitis (SCORAD) [14,15]. The EASI is a medical evaluation of extension and intensity, while the SCORAD also includes a patient assessment of itch and sleeplessness [1,14]; both these parameters have a subjective component that could lead to a high intra- and interobserver variability [16–18].

Available therapies for AD include topical corticosteroids (TCS), calcineurin inhibitors, phototherapy and systemic immunotherapies, such as cyclosporine [1]. Dupilumab, a fully human monoclonal antibody that binds specifically to the shared α chain subunit of the interleukin-4 and interleukin-13 receptors, is associated with clinical improvement in patients with AD, reducing EASI, SCORAD and Dermatology Life Quality Index (DLQI) with an acceptable safety profile [19]. Other biologics (dupilumab, tralokinumab) and JAK-inhibitors are being developed that are showing similar or even better score outcomes [20,21]. Nevertheless, there is scant information about how these treatments modify skin barrier function [7].

The difficulty in objectively assessing AD severity and the introduction of new biologicals in clinical practice highlight the need to find parameters to select the most appropriate treatment and for monitoring outcomes [22,23]. Skin barrier function parameters are easy to evaluate objectively [24,25] and may be suitable tools for assessing the efficacy of AD therapies.

Thus, the aim of this study is to evaluate the impact of dupilumab on skin barrier function and compare it with other treatments in patients with AD.

2. Materials and Methods

2.1. Study Design and Participants

We conducted a prospective observational study in participants recruited from September 2019 to May 2021, in the Department of Dermatology, Virgen de las Nieves University Hospital, Granada, Spain.

Eligible patients were adults diagnosed with AD by a dermatologist according to Hanifin and Rajka criteria [26,27], 18–65 years of age, with an eczematous lesion on the volar forearm, who were scheduled to start a new treatment with TCS, cyclosporine or dupilumab. Only patients with an eczematous lesion on the volar forearm were included because the measurements were always taken in this area to homogenize differences in skin barrier function that may exist in different body areas [25]. The exclusion criteria were: a clinical infection on the measured area; history of cancer; immunological disease or other inflammatory skin disease; incapacity to comply with the study protocol and no signed informed consent form.

Patients were assigned to treatment with TCS, cyclosporine or dupilumab following current clinical recommendations for treating AD [1]. Mild-to-moderate patients who had not received treatment for at least the previous six months were treated with mometasone cream once daily (TCS group). Moderate patients who had not responded to TCS and severe patients were assigned to the cyclosporine group and advised to avoid any topical treatment for one week prior to baseline evaluation. Cyclosporine was initiated at a high dose, 5 mg/kg/day, for a 3–6-week induction phase, followed by gradual tapering of the dose based on clinical response to a dose of 2–3 mg/kg/day in the maintenance phase. Moderate to severe patients who did not respond to cyclosporine or had some contraindication to receiving it were assigned to the dupilumab group. Dupilumab 300 mg was administered subcutaneously every other week following a loading dose of 600 mg. Immunosuppressive agents were discontinued at least 4 weeks prior to starting dupilumab in all patients. Patients receiving cyclosporine and dupilumab were allowed to use TCS twice a week if needed. Both patients starting cyclosporine and dupilumab should not use TCS or calcineurin inhibitors at least 24 h before baseline measurement. Patients in all groups were allowed to use emollients and moisturizers when needed but not 24 h before the baseline measure.

2.2. Outcomes and Measures

The main outcome measure to assess clinical improvement was EASI-50 (50% improvement in EASI) at treatment week 16 [28], and the primary outcome measure to assess skin barrier improvement was TEWL-50 (50% improvement in TEWL) at treatment week 16.

2.2.1. Clinical Assessment

AD severity was assessed by EASI, SCORAD, DLQI, the Investigator Global Assessment (IGA) scale and body surface area (BSA). EASI is calculated by independently assessing body surface involvement in four body regions (head and neck, upper extremities, trunk and lower extremities) and evaluating erythema, induration/papulation/edema, excoriations, and lichenification in each area [29]. SCORAD consists of the evaluation of disease extension, intensity (composed of six items: erythema, edema/papules, effect of scratching, oozing/crust formation, lichenification and dryness) and subjective symptoms (itch, sleeplessness) [15,27]. DLQI evaluates the impact of dermatological conditions and consists of a 10-item questionnaire addressing patient quality of life [30]. All clinical indexes were determined by a dermatologist at baseline and after the 16-week follow-up.

2.2.2. Skin Barrier Function Parameters

TEWL (in $g \cdot h^{-1} \cdot m^{-2}$), skin temperature (in °C), stratum corneum hydration (SCH) (in arbitrary units, AU), and pH were respectively measured using the Tewameter TM 300, Skin-Thermometer ST 500, Corneometer CM 825, and Skin-pH-Meter PH 905, connected to a Multi Probe Adapter (MPA, Courage + Khazaka electronic GmbH, Bilbao, Spain). All parameters were evaluated on an eczematous area on the volar forearm and on a non-involved area 5 cm from the affected area, and the average value from ten measurements in each location was used for analysis. Measurements were taken at baseline and at the 16-week follow-up after resting at least for 30 min in a room with controlled ambient air temperature and humidity, measured with the TFA Lab Thermometer IP65 LT-101, Wertheim, Germany (average air temperature 22 ± 1 °C; ambient air humidity of $45\% \pm 5\%$). No systemic or topical treatments were allowed in the six hours before the measurements were taken.

2.2.3. Other Variables

Age, sex, smoking/alcohol habits, marital status, education level, skin hydration habit, family history of AD, age of disease onset, signs of atopy march, and body mass index (BMI) were recorded by means of a clinical interview and physical examination. Regarding skin hydration habit patients were asked about the frequency they used emollients per week and were classified into those applying them ≤ 4 or >4 times per week.

2.3. Statistical Analysis

Descriptive statistics were used to present the sample characteristics. Continuous data were expressed as the mean (standard deviation) and qualitative data as relative (absolute) frequency. The Shapiro–Wilk test was used to determine the normality of data distribution and Levene's test to check the homogeneity of variance. One-way analysis of variance (ANOVA) was used to compare quantitative variables between different treatments. The Student's *t*-test for paired samples was used to compare differences in parameters before and after using the hand hygiene product. A multivariable logistic regression model was constructed to evaluate variables associated with TEWL-50 and EASI-50. Epidemiological and statistical criteria were used to model variable selection. The effect of each exploratory variable on the model and its significance were studied. If the variable improved the model fit and adequacy (based on the likelihood ratio criteria and the significance of the parameter), it was kept; otherwise, the variable was excluded. The model was checked for pairwise interaction between covariates. Potential confounding covariates were studied using a change of significance in the model's parameters or a change of 30% of its value.

Statistical significance was defined as a two-tailed $p < 0.05$. SPSS version 24.0 (SPSS Inc, Chicago, IL, USA) was used for statistical analyses.

2.4. Ethics

This study was approved by the ethics committee of Hospital Universitario Virgen de las Nieves (HC01/0442-N-20, Impact of topical, systemic or physical treatment on skin homeostasis in patients with skin diseases). The nature of the study was explained to all participants, who agreed to participate by giving their verbal and written consent. All measurements were non-invasive, and the confidentiality of participants' data was strictly preserved.

3. Results

3.1. Baseline Demographic and Clinical Characteristics

Sixty-two individuals were assessed for eligibility, and 46 patients with AD were finally included, 10 of whom were treated with TCS, 14 with cyclosporine and 22 with dupilumab. Demographic and clinical characteristics are shown in Table 1. Patients receiving TCS were younger than those treated with dupilumab and cyclosporine (19.90 vs. 28.95 vs. 35.64 years, $p = 0.042$). Patients in the TCS group were more frequently single than those in the dupilumab and cyclosporine groups (100% vs. 81.8% vs. 63.3%, $p = 0.036$). Dupilumab patients had a longer disease duration than those receiving cyclosporine or TCS (20.67 vs. 8.38 vs. 12.20, $p = 0.004$). After follow-up, 8.7% (4/46) of the patients did not complete the study (1 TCS, 2 cyclosporine, 1 dupilumab), Figure S1.

Table 1. Sociodemographic and clinical characteristics at baseline.

	Topical Corticosteroids (n = 10)	Cyclosporin (n = 14)	Dupilumab (n = 22)	p *
Age (years)	19.90 (3.76)	35.64 (22.42)	28.95 (10.83)	0.042 *
Female sex, % (n)	80% (8)	71.4% (10)	68.2% (15)	0.789
Marital status				
- Single	100% (10)	64.3% (9)	81.8% (18)	
- Married	0	35.7% (5)	4.5% (1)	0.036 *
- Divorced	0	0	13.6% (3)	
Mandatory educational level (yes)	80% (8)	71.4% (10)	88.9% (16)	0.457
Smoking habit (yes)	30% (3)	28.57% (4)	9.1% (2)	0.202
Alcohol consumption (yes)	20% (2)	42.86% (6)	27.3% (6)	0.350
BMI (kg/m^2)	20.55 (0.97)	22.66 (1.71)	22.02 (3.78)	0.566
Frecuecy of emollient use (times per week)	5.12 (1.72)	5.25 (3.50)	6.33 (1.19)	0.378
Skin hydration > 4 times/week (yes)	70% (7)	92.9% (13)	72.72% (16)	0.253
AD family history (yes)	40% (4)	71.43% (10)	68.18% (15)	0.111
Signs of atopic march (yes)	60% (6)	64.29% (9)	68.18% (15)	0.835
Disease evolution (years)	12.20 (7.68)	8.38 (7.45)	20.67 (12.39)	0.004 *
EASI	11.72 (3.91)	15.04 (4.90)	24.60 (5.36)	<0.001 *
SCORAD	37.28 (12.11)	47.41 (9.43)	57.30 (13.83)	<0.001 *
DLQI	12.20 (6.20)	9.29 (4.05)	17.59 (7.28)	0.001
BSA	14.23 (5.31)	22.63 (7.96)	39.54 (18.47)	<0.001
IGA	2.5 (0.53)	3.21 (0.58)	3.73 (0.46)	<0.001

Data are expressed as mean (standard deviation) or relative (absolute) frequency. AD, atopic dermatitis; BMI, body mass index; BSA, body surface area; DLQI, Dermatology Life Quality Index; EASI, Eczema Area Severity Index; IGA, Investigator Global Assessment scale; SCORAD, SCORing Atopic Dermatitis. * p-value after using one-way independent ANOVA to compare continuous variables or Chi-square or Fisher test, as appropriate, to compare qualitative variables.

3.2. Clinical Improvement

At baseline, patients who received dupilumab had more severe disease than those who received cyclosporine and TCS, reflected in higher EASI (24.60 vs. 15.04 vs. 11.72, $p < 0.001$), SCORAD (57.30 vs. 47.41 vs. 37.28, $p < 0.001$), DLQI (17.59 vs. 9.29 vs. 12.20,

$p < 0.001$), BSA (39.54 vs. 22.63 vs. 14.23, $p < 0.001$) and IGA scores (3.73 vs. 3.21 vs. 2.5, $p < 0.001$), Table 1.

The proportion of patients who achieved EASI-50 at week 16 was significantly higher among patients receiving dupilumab than those receiving cyclosporine or topical corticosteroids (81.8% vs. 28.6% vs. 40%, $p = 0.004$), Figure 1. After conducting a multivariate logistic regression model adjusted by age, age of disease onset, sex, smoking habit and skin hydration habit, treatment with dupilumab emerged as an independent factor for achieving EASI-50 (OR = 10.67, $p = 0.026$).

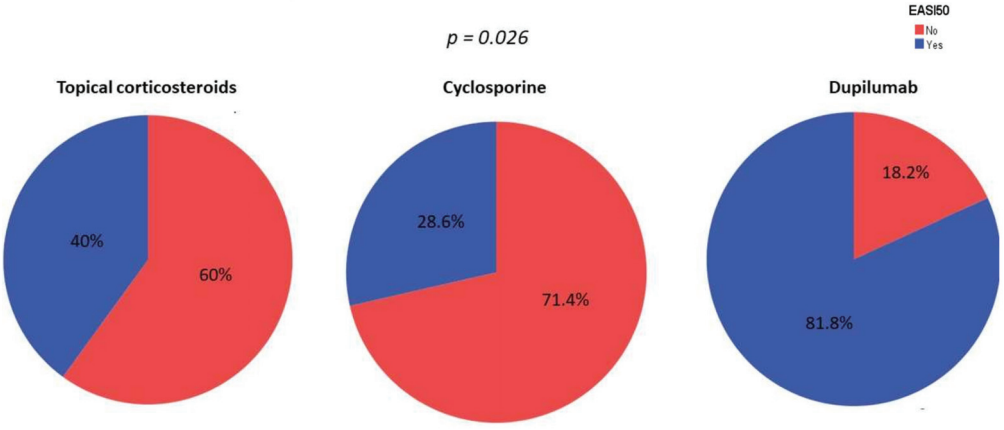

Figure 1. Percentage of patients achieving EASI-50 by treatment. *p*-value after conducting a multivariate logistic regression model adjusted by age, age of disease onset, sex, smoking habit and skin hydration habit.

Patients receiving dupilumab also showed greater improvement in SCORAD (−29.26 vs. −15.69 vs. −12.79, $p < 0.001$), DLQI (−12.52 vs. −2.56 vs. −3.75, $p < 0.001$), BSA and IGA scores than those treated with cyclosporine and TCS (Figure 2, Table S1).

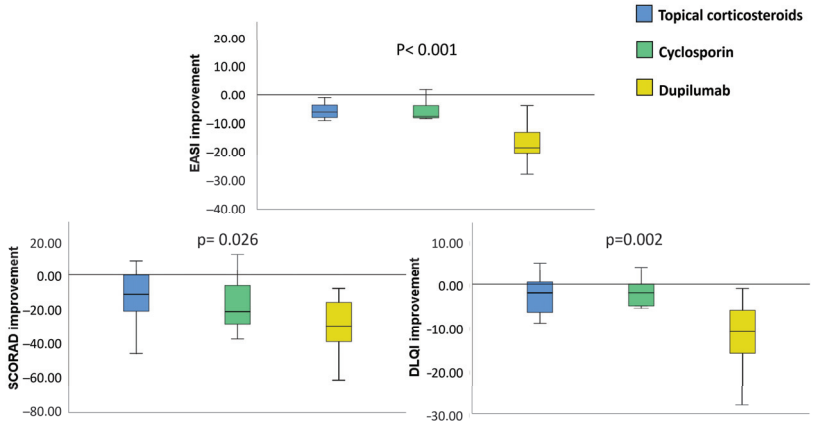

Figure 2. Differences in the improvement in clinical scores by treatment. DLQI, Dermatology Life Quality Index; EASI, Eczema Area Severity Index; SCORAD, SCORing Atopic Dermatitis. *p*-value after using one-way independent ANOVA test to compare changes in scores between different treatments (topical corticosteroids, cyclosporine and dupilumab).

3.3. Skin Barrier Function

Skin barrier function parameters did not differ at baseline between groups, see Table S2. In eczematous lesions, TEWL decreased in patients receiving dupilumab (31.02 vs. 12.10 $g \cdot h^{-1} \cdot m^{-2}$, $p < 0.001$) and TCS (25.30 vs. 14.88 $g \cdot h^{-1} \cdot m^{-2}$, $p = 0.047$) but did not change with cyclosporine. Temperature only decreased in the dupilumab group (32.53 vs. 31.64 °C, $p = 0.009$). SCH increased in patients treated with dupilumab (19.93 vs. 37.73 AU, $p < 0.001$) and TCS (18.24 vs. 40.79 AU, $p = 0.010$), but did not change with cyclosporine. pH did not change in any group. In non-lesioned skin, TEWL (11.87 vs. 8.25 $g \cdot h^{-1} \cdot m^{-2}$, $p = 0.006$) and SCH (32.68 vs. 41.68 AU, $p < 0.001$) only improved in patients receiving dupilumab. Temperature, SCH and pH did not change in non-lesioned skin after receiving any treatment, see Table S2.

The proportion of patients who achieved TEWL-50 at week 16 was greater for patients receiving dupilumab than for those receiving cyclosporine or topical corticosteroids (50% vs. 14.3% vs. 30%, $p = 0.101$). Furthermore, after a multivariate logistic regression model adjusted for age, age of disease onset, sex, smoking habit and skin hydration habit, dupilumab treatment with dupilumab emerged as an independent factor for achieving TEWL-50 ($p = 0.004$), see Figure 3.

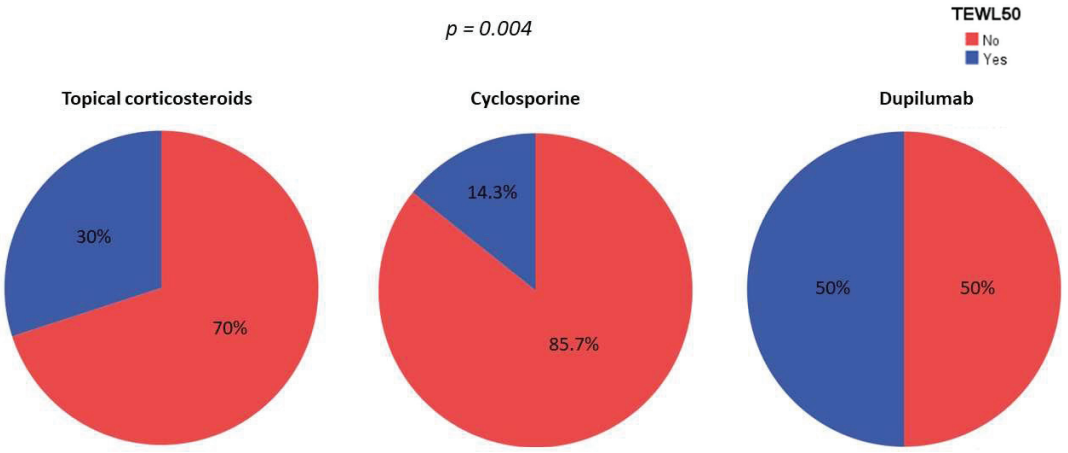

Figure 3. Percentage of patients reaching TEWL-50 by treatment. *p*-value after conducting a multivariable logistic regression model adjusted by age, age of disease onset, sex, smoking habit and skin hydration habit.

Differences between treatments in other skin barrier function changes were also found (Figure 4). The temperature decrease on eczematous lesions was greater after receiving dupilumab than after cyclosporine and TCS (−0.82 vs. +0.49 vs. +0.46 °C, $p = 0.013$). SCH improvement in eczematous lesions was greater in patients treated with dupilumab and TCS than in those receiving cyclosporine, while SCH increases in non-lesioned skin were higher in patients receiving dupilumab compared to TCS and cyclosporine (+11.41 vs. +0.05 vs. +6.46, $p = 0.033$). No differences in pH changes were found between treatments.

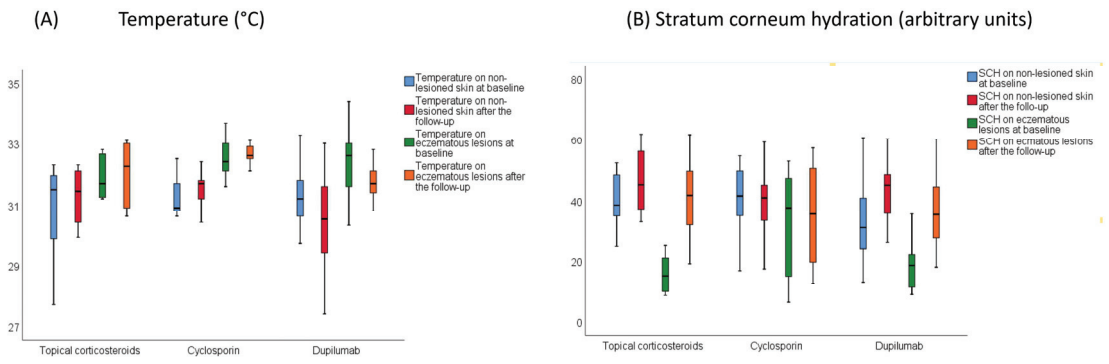

Figure 4. Changes in skin barrier function parameters after receiving each treatment. (**A**) Temperature (**B**) Stratum corneum hydration.

4. Discussion

This study shows that dupilumab is more effective than TCS and cyclosporine in improving both clinical scores, demonstrated by a higher proportion of patients achieving EASI-50, and epidermal barrier function, demonstrated by a higher proportion of participants achieving TEWL-50, after 16 weeks of treatment.

The increased effectiveness of dupilumab in clinical practice was reflected in a higher number of patients who achieved EASI-50 and a higher reduction in SCORAD and DLQI. Clinical improvement with dupilumab was similar to that observed in previous studies in real-life settings (achieving EASI-50 in around 80% at week 12–16) [31–33] and higher than in clinical trials [34]. As in previous studies, we found that dupilumab improves patient quality of life [35]. Although several studies reflect the great impact of dupilumab in improving EASI, SCORAD and DLQI scores, few reports have compared its effectiveness with other systemic therapies [36]. A direct comparison between dupilumab and cyclosporine was needed, as the lack of data meant that it could not be confirmed whether these patients were really improved by dupilumab or if they could also have improved with other treatments. A network meta-analysis of randomized clinical trials showed that cyclosporine and dupilumab were similarly effective and better than placebo in improving EASI [37]. Nevertheless, another meta-analysis found that only baricitinib and dupilumab were more effective after 16 weeks of treatment than placebo while cyclosporine was not [36]. Moreover, a recent indirect comparison observed that dupilumab was more effective than cyclosporine, as manifested by a higher percentage of patients reaching EASI-50 at week 16 [38]. Our study shows that dupilumab is more effective in clinical practice, even when compared to TCS and cyclosporine.

Regarding epidermal barrier function, patients with AD are known to have higher TEWL and lower SCH in both lesioned and non-lesioned skin due to skin barrier impairment than healthy individuals [39,40]. The alkalinization of the pH in AD could also increase skin barrier dysfunction [41]. Moreover, higher temperatures could be a sign of the inflammatory changes involved in this disease [42]. Skin barrier dysfunction in AD patients has been related to proinflammatory cytokine production, mainly IL-4 and IL-13, that inhibits the expression of barrier-related molecules such as filaggrin, involucrin and loricrin, that damage the skin barrier. TCS has been reported to improve clinical inflammatory features and reduce TEWL by decreasing IL-13 production and upregulating filaggrin and loricrin expression [43]. Skin barrier improvement with dupilumab may, then, be explained by the inhibition of IL-4/IL-13 signaling, reducing markers of type 2 inflammation and reversing AD-associated epidermal abnormalities [44]. Improvements in epidermal remodeling and inflammation after dupilumab treatment have also been observed on dynamic optical coherence tomography [45].

Some other studies have also evaluated the impact of dupilumab on skin barrier function [46–50]. They showed that dupilumab reduced TEWL on non-involved and involved skin [46–50]. The impact of dupilumab in SCH differs between studies. Cristaudo et al. found reported a decreasing trend in SCH values in 30 patients with AD after 8 weeks of treatment [46] while Furuhashi et al. found that SCH did not change after 24 weeks of dupilumab treatment in seven patients [47]. We also observed that dupilumab decreases TEWL and increases SCH, reflecting skin barrier recovery. The lack of differences in SCH in other studies may be explained by the shorter follow-up [46] or the limited number of participants [47]. Moreover, we also found that dupilumab decreases temperature, which might be reflecting a reduction in the inflammatory load, while we did not find changes in pH.

Furthermore, this is the first time to our knowledge that the impact of dupilumab on skin barrier function has been compared to other therapies, including cyclosporine and TCS. TCS also increased TEWL and decreased SCH but only in eczematous lesions. This may be because the skin compartment generates a major component of dysregulated systemic cytokines [51]. The lack of changes in skin barrier function after cyclosporine may be because this therapy does not act on the etiopathogenic axis of this disease. We also found that dupilumab is an independent factor for achieving TEWL-50. It has previously been suggested that TEWL could be a predictor for developing AD [9] or even a marker of disease severity [13] and that the decrease in TEWL after dupilumab treatment occurs mainly during the first two weeks [47], indicating that TEWL may likely be an early clinical response marker. Given the rapidly increasing number of drugs available for AD, it is important to identify markers that give an early indication of a lack of response in order to change treatment, reduce the burden of AD on patient quality of life and save healthcare costs.

Our research is limited by the small sample size and concerns surrounding data collection: in Spain, it is compulsory to use cyclosporine before dupilumab treatment, possibly biasing the comparison between patients receiving dupilumab and cyclosporine. This bias would be toward the null hypothesis, as the real difference in skin barrier function between dupilumab and cyclosporine would be even greater than that reported in this study. The main limitation of our study that it is difficult to ensure the comparability between patients receiving each treatment. In that way, patients receiving TCS had a less severe disease that those receiving cyclosporine or dupilimab; even if the proportion of patients achieving EASI-50 in the cyclosporine group was smaller than that in the TCS group, it does not necessarily mean that TCS was better or stronger than cyclosporine. Further clinical trials should be conducted to assess the real difference in clinical and skin barrier function improvement between TCS, cyclosporin and dupilumab. Other skin barrier function parameters, such as lipid content or filaggrin, should be also measured.

5. Conclusions

This is the first study to evaluate the impact of dupilumab on skin barrier function and compare it to other treatments. This research could increase our understanding of the mechanism of action of dupilumab and could help clinicians select the appropriate patients to receive this treatment. Further clinical research should be conducted to determine whether patients who did not achieve TEWL-50 would subsequently fail on dupilumab and if TEWL could be considered a marker of therapeutic response in patients with AD.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/jcm11123341/s1. Figure S1: Participant flow chart. Table S1: Changes in clinical scores by groups. Table S2: Changes in barrier function parameters after each treatment and by groups.

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Informed Consent Statement: Informed consent was obtained from all subjects involved in the study.

Data Availability Statement: The data presented in this study are available from the corresponding author on request.

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References

1. Wollenberg, A.; Christen-Zach, S.; Taieb, A.; Paul, C.; Thyssen, J.P.; De Bruin-Weller, M.; Vestergaard, C.; Seneschal, J.; Werfel, T.; Cork, M.J.; et al. ETFAD/EADV Eczema task force 2020 position paper on diagnosis and treatment of atopic dermatitis in adults and children. *J. Eur. Acad. Dermatol. Venereol.* **2020**, *34*, 2717–2744. [CrossRef]
2. Stander, S. Atopic Dermatitis. *N. Engl. J. Med.* **2021**, *384*, 1136–1143. [CrossRef] [PubMed]
3. Bylund, S.; Kobyletzki, L.B.; Svalstedt, M.; Svensson, A. Prevalence and Incidence of Atopic Dermatitis: A Systematic Review. *Acta Derm. Venereol.* **2020**, *100*, adv00160. [CrossRef] [PubMed]
4. Lloyd-Lavery, A.; Solman, L.; Grindlay, D.J.C.; Rogers, N.K.; Thomas, K.S.; Harman, K.E. What's new in atopic eczema? An analysis of systematic reviews published in 2016. Part 2: Epidemiology, aetiology and risk factors. *Clin. Exp. Dermatol.* **2019**, *44*, 370–375. [CrossRef] [PubMed]
5. Esparza-Gordillo, J.; Dekio, I. *Atopic Dermatitis—Disease Etiology and Clinical Management [Internet]*; IntechOpen: London, UK, 2012; 416p, Available online: https://www.intechopen.com/books/941 (accessed on 7 June 2022). [CrossRef]
6. Ali, F.; Vyas, J.; Finlay, A.Y. Counting the Burden: Atopic Dermatitis and Health-related Quality of Life. *Acta Derm. Venereol.* **2020**, *100*, adv00161. [CrossRef]
7. Luger, T.; Amagai, M.; Dreno, B.; Dagnelie, M.A.; Liao, W.; Kabashima, K.; Schikowski, T.; Proksch, E.; Elias, P.M.; Simon, M.; et al. Atopic dermatitis: Role of the skin barrier, environment, microbiome, and therapeutic agents. *J. Dermatol. Sci.* **2021**, *102*, 142–157. [CrossRef]
8. Kim, J.; Kim, B.E.; Leung, D.Y.M. Pathophysiology of atopic dermatitis: Clinical implications. *Allergy Asthma Proc.* **2019**, *40*, 84–92. [CrossRef]
9. Kelleher, M.; Dunn-Galvin, A.; Hourihane, J.O.; Murray, D.; Campbell, L.E.; McLean, W.H.I.; Irvine, A.D. Skin barrier dysfunction measured by transepidermal water loss at 2 days and 2 months predates and predicts atopic dermatitis at 1 year. *J. Allergy Clin. Immunol.* **2015**, *135*, 930–935.e1. [CrossRef]
10. David Boothe, W.; Tarbox, J.A.; Tarbox, M.B. Atopic Dermatitis: Pathophysiology. *Adv. Exp. Med. Biol.* **2017**, *1027*, 21–37. [CrossRef]
11. Tsakok, T.; Woolf, R.; Smith, C.H.; Weidinger, S.; Flohr, C. Atopic dermatitis: The skin barrier and beyond. *Br. J. Dermatol.* **2019**, *180*, 464–474. [CrossRef]
12. Flohr, C.; Perkin, M.; Logan, K.; Marrs, T.; Radulovic, S.; Campbell, L.E.; MacCallum, S.F.; McLean, W.H.I.; Lack, G. Atopic dermatitis and disease severity are the main risk factors for food sensitization in exclusively breastfed infants. *J. Investig. Dermatol.* **2014**, *134*, 345–350. [CrossRef] [PubMed]
13. Montero-Vilchez, T.; Segura-Fernandez-Nogueras, M.V.; Perez-Rodriguez, I.; Soler-Gongora, M.; Martinez-Lopez, A.; Fernandez-Gonzalez, A.; Molina-Leyva, A.; Arias-Santiago, S. Skin Barrier Function in Psoriasis and Atopic Dermatitis: Transepidermal Water Loss and Temperature as Useful Tools to Assess Disease Severity. *J. Clin. Med.* **2021**, *10*, 359. [CrossRef] [PubMed]
14. Chopra, R.; Silverberg, J.I. Assessing the severity of atopic dermatitis in clinical trials and practice. *Clin. Dermatol.* **2018**, *36*, 606–615. [CrossRef] [PubMed]

15. Severity scoring of atopic dermatitis: The SCORAD index. Consensus Report of the European Task Force on Atopic Dermatitis. *Dermatology* **1993**, *186*, 23–31. [CrossRef] [PubMed]
16. Zhao, C.Y.; Wijayanti, A.; Doria, M.C.; Harris, A.G.; Jain, S.V.; Legaspi, K.N.; Dlova, N.C.; Law, M.G.; Murrell, D.F. The reliability and validity of outcome measures for atopic dermatitis in patients with pigmented skin: A grey area. *Int. J. Womens Dermatol.* **2015**, *1*, 150–154. [CrossRef]
17. Hon, K.L.; Kung, J.S.C.; Ng, W.G.; Tsang, K.Y.C.; Cheng, N.; Leung, T.F. Are skin equipment for assessing childhood eczema any good? *J. Dermatol. Treat.* **2021**, *32*, 45–48. [CrossRef]
18. Chopra, R.; Vakharia, P.P.; Sacotte, R.; Patel, N.; Immaneni, S.; White, T.; Kantor, R.; Hsu, D.Y.; Silverberg, J.I. Relationship between EASI and SCORAD severity assessments for atopic dermatitis. *J. Allergy Clin. Immunol.* **2017**, *140*, 1708–1710.e1. [CrossRef]
19. Halling, A.S.; Loft, N.; Silverberg, J.I.; Guttman-Yassky, E.; Thyssen, J.P. Real-world evidence of dupilumab efficacy and risk of adverse events: A systematic review and meta-analysis. *J. Am. Acad. Dermatol.* **2021**, *84*, 139–147. [CrossRef]
20. Pereyra-Rodriguez, J.J.; Alcantara-Luna, S.; Dominguez-Cruz, J.; Galan-Gutierrez, M.; Ruiz-Villaverde, R.; Vilar-Palomo, S.; Armario-Hita, J.C. Short-Term Effectiveness and Safety of Biologics and Small Molecule Drugs for Moderate to Severe Atopic Dermatitis: A Systematic Review and Network Meta-Analysis. *Life* **2021**, *11*, 927. [CrossRef]
21. Blauvelt, A.; Teixeira, H.D.; Simpson, E.L.; Costanzo, A.; De Bruin-Weller, M.; Barbarot, S.; Prajapati, V.H.; Lio, P.; Hu, X.; Wu, T.; et al. Efficacy and Safety of Upadacitinib vs. Dupilumab in Adults With Moderate-to-Severe Atopic Dermatitis: A Randomized Clinical Trial. *JAMA Dermatol.* **2021**, *157*, 1047–1055. [CrossRef]
22. Nakahara, T.; Izuhara, K.; Onozuka, D.; Nunomura, S.; Tamagawa-Mineoka, R.; Masuda, K.; Ichiyama, S.; Saeki, H.; Kabata, Y.; Abe, R.; et al. Exploration of biomarkers to predict clinical improvement of atopic dermatitis in patients treated with dupilumab: A study protocol. *Medicine* **2020**, *99*, e22043. [CrossRef] [PubMed]
23. Breiteneder, H.; Peng, Y.Q.; Agache, I.; Diamant, Z.; Eiwegger, T.; Fokkens, W.J.; Traidl-Hoffmann, C.; Nadeau, K.; O'Hehir, R.E.; O'Mahony, L.; et al. Biomarkers for diagnosis and prediction of therapy responses in allergic diseases and asthma. *Allergy* **2020**, *75*, 3039–3068. [CrossRef] [PubMed]
24. Alexander, H.; Brown, S.; Danby, S.; Flohr, C. Research Techniques Made Simple: Transepidermal Water Loss Measurement as a Research Tool. *J. Investig. Dermatol.* **2018**, *138*, 2295–2300.e1. [CrossRef] [PubMed]
25. Espinosa-Rueda, M.I.; Montero-Vilchez, T.; Martinez-Lopez, A.; Molina-Leyva, A.; Sierra-Sanchez, A.; Arias-Santiago, S.; Buendia-Eisman, A. Cutaneous homeostasis and epidermal barrier function in a young healthy Caucasian population. *Eur. J. Dermatol.* **2021**, *31*, 176–182. [CrossRef]
26. Rudzki, E.; Samochocki, Z.; Rebandel, P.; Saciuk, E.; Galecki, W.; Raczka, A.; Szmurlo, A. Frequency and significance of the major and minor features of Hanifin and Rajka among patients with atopic dermatitis. *Dermatology* **1994**, *189*, 41–46. [CrossRef]
27. Oranje, A.P.; Glazenburg, E.J.; Wolkerstorfer, A.; De Waard-van der Spek, F.B. Practical issues on interpretation of scoring atopic dermatitis: The SCORAD index, objective SCORAD and the three-item severity score. *Br. J. Dermatol.* **2007**, *157*, 645–648. [CrossRef]
28. Schmitt, J.; Spuls, P.I.; Thomas, K.S.; Simpson, E.; Furue, M.; Deckert, S.; Dohil, M.; Apfelbacher, C.; Singh, J.A.; Chalmers, J.; et al. The Harmonising Outcome Measures for Eczema (HOME) statement to assess clinical signs of atopic eczema in trials. *J. Allergy Clin. Immunol.* **2014**, *134*, 800–807. [CrossRef]
29. Hanifin, J.M.; Thurston, M.; Omoto, M.; Cherill, R.; Tofte, S.J.; Graeber, M. The eczema area and severity index (EASI): Assessment of reliability in atopic dermatitis. EASI Evaluator Group. *Exp. Dermatol.* **2001**, *10*, 11–18. [CrossRef]
30. Finlay, A.Y.; Khan, G.K. Dermatology Life Quality Index (DLQI)–a simple practical measure for routine clinical use. *Clin. Exp. Dermatol.* **1994**, *19*, 210–216. [CrossRef]
31. Li, Y.; Lu, J.; Chen, R.; Wang, Y.; Ding, Y.; Xu, S.; Zou, Y.; Yi, X.; Shi, Y. The efficacy and safety of dupilumab for the treatment of atopic dermatitis among Chinese patients in clinical practice: A single-center retrospective study. *Dermatol. Ther.* **2022**, *35*, e15385. [CrossRef]
32. Jahn, S.; Fohr, J.; Diamanti, E.; Herbst, M. Treatment of atopic dermatitis with dupilumab: A retrospective cohort analysis from dermatological practice. *Hautarzt* **2021**, *72*, 1071–1078. [CrossRef] [PubMed]
33. Lee, Y.; Kim, M.E.; Nahm, D.H. Real Clinical Practice Data of Monthly Dupilumab Therapy in Adult Patients With Moderate-to-Severe Atopic Dermatitis: Clinical Efficacy and Predictive Markers for a Favorable Clinical Response. *Allergy Asthma Immunol. Res.* **2021**, *13*, 733–745. [CrossRef] [PubMed]
34. Silverberg, J.I.; Thyssen, J.P.; Fahrbach, K.; Mickle, K.; Cappelleri, J.C.; Romero, W.; Cameron, M.C.; Myers, D.E.; Clibborn, C.; DiBonaventura, M. Comparative efficacy and safety of systemic therapies used in moderate-to-severe atopic dermatitis: A systematic literature review and network meta-analysis. *J. Eur. Acad. Dermatol. Venereol.* **2021**, *35*, 1797–1810. [CrossRef]
35. Nettis, E.; Ferrucci, S.M.; Ortoncelli, M.; Pellacani, G.; Foti, C.; Di Leo, E.; Patruno, C.; Rongioletti, F.; Argenziano, G.; Macchia, L.; et al. Use of Dupilumab for 543 Adult Patients with Moderate-To-Severe Atopic Dermatitis: A Multicenter, Retrospective Study. *J. Investig. Allergol. Clin. Immunol.* **2022**, *32*, 124–132. [CrossRef] [PubMed]
36. Siegels, D.; Heratizadeh, A.; Abraham, S.; Binnmyr, J.; Brockow, K.; Irvine, A.D.; Halken, S.; Mortz, C.G.; Flohr, C.; Schmid-Grendelmeier, P.; et al. Systemic treatments in the management of atopic dermatitis: A systematic review and meta-analysis. *Allergy* **2021**, *76*, 1053–1076. [CrossRef] [PubMed]

37. Drucker, A.M.; Ellis, A.G.; Bohdanowicz, M.; Mashayekhi, S.; Yiu, Z.Z.N.; Rochwerg, B.; Di Giorgio, S.; Arents, B.W.M.; Burton, T.; Spuls, P.I.; et al. Systemic Immunomodulatory Treatments for Patients With Atopic Dermatitis: A Systematic Review and Network Meta-analysis. *JAMA Dermatol.* **2020**, *156*, 659–667. [CrossRef] [PubMed]
38. Ariens, L.F.M.; Gadkari, A.; Van Os-Medendorp, H.; Ayyagari, R.; Terasawa, E.; Kuznik, A.; Chen, Z.; Bego-Le Bagousse, G.; Lu, Y.; Rizova, E.; et al. Dupilumab Versus Cyclosporine for the Treatment of Moderate-to-Severe Atopic Dermatitis in Adults: Indirect Comparison Using the Eczema Area and Severity Index. *Acta Derm. Venereol.* **2019**, *99*, 851–857. [CrossRef]
39. Montero-Vilchez, T.A.-O.; Cuenca-Barrales, C.A.-O.; Rodriguez-Pozo, J.A.; Diaz-Calvillo, P.; Tercedor-Sanchez, J.A.-O.X.; Martinez-Lopez, A.; Molina-Leyva, A.A.-O.; Arias-Santiago, S.A.-O. Epidermal Barrier Function and Skin Homeostasis in Atopic Dermatitis: The Impact of Age. *Life* **2022**, *12*, 132. [CrossRef]
40. Thyssen, J.P.; Kezic, S. Causes of epidermal filaggrin reduction and their role in the pathogenesis of atopic dermatitis. *J. Allergy Clin. Immunol.* **2014**, *134*, 792–799. [CrossRef]
41. Danby, S.G.; Cork, M.J. pH in Atopic Dermatitis. *Curr. Probl. Dermatol.* **2018**, *54*, 95–107. [CrossRef]
42. Brunner, P.M.; Guttman-Yassky, E.; Leung, D.Y. The immunology of atopic dermatitis and its reversibility with broad-spectrum and targeted therapies. *J. Allergy Clin. Immunol.* **2017**, *139*, S65–S76. [CrossRef] [PubMed]
43. Guttman-Yassky, E.; Ungar, B.; Malik, K.; Dickstein, D.; Suprun, M.; Estrada, Y.D.; Xu, H.; Peng, X.; Oliva, M.; Todd, D.; et al. Molecular signatures order the potency of topically applied anti-inflammatory drugs in patients with atopic dermatitis. *J. Allergy Clin. Immunol.* **2017**, *140*, 1032–1042.e13. [CrossRef]
44. Guttman-Yassky, E.; Bissonnette, R.; Ungar, B.; Suarez-Farinas, M.; Ardeleanu, M.; Esaki, H.; Suprun, M.; Estrada, Y.; Xu, H.; Peng, X.; et al. Dupilumab progressively improves systemic and cutaneous abnormalities in patients with atopic dermatitis. *J Allergy Clin. Immunol.* **2019**, *143*, 155–172. [CrossRef] [PubMed]
45. Manfredini, M.; Liberati, S.; Ciardo, S.; Bonzano, L.; Guanti, M.; Chester, J.; Kaleci, S.; Pellacani, G. Microscopic and functional changes observed with dynamic optical coherence tomography for severe refractory atopic dermatitis treated with dupilumab. *Skin Res. Technol.* **2020**, *26*, 779–787. [CrossRef] [PubMed]
46. Cristaudo, A.; Pigliacelli, F.; Sperati, F.; Orsini, D.; Cameli, N.; Morrone, A.; Mariano, M. Instrumental evaluation of skin barrier function and clinical outcomes during dupilumab treatment for atopic dermatitis: An observational study. *Skin Res. Technol.* **2021**, *27*, 810–813. [CrossRef]
47. Furuhashi, T.; Oda, T.; Torii, K.; Nishida, E.; Morita, A. Dupilumab probably reduces transepidermal water loss but does not increase stratum corneum hydration in atopic dermatitis. *J. Dermatol.* **2021**, *48*, e74–e75. [CrossRef]
48. Berdyshev, E.; Goleva, E.; Bissonnette, R.; Bronova, I.; Bronoff, A.S.; Richers, B.; Garcia, S.; Gama, M.R.; Taylor, P.; Bologna, G.; et al. Dupilumab treatment significantly improves skin barrier function in adult and adolescent patients with moderate to severe atopic dermatitis. *J. Allergy Clin. Immunol.* **2022**, *149*, AB10. [CrossRef]
49. Ferrucci, S.; Romagnuolo, M.; Maronese, C.A.; Germiniasi, F.; Tavecchio, S.; Angileri, L.; Casazza, G.; Marzano, A.V.; Genovese, G. Skin barrier status during dupilumab treatment in patients with severe atopic dermatitis. *Ther. Adv. Chronic Dis.* **2021**, *12*, 20406223211058332. [CrossRef]
50. Lee, S.J.; Kim, S.E.; Shin, K.O.; Park, K.; Lee, S.E. Dupilumab Therapy Improves Stratum Corneum Hydration and Skin Dysbiosis in Patients With Atopic Dermatitis. *Allergy Asthma Immunol. Res.* **2021**, *13*, 762–775. [CrossRef]
51. McAleer, M.A.; Jakasa, I.; Stefanovic, N.; McLean, W.H.I.; Kezic, S.; Irvine, A.D. Topical corticosteroids normalize both skin and systemic inflammatory markers in infant atopic dermatitis. *Br. J. Dermatol.* **2021**, *185*, 153–163. [CrossRef]

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Review

Use of Dexpanthenol for Atopic Dermatitis—Benefits and Recommendations Based on Current Evidence

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Abstract: Background: Atopic dermatitis (AD) is an inflammatory skin disease of multiple phenotypes and endotypes, and is highly prevalent in children. Many people of all ages, including active adolescents, pregnant women, and the elderly, suffer from AD, experiencing chronicity, flares, and unexpected relapse. Dexpanthenol has multiple pharmacological effects and has been employed to treat various skin disorders such as AD. We aimed to summarize the up-to-date evidence relating to dexpanthenol and to provide a consensus on how to use dexpanthenol effectively for the treatment of AD. Methods: The evidence to date on the application and efficacy of dexpanthenol in AD was reviewed. The literature search focused on dexpanthenol use and the improvement of skin barrier function, the prevention of acute flares, and its topical corticosteroid (TCS) sparing effects. Evidence and recommendations for special groups such as pregnant women, and the effects of dexpanthenol and emollient plus in maintenance therapy, were also summarized. Results: Dexpanthenol is effective and well-tolerated for the treatment of AD. Dexpanthenol improves skin barrier function, reduces acute and frequent flares, has a significant TCS sparing effect, and enhances wound healing for skin lesions. Conclusion: This review article provides helpful advice for clinicians and patients on the proper maintenance treatment of AD. Dexpanthenol, as an active ingredient in ointments or emollients, is suitable for the treatment and maintenance of AD. This paper will guide dermatologists and clinicians to consider dexpanthenol as a treatment option for mild to moderate AD.

Keywords: atopic dermatitis; dexpanthenol; topical corticosteroid; emollient

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1. Introduction

Atopic dermatitis (AD) is a chronic inflammatory skin disorder characterized by pruritus and recurrent inflammation [1]. Patients with AD commonly suffer from many symptoms and signs, including pruritis, pain, erythema, excoriation, and sleep disturbances [2–4]. Difficulties during outdoor activities and periorbital hyperpigmentation are common complaints of active adolescents and women, respectively [5–8]. The treatment of AD might be particularly challenging for certain patients, including those with severe AD [9–13], frequent relapses, extensive area of involvement [14,15], and steroid-phobia, those without experience of full recovery, thereby making rapport difficult [16,17], and the elderly with weak, thin skin.

Most guidelines recommend the use of emollients in conjunction with topical corticosteroids (TCS) for the initial treatment of this intractable disease. Moisturizer is used synonymously with emollient and refers to as a product that moisturizes and smooths the skin, whereas humectant increases or maintains hydration of the skin [18]. Dexpanthenol is a stable alcohol analog of pantothenic acid (vitamin B5) with moisturizing and wound

healing efficacy. Increasing evidence has revealed that topical dexpanthenol can be employed as an effective and well-tolerated agent for AD flare and maintenance [19]. Based on the published evidence and Korean dermatologists' expert consensus, we aimed to review the experiences, and recommendations on the management of AD, especially in terms of dexpanthenol use.

2. Overview of AD and Treatment Options

2.1. Prevalence of AD

AD is highly prevalent in children. It affects 15–20% of children (ISAAC study) and up to 3% of adults worldwide [20]. Regions with a high prevalence of AD are characterized by urbanization and industrialization [20–23]. A recent study has reported that the prevalence of AD in infants aged 1–12 months was 30.48% in China [24]. The prevalence rate based on the Korea National Health and Nutrition Examination Survey (2010–2012) was highest at 3.5% for men and 4.3% for women aged 19–29 years and declined sharply in people aged 30 and above [25]. In Japan, the prevalence of childhood AD was 12–13% in the mainland [22]. As AD has an overwhelmingly high prevalence in children globally, it is necessary to provide effective, safe, and well-tolerated agents that are convenient for daily use.

2.2. Pathophysiology of AD

The development of AD is a multifactorial process involving immunologic defects, dysfunctional skin barrier, genetic variations, and environmental factors [2,26,27]. A biphasic inflammation pattern is frequently observed in the disease course of AD. Acute flares are triggered by a Th2-biased immune response, while Th1/Th22 deviation is predominantly responsible for chronic lesions [28]. Stratum corneum (SC), supported by a lamellar-structured extracellular lipid matrix consisting of ceramides, cholesterol, and free fatty acids, plays an indispensable role in preventing transcutaneous water loss and bacterial invasion. Defective skin barrier function, leading to increased transepidermal water loss (TEWL) and decreased SC hydration (dry skin), is a characteristic feature of AD. An impaired skin barrier plays a significant role in various skin conditions, such as dry skin (as a condition itself), sensitive skin, seborrheic dermatitis, contact dermatitis, or AD [29,30].

2.3. Burdens of AD

The burden of AD arises from not only the symptoms, but also the chronic course of the disease. AD can involve physical, social, and mental impacts, ultimately worsening a patient's quality of life. Many patients with AD suffer from itching and pain, leading to significant sleep disturbance, anxiety, and depression [31]. One of the most challenging parts of AD treatment is that it is hard to prevent flares completely and overcome the disease entirely. Recent evidence indicates that AD is not only a dermatological disease but also an inflammatory disease that extends beyond the skin. Patients with AD have a greater risk of cardiovascular disorders than healthy controls, including stroke, myocardial infarction, angina, and peripheral vascular disease [32].

Furthermore, out-of-pocket health care expenses associated with AD are a significant burden on patients with AD [33]. The cost of sophisticated emollient therapies often makes patients hesitant to use the recommended amount of 250 g/week for adults [34].

2.4. Treatment Options

Based on recent guidelines and consensus, topical emollient/moisturizer, TCS, topical calcineurin inhibitors (TCI), topical phosphodiesterase 4 inhibitors, oral immunosuppressants, and biologics are current effective treatment options for AD [34–38]. TCS has long been the first criterion of choice for AD therapeutics, generally in combination with topical moisturizers. Despite advances in the development of systemic drugs such as dupilumab, topical therapies continue to be essential for skin barrier dysfunction and for the delivery of anti-inflammatory therapeutics [39].

The daily use of topical moisturizers may help manage AD or can decrease the frequency of flare recurrence. Topical moisturizers might have a role as skincare products during post-inflammatory maintenance stages due to their established skin hydration, skin barrier restoration potential, and wound healing effects [19,40,41]. Guidelines in Asia, the USA, and Europe recommend the daily application of moisturizers as first-line therapy [35,36,39]. Moisturizers should be selected depending on skin type, degree of dryness, and the humidity of the climate [42].

For mild-to-moderate AD, moisturizers, TCS, and antihistamines are generally recommended. For severe AD, more potent TCS, TCIs, systemic immunosuppressants, biologics, and phototherapy are considered. The treatment goal is to achieve absent or mild symptoms without medication, and to reduce or eliminate discomfort in performing daily activities, and slight symptoms can be controlled by moisturizers [43]. In any case, moisturizers and patient education are necessary for the management of AD.

2.5. TCS and TCI Treatment for AD

TCS is recognized as a mainstay for AD treatment for mild-to-severe symptoms. TCS produces anti-inflammatory, antipruritic, and vasoconstriction effects via interaction with steroid receptors. The inflammatory cascades during the flares of AD symptoms are suppressed, along with the inhibition of the release of inflammatory mediators. Recent guidelines on AD management from the American Academy of Dermatology, the Joint Task Force, European Task Force on Atopic Dermatitis (ETFAD), and Asia, including Korea, Japan, and China, have recognized TCS and emollients as initial therapy options for targeting inflammation [23,36,39,43–45]. For preventive purposes, intermittent proactive application with TCS could help treat frequent flares [39]. However, topical steroids sometimes cause side effects; e.g., a human and murine study has revealed that the short-term (three days) use of TCS (clobetasol 0.05%) could disrupt the epidermal barrier by inhibiting the epidermal synthesis of fatty acids and impairing SC cohesion and integrity, delaying the recovery of the epidermal barrier [46].

TCIs, for example, tacrolimus ointment and pimecrolimus cream, are non-steroid anti-inflammatory drugs. TCIs have been proven effective in short-term, long-term, and proactive treatment of AD. In contrast to TCS, TCIs are not associated with skin atrophy, glaucoma, or cataract, which favors their use in delicate and sensitive areas and for long-term management. TCIs are well-tolerated, but some patients experience a burning sensation and transient worsening of skin conditions, particularly during acute flares [39].

3. How Dexpanthenol Can Help in AD

3.1. Property and Mechanism of Action of Dexpanthenol

Dexpanthenol is the dextrorotatory isomer of panthenol, and only the dextro-form is biologically active. Panthenol (provitamin B5) and pantothenic acid (vitamin B5) have a similar structure, and the oxidation of panthenol produces pantothenic acid. All animals need pantothenic acid to synthesize coenzyme A (CoA), which plays a crucial role in the oxidation and synthesis of fatty acids [40]. Dexpanthenol is an odorless, transparent, colorless, and highly viscous liquid at room temperature. It is freely soluble in water and alcohol. Its physical properties make it easy to formulate pharmaceutical dosage forms, such as ointments, gels, creams, and hydrogels. Dexpanthenol, pantothenic acid, and their derivatives, are regarded as safe by Cosmetic Ingredient Review [47], and dexpanthenol has been approved by the Food and Drug Administration and the European Commission on Cosmetics.

Since dexpanthenol is highly hygroscopic, it can penetrate easily into the skin and serve as a moisturizer or humectant to maintain the normal skin barrier properties, smoothness and skin elasticity [19,40]. Several in vivo and in vitro studies have shown that dexpanthenol promotes fibroblast proliferation, accelerates re-epithelization, moisturizes the skin, restores the skin barrier, and heals wounds [40,48,49]. In animal studies, dexpanthenol demonstrated cell proliferation and epithelium protection [50–52]. Owing to these effects,

the combination of dexpanthenol with nasal decongestants could relieve symptoms in patients with acute rhinitis [53]. Dexpanthenol protected against lipopolysaccharide-induced acute lung injury in mice [54].

3.2. Effect of Dexpanthenol on Skin Barrier Function

The skin barrier serves as frontline protection, so its intact function and restoration are implicated in various skin conditions including dry skin, sensitive skin, seborrheic dermatitis, AD, and contact dermatitis. Preventive skin hygiene, such as stabilizing skin barrier function with topical treatment, is critical in the care of patients with AD [55]. In most guidelines and consensus, a daily, frequent, regular application of moisturizers, which can help to enhance the skin barrier function, is recommended or required [23,36,39,43,56,57]. Asian countries also consider moisturizers as an important skincare method to provide better skin barrier function [23,36,43,57,58]. Emollients may be composed of humectants for promoting SC hydration and occlusives for reducing moisture evaporation. Although emollients are the basic therapy for skin barrier dysfunction, the direct sole use of emollients on inflamed skin areas is poorly tolerated, and treating the acute flare first is recommended [39].

Due to its extremely hygroscopic characteristics, dexpanthenol provides notable humectant effects. Topical dexpanthenol improves skin hydration and reduces transepidermal water loss (TEWL), thus maintaining the skin's smoothness and elasticity [40,41,59]. According to the evaluation of average moisture retention for 5 h, dexpanthenol mediates sustained tissue moisturizing effects [60].

Topical 2.5% dexpanthenol formulated in lipophilic vehicles was applied to the skin of 60 healthy volunteers in a double-blind, randomized controlled trial. Dexpanthenol application twice a day for seven days significantly improved SC hydration and reduced TEWL, compared with vehicle controls [41]. In another clinical study, the effect of dexpanthenol cream on skin barrier repair significantly increased after sodium lauryl sulfate (SLS)-induced irritation. After application of dexpanthenol cream for seven days, the skin barrier function was restored. Significant differences were observed between dexpanthenol use and placebo treatment [61]. In addition, SC hydration at dexpanthenol-treated sites remained steady following seven-day treatment with SLS [62]. The hydrating effect may be interrelated with its capacity to regenerate the epidermal barrier [63]. Repairing the skin barrier or preventing barrier dysfunction are essential strategies for reducing the risks for eczema [64].

3.3. Effect of Dexpanthenol on AD Flares

As first-line therapy for acute flares, emollients [65] and TCS [58] are recommended for treatment and remission of AD. For acutely inflamed flare lesions, the guidelines indicate that treatment with anti-inflammatory topicals, such as TCS or TCIs, is required first, rather than emollients alone [35,39]. For acute flares, especially oozing and erosive lesions, the 'wet-wrap' treatment has been recommended by the ETFAD [39]. For patients with moderate to severe AD, wet-wrap therapy containing emollients with or without TCS could be recommended to relieve pruritis, severity, and improve hydration during flares [35,66]. Wet-wrap therapy is highly effective and could improve tolerance, especially for patients with acute, oozing, and erosive lesions, and for children [44].

Emollients may cause irritation when directly used on inflamed skin. Acute flares should always be treated first with appropriate TCS, followed by emollients and emollients on the surrounding skin [39]. Two conditions are required to ensure the effectiveness of emollients—control of acute flares and proper formulation of the emollient [67]. The order of application of TCS and emollients did not result in significant variation of treatment outcomes. Therefore, emollients can be applied before or after TCS [68]. Application of topical agents is recommended within a few minutes following showering or bathing while a small amount of moisture remains. The consensus is that the topical agent should be left for an appropriate period to allow complete absorption before applying another agent. The guidelines recommend the "soak and smear" technique when using topical agents

(emollients and/or TCS) to maximize the absorption of active ingredients, which penetrate the epidermal layer via the expanded pores resulting from bathing [29,35,69,70].

Applying emollients as freely and frequently as possible is recommended, preferably every 4 h or at least 3–4 times per day. The ETFAD recommendations indicate that a sufficient quantity of emollients, at least 30 g/day or 1 kg/month for an adult with AD, should be applied in a 'soak and smear' or 'soak and seal' technique [44].

3.4. TCS Sparing Effect of Dexpanthenol

Previous results have suggested that the effectiveness of dexpanthenol is comparable to that of TCS. The topical application of moisturizers in adequate amounts, irregularly or continuously, was proved effective in sparing the use of TCS as short- or long-term treatment, and in maintaining the remission obtained with corticosteroids [36,71–73]. In practical guidance from a national expert panel in Italy, the TCS sparing effect was confirmed when patients were administered moisturizers and emollients intermittently or continuously in appropriate amounts [71]. The supply of hydration was also proved by the sustained remission of atopic lesions obtained with TCS treatment. Optimal skin hydration could reduce skin inflammation and the frequency of flares [71]. Eventually, the amount of TCS was decreased following the topical application of moisturizers [36]. The application of both agents twice daily is the basic recommendation of most guidelines or consensus [36,39,42,69].

Regular daily use of topical emollients could reduce the amount of TCS for short- and long-term treatment in mild-to-moderate AD [56]. Since the topical application of 5% dexpanthenol showed comparable efficacy with low potency TCS, dexpanthenol could be a substitute for TCS [73]. According to the evidence of steroid-sparing, the expert panel recommended that when dexpanthenol is used on a daily basis, TCS can be used every other day, particularly for infants, children, or patients with potential TCS side effects or steroid-phobia.

For severe AD, TCS or TCI is required to achieve effective treatment. Proper guidance, persuasive education, and basic treatment such as the use of dexpanthenol would be required for patients who abuse TCS, including addiction to high potency TCS, or have steroid-phobia [74].

Overall, the panel recommended that patients with atopic dermatitis should use TCS and dexpanthenol alternatively, especially if the disease is mild to moderate.

3.5. Special Populations

Schmutz et al. reported non-inferiority in maintaining TEWL scores in acute radiation dermatitis (0.1% methylprednisolone cream vs. 0.5% dexpanthenol cream) [75]. As shown in full-thickness 3D skin models representing acute radiodermatitis and mucositis, skin impairment seven days after radiotherapy demonstrated a completely restored epidermal part after treatment with dexpanthenol-containing ointment or liquid [76].

Gestational AD is one of the most common skin diseases during pregnancy. According to Japanese guidelines, TCS is considered safe for both pregnancy and breastfeeding, since the absorption of TCS into the bloodstream is low [43]. In the Taiwanese consensus, the application of TCS during pregnancy was also disclosed as safe, except for fluticasone propionate due to its metabolic characterization [57]. Although normal use of lower potency TCS is regarded as safe, low birth weight might be related to long-term use of higher potency TCS at high doses ($\geq$300 g) [43]. Dexpanthenol has therapeutic effects on nipple trauma through epithelialization and granulation [77]. During the lactation period, TCS should be smeared after breastfeeding, followed by the cleaning of nipples before feeding [57]. The use of emollients and TCS with a moderate-to-low potency is the ideal treatment for this area [78]. Skin damage can be caused by ablative laser therapy, microneedling, or tattooing. To facilitate wound healing, dexpanthenol reduces inflammation, and promotes cell proliferation and epithelial remodeling [49]. Clinically, dexpanthenol demonstrated superior re-epithelialization rates compared to standard treatments such as petroleum

jelly. Therefore, topical dexpanthenol is recommended as an effective treatment option for superficial skin damage in the early stages [49]. Among patients who received laser corneal surface ablation, 2% dexpanthenol resulted in significantly better vision and reduced residual cylinder after seven days, compared to artificial teardrops [79]. Topical application of 5% dexpanthenol to freshly tattooed skin restored the skin barrier, as demonstrated by TEWL [80]. Throat pain and tonsillar wound healing after tonsillectomy were significantly improved in dexpanthenol-treated patients, via its anti-inflammation, skin hydration, and mucosal protection properties [81]. Dexpanthenol could prevent the occurrence of postoperative sore throat [82], and promoted skin healing at the laser-irradiated site of photo-damaged skin [83]. Dexpanthenol produced significantly improved results and re-epithelialization earlier after laser therapy. Hence, dexpanthenol could be a promising alternative to routinely used treatments for wound healing.

Furthermore, cheilitis associated with isotretinoin treatment was markedly improved after topical application of 5% dexpanthenol cream [84]. The expert panel recommended dexpanthenol ointment as an effective and well-tolerated treatment for cheilitis, due to its hygroscopic activity and moisturizing formulation. Another study reported that 0.5% dexpanthenol cream in addition to TCS delayed the development of acute radiation dermatitis, in terms of clinical scores and TEWL values [75].

3.6. Effect of Dexpanthenol on AD Maintenance

AD depicts various phenotypes and endotypes depending on age, chronicity, atopic status, or ethnicity [28,85]. For long-term maintenance therapy to maintain adequate skin hydration and prevent flares, moisturizers such as dexpanthenol ointment should be continuously used, at least twice daily, after the induction of remission by TCS [39,58].

In the recent study involving infants and children with stabilized mild AD [86], a dexpanthenol medical device cream was applied 2–3 times daily in the stabilization phase, until severity was reduced. Then, a topical panthenol-containing cosmetic emollient was used twice daily during the maintenance phase. At the end of the three-month study, the proportions of patients without flares in the dexpanthenol and reference groups were 96% and 77%, respectively. In healthy subjects, the same dexpanthenol-containing emollient was effective in reducing TEWL and enhancing skin hydration. Moreover, Raman spectroscopy revealed that dexpanthenol-containing emollient was associated with sustained and deep skin moisturization and improved intercellular lipid lamellae organization [48,87]. Higher water distribution was observed by the relocation of the water molecules from more superficial to deeper layers of the SC, which resulted in deeper moisturization [87]. Dexpanthenol formulation effectively increased skin hydration and was well-tolerated in healthy infants between 3 and 25 months old without significant change in mean cutaneous tolerability scores [87].

3.7. Emollient Plus in Maintenance

Emollients are mostly recommended as the first-line therapy for AD, even when the disease is clear or almost clear, as well as during acute flares and remission [39,43,65,70,88]. Proksch et al. proposed that emollients should include a proper combination of humectants, physiological and non-physiological lipids, antipruritics, and multifunctional components such as dexpanthenol [89]. Selecting an appropriate emollient for patients with AD would improve acceptability and adherence for emollient treatment. A physician's recommendation is the primary consideration for patients when choosing an emollient; therefore, doctors should provide evidence-based information on these emollients [90].

Traditionally, emollients are generally considered to be topical formulations without active pharmaceutical ingredients. "Emollient plus" has been defined to include topical formulations with vehicle-type substances and additional active, non-medicated substances [39]. The active, non-medicated substances are active ingredients that do not qualify as topical drugs [66], which include saponins, flavonoids, vitamins such as riboflavin and niacin, and beneficial bacterial lysates. Some of these active ingredients were

found to improve skin protection, relieve pruritis, exert an anti-inflammatory reaction, exhibit antioxidant properties, and provide biologically essential lipids and antimicrobial activities [91–93]. Dexpanthenol is approved as a cosmetic ingredient with established safety, and it has evidence-based potent skin-hydration and wound-healing effects [19,40,41]. Products containing dexpanthenol as active ingredients can be considered "emollient plus".

Dexpanthenol accelerated the wound healing process (by a factor of 1.52 vs. the vehicle) and promoted fibroblast proliferation, in vivo and in vitro [40]. Another double-blind study monitored by histological examination revealed that dexpanthenol accelerated the wound-healing process [94]. In a randomized controlled trial, wound healing effects of water-filtered infrared-A (IRA) and/or dexpanthenol were examined in 12 healthy subjects using an acute wound model. Measured by laser scanning microscopy, the fastest SC formation was observed when water-filtered IRA irradiation was combined with dexpanthenol cream [95].

For healthy volunteers with dry skin, topical dexpanthenol-containing emollients (oil-in-water formulation) were topically applied like cosmetic products for daily care over four weeks [59]. The dexpanthenol formulation induced a significant increase in skin elasticity as measured by Cutometer® MPA580 (Courage & Khazaka, Cologne, Germany), skin hydration, TEWL, and SC lipid contents. Use of the dexpanthenol formulation once daily for over 28 days was well-tolerated in healthy adults.

Other formulations with dexpanthenol were tested in healthy adult women who underwent non-ablative laser resurfacing, laser depilation, or chemical peel [96]. The tested formulations maintained skin integrity, promoted recovery of damaged skin, and reduced erythema, and were associated with significantly decreased TEWL and dermal temperature. In the field of aesthetic dermatology, these dexpanthenol-containing formulations were well appreciated and would be an appropriate option for post-procedural care.

Compared with drug-free vehicles, panthenol-containing emollient plus can provide additional benefits such as accelerated wound healing, more prominent skin hydration, reduced skin redness from inflammation, and improvement to rough skin [62]. 5% dexpanthenol cream was superior to placebo in terms of SC hydration and protection against skin irritation in 23 healthy participants after exposure to SLS.

4. Conclusions

The treatment of AD requires long-term, risk-based stepwise management. Moisturizers are the first-line or basic therapy for AD treatment across various national guidelines and consensuses. Topical application of dexpanthenol significantly improved SC hydration and skin barrier function compared with the control. Appropriate use of dexpanthenol ointments during acute dermatitis flares is useful for minimizing epidermal disruption caused by TCS. The regular use of a dexpanthenol ointment subsequent to remission of AD flares has a steroid-sparing effect. The current evidence reveals that 5% dexpanthenol ointment has a good efficacy, safety, and tolerability profile, and is suitable for use during pregnancy and lactation.

Panthenols rapidly convert to pantothenic acid, resulting in very low toxicity. Allergic or irritant reactions to dexpanthenol have been reported, but overall it is generally well-tolerated [40,97]. As with all medications, proper use of dexpanthenol should be discussed with physicians.

Dexpanthenol-containing emollients, especially water-in-oil formulations, are considered "emollient plus" for AD treatment and provide improved skin hydration and wound healing effects compared with conventional emollients. Summary of the current evidence indicates that dexpanthenol might be a suitable ingredient for flare control and maintenance of AD. Physicians should consider prescribing an emollient plus over TCS if patients have steroid-phobia or show signs of TCS side effects. Because atopic dermatitis needs long-term management, dexpanthenol could be a promising ingredient for patients.

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References

1. Abramovits, W.; Hebert, A.A.; Boguniewicz, M.; Kempers, S.E.; Tschen, E.; Jarratt, M.T.; Lucky, A.W.; Cornelison, R.L.; Swinyer, L.J.; Jones, T.M. Patient-reported outcomes from a multicenter, randomized, vehicle-controlled clinical study of MAS063DP (Atopiclair) in the management of mild-to-moderate atopic dermatitis in adults. *J. Dermatol. Treat.* **2008**, *19*, 327–332. [CrossRef] [PubMed]
2. Yew, Y.W.; Thyssen, J.P.; Silverberg, J.I. A systematic review and meta-analysis of the regional and age-related differences in atopic dermatitis clinical characteristics. *J. Am. Acad. Dermatol.* **2019**, *80*, 390–401. [CrossRef] [PubMed]
3. Darsow, U.; Raap, U.; Stander, S. Atopic Dermatitis. In *Itch: Mechanisms and Treatment*; Carstens, E., Akiyama, T., Eds.; CRC Press: Boca Raton, FL, USA, 2014.
4. Langan, S.M.; Irvine, A.D.; Weidinger, S. Atopic dermatitis. *Lancet* **2020**, *396*, 345–360. [CrossRef]
5. Drucker, A.M.; Wang, A.R.; Li, W.Q.; Sevetson, E.; Block, J.K.; Qureshi, A.A. The Burden of Atopic Dermatitis: Summary of a Report for the National Eczema Association. *J. Investig. Dermatol.* **2017**, *137*, 26–30. [CrossRef]
6. Ben-Gashir, M.A.; Seed, P.T.; Hay, R.J. Quality of life and disease severity are correlated in children with atopic dermatitis. *Br. J. Dermatol.* **2004**, *150*, 284–290. [CrossRef]
7. Sarkar, R.; Ranjan, R.; Garg, S.; Garg, V.K.; Sonthalia, S.; Bansal, S. Periorbital Hyperpigmentation: A Comprehensive Review. *J. Clin. Aesthet. Dermatol.* **2016**, *9*, 49–55.
8. Poladian, K.; De Souza, B.; McMichael, A.J. Atopic dermatitis in adolescents with skin of color. *Cutis* **2019**, *104*, 164–168.
9. Pei, A.Y.; Chan, H.H.; Ho, K.M. The effectiveness of wet wrap dressings using 0.1% mometasone furoate and 0.005% fluticasone proprionate ointments in the treatment of moderate to severe atopic dermatitis in children. *Pediatr. Dermatol.* **2001**, *18*, 343–348. [CrossRef]
10. Wolkerstorfer, A.; Visser, R.L.; de Waard-van der Spek, F.B.; Mulder, P.G.; Oranje, A.P. Efficacy and safety of wet-wrap dressings in children with severe atopic dermatitis: Influence of corticosteroid dilution. *Br. J. Dermatol.* **2000**, *143*, 999–1004. [CrossRef] [PubMed]
11. Nicol, N.H.; Boguniewicz, M. Wet Wrap Therapy in Moderate to Severe Atopic Dermatitis. *Immunol. Allergy Clin. N. Am.* **2017**, *37*, 123–139. [CrossRef]
12. Boguniewicz, M.; Alexis, A.F.; Beck, L.A.; Block, J.; Eichenfield, L.F.; Fonacier, L.; Guttman-Yassky, E.; Paller, A.S.; Pariser, D.; Silverberg, J.I.; et al. Expert Perspectives on Management of Moderate-to-Severe Atopic Dermatitis: A Multidisciplinary Consensus Addressing Current and Emerging Therapies. *J. Allergy Clin. Immunol. Pract.* **2017**, *5*, 1519–1531. [CrossRef] [PubMed]
13. Yum, H.Y.; Kim, H.H.; Kim, H.J.; Kim, W.K.; Lee, S.Y.; Li, K.; Lee, D.H.; The KAAACI Work Group on Severe/Recalcitrant Atopic Dermatitis. Current Management of Moderate-to-Severe Atopic Dermatitis: A Survey of Allergists, Pediatric Allergists and Dermatologists in Korea. *Allergy Asthma Immunol. Res.* **2018**, *10*, 253–259. [CrossRef] [PubMed]
14. Usatine, R.P.; Riojas, M. Diagnosis and management of contact dermatitis. *Am. Fam. Physician* **2010**, *82*, 249–255. [PubMed]
15. Maarouf, M.; Hendricks, A.J.; Shi, V.Y. Bathing Additives for Atopic Dermatitis-A Systematic Review. *Dermatitis* **2019**, *30*, 191–197. [CrossRef]
16. Aubert-Wastiaux, H.; Moret, L.; Le Rhun, A.; Fontenoy, A.M.; Nguyen, J.M.; Leux, C.; Misery, L.; Young, P.; Chastaing, M.; Danou, N.; et al. Topical corticosteroid phobia in atopic dermatitis: A study of its nature, origins and frequency. *Br. J. Dermatol.* **2011**, *165*, 808–814. [CrossRef]
17. Weidinger, S.; Novak, N. Atopic dermatitis. *Lancet* **2016**, *387*, 1109–1122. [CrossRef]
18. Loden, M. The clinical benefit of moisturizers. *J. Eur. Acad. Dermatol. Venereol.* **2005**, *19*, 672–688. [CrossRef]
19. Proksch, E.; de Bony, R.; Trapp, S.; Boudon, S. Topical use of dexpanthenol: A 70th anniversary article. *J. Dermatol. Treat.* **2017**, *28*, 766–773. [CrossRef]
20. Nutten, S. Atopic dermatitis: Global epidemiology and risk factors. *Ann. Nutr. Metab.* **2015**, *66* (Suppl. S1), 8–16. [CrossRef]
21. Ha, J.; Lee, S.W.; Yon, D.K. Ten-year trends and prevalence of asthma, allergic rhinitis, and atopic dermatitis among the Korean population, 2008–2017. *Clin. Exp. Pediatr.* **2020**, *63*, 278–283. [CrossRef]
22. Takeuchi, S.; Esaki, H.; Furue, M. Epidemiology of atopic dermatitis in Japan. *J. Dermatol.* **2014**, *41*, 200–204. [CrossRef]
23. Yao, X.; Song, Z.-Q.; Li, W.; Liang, Y.-S.; Zhao, Y.; Cao, H.; Chen, T.; Chen, X.; Feng, A.-P.; Geng, S.-M.; et al. Guidelines for Diagnosis and Treatment of Atopic Dermatitis in China (2020)#. *Int. J. Dermatol. Venereol.* **2021**, *4*, 1–9. [CrossRef]
24. Guo, Y.; Zhang, H.; Liu, Q.; Wei, F.; Tang, J.; Li, P.; Han, X.; Zou, X.; Xu, G.; Xu, Z.; et al. Phenotypic analysis of atopic dermatitis in children aged 1-12 months: Elaboration of novel diagnostic criteria for infants in China and estimation of prevalence. *J. Eur. Acad. Dermatol. Venereol.* **2019**, *33*, 1569–1576. [CrossRef] [PubMed]

25. Im, D.; Yang, Y.S.; Choi, H.R.; Choi, S.; Nahm, H.; Han, K.; Hong, S.-C.; Kim, J.K.; Cho, J.H. Prevalence of Allergic Disease in Korean Adults: Results from the Korea National Health and Nutrition Examination Survey (2010–2012). *Korean J. Otorhinolaryngol. -Head Neck Surg.* **2017**, *60*, 504–511. [CrossRef]
26. Sroka-Tomaszewska, J.; Trzeciak, M. Molecular Mechanisms of Atopic Dermatitis Pathogenesis. *Int. J. Mol. Sci.* **2021**, *22*, 4130. [CrossRef]
27. Fortson, E.; Feldman, S.; Strowd, L. *Management of Atopic Dermatitis*; Springer: New York, NY, USA, 2017.
28. Czarnowicki, T.; He, H.; Krueger, J.G.; Guttman-Yassky, E. Atopic dermatitis endotypes and implications for targeted therapeutics. *J. Allergy Clin. Immunol.* **2019**, *143*, 1–11. [CrossRef]
29. Purnamawati, S.; Indrastuti, N.; Danarti, R.; Saefudin, T. The Role of Moisturizers in Addressing Various Kinds of Dermatitis: A Review. *Clin. Med. Res.* **2017**, *15*, 75–87. [CrossRef]
30. Wu, Y.; Wangari-Olivero, J.; Zhen, Y. ARTICLE: Compromised Skin Barrier and Sensitive Skin in Diverse Populations. *J. Drugs Dermatol.* **2021**, *20*, s17–s22. [CrossRef]
31. Bridgman, A.C.; Block, J.K.; Drucker, A.M. The multidimensional burden of atopic dermatitis: An update. *Ann. Allergy Asthma Immunol.* **2018**, *120*, 603–606. [CrossRef]
32. Jung, H.J.; Lee, D.H.; Park, M.Y.; Ahn, J. Cardiovascular comorbidities of atopic dermatitis: Using National Health Insurance data in Korea. *Allergy Asthma Clin. Immunol.* **2021**, *17*, 94. [CrossRef]
33. Smith Begolka, W.; Chovatiya, R.; Thibau, I.J.; Silverberg, J.I. Financial Burden of Atopic Dermatitis Out-of-Pocket Health Care Expenses in the United States. *Dermatitis* **2021**, *32*, S62–S70. [CrossRef]
34. Wollenberg, A.; Barbarot, S.; Bieber, T.; Christen-Zaech, S.; Deleuran, M.; Fink-Wagner, A.; Gieler, U.; Girolomoni, G.; Lau, S.; Muraro, A.; et al. Consensus-based European guidelines for treatment of atopic eczema (atopic dermatitis) in adults and children: Part II. *J. Eur. Acad. Dermatol. Venereol.* **2018**, *32*, 850–878. [CrossRef]
35. Eichenfield, L.F.; Tom, W.L.; Berger, T.G.; Krol, A.; Paller, A.S.; Schwarzenberger, K.; Bergman, J.N.; Chamlin, S.L.; Cohen, D.E.; Cooper, K.D.; et al. Guidelines of care for the management of atopic dermatitis: Section 2. Management and treatment of atopic dermatitis with topical therapies. *J. Am. Acad. Dermatol.* **2014**, *71*, 116–132. [CrossRef]
36. Kim, J.E.; Kim, H.J.; Lew, B.L.; Lee, K.H.; Hong, S.P.; Jang, Y.H.; Park, K.Y.; Seo, S.J.; Bae, J.M.; Choi, E.H.; et al. Consensus Guidelines for the Treatment of Atopic Dermatitis in Korea (Part I): General Management and Topical Treatment. *Ann. Dermatol.* **2015**, *27*, 563–577. [CrossRef]
37. Lee, J.H.; Kim, J.E.; Park, G.H.; Bae, J.M.; Byun, J.Y.; Shin, M.K.; Han, T.Y.; Hong, S.P.; Jang, Y.H.; Kim, H.O.; et al. Consensus Update for Systemic Treatment of Atopic Dermatitis. *Ann. Dermatol.* **2021**, *33*, 497–514. [CrossRef]
38. Sidbury, R.; Davis, D.M.; Cohen, D.E.; Cordoro, K.M.; Berger, T.G.; Bergman, J.N.; Chamlin, S.L.; Cooper, K.D.; Feldman, S.R.; Hanifin, J.M.; et al. Guidelines of care for the management of atopic dermatitis: Section 3. Management and treatment with phototherapy and systemic agents. *J. Am. Acad. Dermatol.* **2014**, *71*, 327–349. [CrossRef]
39. Piquero-Casals, J.; Carrascosa, J.M.; Morgado-Carrasco, D.; Narda, M.; Trullas, C.; Granger, C.; Fabbrocini, G. The Role of Photoprotection in Optimizing the Treatment of Atopic Dermatitis. *Dermatol. Ther.* **2021**, *11*, 315–325. [CrossRef]
40. Ebner, F.; Heller, A.; Rippke, F.; Tausch, I. Topical use of dexpanthenol in skin disorders. *Am. J. Clin. Dermatol.* **2002**, *3*, 427–433. [CrossRef]
41. Gehring, W.; Gloor, M. Effect of topically applied dexpanthenol on epidermal barrier function and stratum corneum hydration. Results of a human in vivo study. *Arzneimittelforschung* **2000**, *50*, 659–663. [CrossRef]
42. Giam, Y.C.; Hebert, A.A.; Dizon, M.V.; Van Bever, H.; Tiongco-Recto, M.; Kim, K.H.; Soebono, H.; Munasir, Z.; Diana, I.A.; Luk, D.C.K. A review on the role of moisturizers for atopic dermatitis. *Asia Pac. Allergy* **2016**, *6*, 120–128. [CrossRef]
43. Katoh, N.; Ohya, Y.; Ikeda, M.; Ebihara, T.; Katayama, I.; Saeki, H.; Shimojo, N.; Tanaka, A.; Nakahara, T.; Nagao, M.; et al. Japanese guidelines for atopic dermatitis 2020. *Allergol. Int.* **2020**, *69*, 356–369. [CrossRef] [PubMed]
44. Wollenberg, A.; Christen-Zach, S.; Taieb, A.; Paul, C.; Thyssen, J.P.; de Bruin-Weller, M.; Vestergaard, C.; Seneschal, J.; Werfel, T.; Cork, M.J.; et al. ETFAD/EADV Eczema task force 2020 position paper on diagnosis and treatment of atopic dermatitis in adults and children. *J. Eur. Acad. Dermatol. Venereol.* **2020**, *34*, 2717–2744. [CrossRef] [PubMed]
45. Eichenfield, L.F.; Ahluwalia, J.; Waldman, A.; Borok, J.; Udkoff, J.; Boguniewicz, M. Current guidelines for the evaluation and management of atopic dermatitis: A comparison of the Joint Task Force Practice Parameter and American Academy of Dermatology guidelines. *J. Allergy Clin. Immunol.* **2017**, *139*, S49–S57. [CrossRef] [PubMed]
46. Kao, J.S.; Fluhr, J.W.; Man, M.Q.; Fowler, A.J.; Hachem, J.P.; Crumrine, D.; Ahn, S.K.; Brown, B.E.; Elias, P.M.; Feingold, K.R. Short-term glucocorticoid treatment compromises both permeability barrier homeostasis and stratum corneum integrity: Inhibition of epidermal lipid synthesis accounts for functional abnormalities. *J. Investig. Dermatol.* **2003**, *120*, 456–464. [CrossRef]
47. Scott, L.N.; Fiume, M. *Safety Assessment of Panthenol, Pantothenic Acid, and Derivatives as Used in Cosmetics*; Cosmetic Ingredient Review: Washington, DC, USA, 2017.
48. Stettler, H.; Kurka, P.; Lunau, N.; Manger, C.; Bohling, A.; Bielfeldt, S.; Wilhelm, K.P.; Dahnhardt-Pfeiffer, S.; Dahnhardt, D.; Brill, F.H.H.; et al. A new topical panthenol-containing emollient: Results from two randomized controlled studies assessing its skin moisturization and barrier restoration potential, and the effect on skin microflora. *J. Dermatol. Treat.* **2017**, *28*, 173–180. [CrossRef]
49. Gorski, J.; Proksch, E.; Baron, J.M.; Schmid, D.; Zhang, L. Dexpanthenol in Wound Healing after Medical and Cosmetic Interventions (Postprocedure Wound Healing). *Pharmaceuticals* **2020**, *13*, 138. [CrossRef]

50. Etensel, B.; Ozkisacik, S.; Ozkara, E.; Karul, A.; Oztan, O.; Yazici, M.; Gürsoy, H. Dexpanthenol attenuates lipid peroxidation and testicular damage at experimental ischemia and reperfusion injury. *Pediatr. Surg. Int.* **2007**, *23*, 177–181. [CrossRef]
51. Altintas, R.; Parlakpinar, H.; Beytur, A.; Vardi, N.; Polat, A.; Sagir, M.; Odabas, G.P. Protective effect of dexpanthenol on ischemia-reperfusion-induced renal injury in rats. *Kidney Blood Press. Res.* **2012**, *36*, 220–230. [CrossRef]
52. Slyshenkov, V.S.; Omelyanchik, S.N.; Moiseenok, A.G.; Trebukhina, R.V.; Wojtczak, L. Pantothenol protects rats against some deleterious effects of gamma radiation. *Free Radic. Biol. Med.* **1998**, *24*, 894–899. [CrossRef]
53. Mosges, R.; Shah-Hosseini, K.; Hucke, H.P.; Joisten, M.J. Dexpanthenol: An Overview of its Contribution to Symptom Relief in Acute Rhinitis Treated with Decongestant Nasal Sprays. *Adv. Ther.* **2017**, *34*, 1850–1858. [CrossRef]
54. Li-Mei, W.; Jie, T.; Shan-He, W.; Dong-Mei, M.; Peng-Jiu, Y. Anti-inflammatory and Anti-oxidative Effects of Dexpanthenol on Lipopolysaccharide Induced Acute Lung Injury in Mice. *Inflammation* **2016**, *39*, 1757–1763. [CrossRef]
55. Girolomoni, G.; de Bruin-Weller, M.; Aoki, V.; Kabashima, K.; Deleuran, M.; Puig, L.; Bansal, A.; Rossi, A.B. Nomenclature and clinical phenotypes of atopic dermatitis. *Ther. Adv. Chronic. Dis.* **2021**, *12*, 20406223211002979. [CrossRef]
56. Damiani, G.; Calzavara-Pinton, P.; Stingeni, L.; Hansel, K.; Cusano, F.; "Skin Allergy" Group of SIDeMaST; "ADOI" (Associazione Dermatologi Ospedalieri Italiani); "SIDAPA" (Società Italiana di Dermatologia Allergologica, Professionale e Ambientale); Pigatto, P.D.M.; Agostinelli, D.; et al. Italian guidelines for therapy of atopic dermatitis-Adapted from consensus-based European guidelines for treatment of atopic eczema (atopic dermatitis). *Dermatol. Ther.* **2019**, *32*, e13121. [CrossRef]
57. Chan, T.C.; Wu, N.-L.; Wong, L.-S.; Cho, Y.-T.; Yang, C.-Y.; Yu, Y.; Lai, P.-J.; Chang, Y.-T.; Shih, I.-H.; Lee, C.-H.; et al. Taiwanese Dermatological Association consensus for the management of atopic dermatitis: A 2020 update. *J. Formos. Med. Assoc.* **2021**, *120*, 429–442. [CrossRef]
58. Luk, D.; Hon, K.L.E.; Dizon, M.V.C.; Leong, K.F.; Tay, Y.K.; Koh, M.J.; Chandran, N.S.; Wananukul, S.; Chatproedprai, S.; Luger, T. Practical Recommendations for the Topical Treatment of Atopic Dermatitis in South and East Asia. *Dermatol. Ther.* **2021**, *11*, 275–291. [CrossRef]
59. Stettler, H.; de Salvo, R.; Olsavszky, R.; Nanu, E.A.; Dumitru, V.; Trapp, S. Performance and Tolerability of a New Topical Dexpanthenol-Containing Emollient Line in Subjects with Dry Skin: Results from Three Randomized Studies. *Cosmetics* **2021**, *8*, 18. [CrossRef]
60. Ruther, L.; Voss, W. Hydrogel or ointment? Comparison of five different galenics regarding tissue breathability and transepidermal water loss. *Heliyon* **2021**, *7*, e06071. [CrossRef]
61. Proksch, E.; Nissen, H.P. Dexpanthenol enhances skin barrier repair and reduces inflammation after sodium lauryl sulphate-induced irritation. *J. Dermatol. Treat.* **2002**, *13*, 173–178. [CrossRef]
62. Biro, K.; Thaci, D.; Ochsendorf, F.R.; Kaufmann, R.; Boehncke, W.H. Efficacy of dexpanthenol in skin protection against irritation: A double-blind, placebo-controlled study. *Contact Dermat.* **2003**, *49*, 80–84. [CrossRef]
63. Loden, M. Treatments Improving Skin Barrier Function. *Curr. Probl. Dermatol.* **2016**, *49*, 112–122. [CrossRef]
64. Elias, P.M.; Hatano, Y.; Williams, M.L. Basis for the barrier abnormality in atopic dermatitis: Outside-inside-outside pathogenic mechanisms. *J. Allergy Clin. Immunol.* **2008**, *121*, 1337–1343. [CrossRef] [PubMed]
65. Ng, J.P.; Liew, H.M.; Ang, S.B. Use of emollients in atopic dermatitis. *J. Eur. Acad. Dermatol. Venereol.* **2015**, *29*, 854–857. [CrossRef] [PubMed]
66. LePoidevin, L.M.; Lee, D.E.; Shi, V.Y. A comparison of international management guidelines for atopic dermatitis. *Pediatr. Dermatol.* **2019**, *36*, 36–65. [CrossRef] [PubMed]
67. Gelmetti, C.; Metz, M.; Proksch, E. Expert panel on best practices in atopic dermatitis management: Outcome and recommendations. *KOM Dermatol.* **2015**, *10*, 1–4.
68. Ng, S.Y.; Begum, S.; Chong, S.Y. Does Order of Application of Emollient and Topical Corticosteroids Make a Difference in the Severity of Atopic Eczema in Children? *Pediatr. Dermatol.* **2016**, *33*, 160–164. [CrossRef]
69. Glines, K.R.; Stiff, K.M.; Freeze, M.; Cline, A.; Strowd, L.C.; Feldman, S.R. An update on the topical and oral therapy options for treating pediatric atopic dermatitis. *Expert Opin. Pharmacother.* **2019**, *20*, 621–629. [CrossRef]
70. Schneider, L.; Tilles, S.; Lio, P.; Boguniewicz, M.; Beck, L.; LeBovidge, J.; Novak, N.; Bernstein, D.; Blessing-Moore, J.; Khan, D.; et al. Atopic dermatitis: A practice parameter update 2012. *J. Allergy Clin. Immunol.* **2013**, *131*, 295–299. [CrossRef]
71. Chiricozzi, A.; Belloni Fortina, A.; Galli, E.; Girolomoni, G.; Neri, I.; Ricci, G.; Romanelli, M.; Peroni, D. Current therapeutic paradigm in pediatric atopic dermatitis: Practical guidance from a national expert panel. *Allergol. Immunopathol.* **2019**, *47*, 194–206. [CrossRef]
72. Harcharik, S.; Emer, J. Steroid-sparing properties of emollients in dermatology. *Ski. Ther. Lett.* **2014**, *19*, 5–10.
73. Udompataikul, M.; Limpa-o-vart, D. Comparative trial of 5% dexpanthenol in water-in-oil formulation with 1% hydrocortisone ointment in the treatment of childhood atopic dermatitis: A pilot study. *J. Drugs Dermatol.* **2012**, *11*, 366–374.
74. Siegfried, E.C.; Jaworski, J.C.; Kaiser, J.D.; Hebert, A.A. Systematic review of published trials: Long-term safety of topical corticosteroids and topical calcineurin inhibitors in pediatric patients with atopic dermatitis. *BMC Pediatr.* **2016**, *16*, 75. [CrossRef]
75. Schmuth, M.; Wimmer, M.A.; Hofer, S.; Sztankay, A.; Weinlich, G.; Linder, D.M.; Elias, P.M.; Fritsch, P.O.; Fritsch, E. Topical corticosteroid therapy for acute radiation dermatitis: A prospective, randomized, double-blind study. *Br. J. Dermatol.* **2002**, *146*, 983–991. [CrossRef]

76. Huth, S.; Marquardt, Y.; Huth, L.; Schmitt, L.; Prescher, K.; Winterhalder, P.; Steiner, T.; Holzle, F.; Eble, M.; Malte Baron, J. Molecular effects of photon irradiation and subsequent aftercare treatment with dexpanthenol-containing ointment or liquid in 3D models of human skin and non-keratinized oral mucosa. *Exp. Dermatol.* **2021**, *30*, 745–750. [CrossRef]
77. Shanazi, M.; Farshbaf Khalili, A.; Kamalifard, M.; Asghari Jafarabadi, M.; Masoudin, K.; Esmaeli, F. Comparison of the Effects of Lanolin, Peppermint, and Dexpanthenol Creams on Treatment of Traumatic Nipples in Breastfeeding Mothers. *J. Caring Sci.* **2015**, *4*, 297–307. [CrossRef]
78. Weatherhead, S.; Robson, S.C.; Reynolds, N.J. Eczema in pregnancy. *BMJ* **2007**, *335*, 152–154. [CrossRef]
79. Hamdi, I.M. Effect of D-Panthenol on Corneal Epithelial Healing after Surface Laser Ablation. *J. Ophthalmol.* **2018**, *2018*, 6537413. [CrossRef]
80. Olsavszky, R.; Nanu, E.A.; Macura-Biegun, A.; Kurka, P.; Trapp, S. Skin barrier restoration upon topical use of two 5% dexpanthenol water-in-oil formulations on freshly tattooed skin: Results from a single-blind prospective study. *Wounds Int.* **2019**, *10*, 33–39.
81. Celebi, S.; Tepe, C.; Yelken, K.; Celik, O. Efficacy of dexpanthenol for pediatric post-tonsillectomy pain and wound healing. *Ann. Otol. Rhinol. Laryngol.* **2013**, *122*, 464–467. [CrossRef]
82. Gulhas, N.; Canpolat, H.; Cicek, M.; Yologlu, S.; Togal, T.; Durmus, M.; Ozcan Ersoy, M. Dexpanthenol pastille and benzydamine hydrochloride spray for the prevention of post-operative sore throat. *Acta Anaesthesiol. Scand.* **2007**, *51*, 239–243. [CrossRef]
83. Heise, R.; Schmitt, L.; Huth, L.; Krings, L.; Kluwig, D.; Katsoulari, K.-V.; Steiner, T.; Hölzle, F.; Malte, J.; Huth, S. Accelerated wound healing with a dexpanthenol-containing ointment after fractional ablative CO_2 laser resurfacing of photo-damaged skin in a randomized prospective clinical trial. *Cutan. Ocul. Toxicol.* **2019**, *38*, 274–278. [CrossRef]
84. Romiti, R.; Romiti, N. Dexpanthenol cream significantly improves mucocutaneous side effects associated with isotretinoin therapy. *Pediatr. Dermatol.* **2002**, *19*, 368. [CrossRef] [PubMed]
85. Tokura, Y.; Hayano, S. Subtypes of atopic dermatitis: From phenotype to endotype. *Allergol. Int.* **2022**, *71*, 14–24. [CrossRef] [PubMed]
86. Stettler, H.; Kurka, P.; Kandzora, J.; Pavel, V.; Breuer, M.; Macura-Biegun, A. A new topical panthenol-containing emollient for maintenance treatment of childhood atopic dermatitis: Results from a multicenter prospective study. *J. Dermatol. Treat.* **2017**, *28*, 774–779. [CrossRef] [PubMed]
87. Stettler, H.; Kurka, P.; Wagner, C.; Sznurkowska, K.; Czernicka, O.; Bohling, A.; Bielfeldt, S.; Wilhelm, K.P.; Lenz, H. A new topical panthenol-containing emollient: Skin-moisturizing effect following single and prolonged usage in healthy adults, and tolerability in healthy infants. *J. Dermatol. Treat.* **2017**, *28*, 251–257. [CrossRef]
88. National Institute for Health and Care Excellence (NICE). *Atopic Eczema in under 12s: Diagnosis and Management*; Guidelines, N.C., Ed.; NICE: London, UK, 2021.
89. Proksch, E.; Berardesca, E.; Misery, L.; Engblom, J.; Bouwstra, J. Dry skin management: Practical approach in light of latest research on skin structure and function. *J. Dermatol. Treat.* **2020**, *31*, 716–722. [CrossRef]
90. Hon, K.L.; Kung, J.S.C.; Ng, W.G.G.; Leung, T.F. Emollient treatment of atopic dermatitis: Latest evidence and clinical considerations. *Drugs Context* **2018**, *7*, 212530. [CrossRef]
91. Hebert, A.A.; Rippke, F.; Weber, T.M.; Nicol, N.H. Efficacy of Nonprescription Moisturizers for Atopic Dermatitis: An Updated Review of Clinical Evidence. *Am. J. Clin. Dermatol.* **2020**, *21*, 641–655. [CrossRef]
92. Spada, F.; Barnes, T.M.; Greive, K.A. Emollient formulations containing antiseptics reduce effectively the level of Staphylococcus aureus on skin. *Clin. Cosmet. Investig. Dermatol.* **2019**, *12*, 639–645. [CrossRef]
93. Deleuran, M.; Georgescu, V.; Jean-Decoster, C. An Emollient Containing Aquaphilus dolomiae Extract is Effective in the Management of Xerosis and Pruritus: An International, Real-World Study. *Dermatol. Ther.* **2020**, *10*, 1013–1029. [CrossRef]
94. Pugliese, P.; Farina, J.; Chautems, Y. Efficacy of dexpanthenol in wound healing: Double-blind assessment of excised wound tissue by ultrasound and histologic examination. *Nouv. Dermatol.* **1995**, *14*, 130–138.
95. Hartel, M.; Illing, P.; Mercer, J.B.; Lademann, J.; Daeschlein, G.; Hoffmann, G. Therapy of acute wounds with water-filtered infrared-A (wIRA). *GMS Krankenhhyg. Interdiszip.* **2007**, *2*, Doc53.
96. Addor, F.A.S.A.; de Souza, M.C.; Trapp, S.; Peltier, E.; Canosa, J.M. Efficacy and Safety of Topical Dexpanthenol-Containing Spray and Cream in the Recovery of the Skin Integrity Compared with Petroleum Jelly after Dermatologic Aesthetic Procedures. *Cosmetics* **2021**, *8*, 87. [CrossRef]
97. Stables, G.I.; Wilkinson, S.M. Allergic contact dermatitis due to panthenol. *Contact Dermat.* **1998**, *38*, 236–237. [CrossRef]

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Mesenchymal Stem Cells Profile in Adult Atopic Dermatitis and Effect of IL4-IL13 Inflammatory Pathway Inhibition In Vivo: Prospective Case-Control Study

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Abstract: Atopic dermatitis (AD) is an inflammatory disease that typically begins in childhood and may persist into adulthood, becoming a lifelong condition. The major inflammatory mediators of AD are known to be interleukin IL4 and IL13, so Dupilumab, which is able to inhibit both interleukins by blocking the shared IL4Rα subunit, has become an attractive option for treating AD. Mesenchymal stem cells (MSCs) are involved in the onset and development of AD by secreting specific interleukins. The aim of this study was to isolate MSCs from healthy controls (C-MSCs) and patients with AD before (AD-MSCs T0) and after 16 weeks of treatment with Dupilumab (AD-MSCs T16); to evaluate the expression mainly of IL4 and IL13 and of other inflammatory cytokines in C-MSCs, AD-MSCs at T0 and at T16; and to evaluate the efficacy of Dupilumab on MSCs immunobiology. C- and AD-MSCs (T0, T16) were isolated from skin specimens and characterized; the expression/secretion of IL4 and IL13 was evaluated using immuno-cytochemistry (ICC), indirect immune-fluorescence (IIF) and an ELISA test; secretion of IL2, IL4, IL5, IL6, IL10, IL12, IL13, IL17A, Interferon gamma (IFNγ), Tumor necrosis factor alpha (TNFα), Granulocyte Colony-Stimulating Factor (G-CSF), and Transforming Growth Factor beta1 (TGFβ1) were measured with ELISA. IL13 and IL6 were over-expressed, while IL4 was down-regulated in AD-MSCs at T0 compared to C-MSCs. IL6 and IL13 expression was restored after 16 weeks of Dupilumab treatment, while no significant effects on IL4 expression were noted. Finally, IL2, IL5, IL10, IL12, IL17A, INFγ, TNFα, G-CSF, and TGFβ1 were similarly secreted by C- and AD-MSCs. Although Dupilumab blocks the IL4Rα subunit shared by IL4 and IL13, it is evident that its real target is IL13, and its ability to target IL13 in MSCs reinforces the evidence, already known in differentiated cells, of the central role IL13 rather than IL4 in the development of AD. The inflammatory cascade in AD begins at the mesenchymal level, so an upstream therapeutic intervention, able to modify the immunobiology of atopic MSCs, could potentially change the natural history of the disease.

Keywords: psoriasis; atopic dermatitis; mesenchymal stem cells; dupilumab; biologics; therapy; regenerative medicine

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1. Introduction

Atopic dermatitis (AD) is a systemic and immune-allergic inflammatory skin disease; it usually appears in early childhood (15% to 30%) and generally resolves before puberty.

However, in more than half of patients, it can persist into adulthood, becoming a permanent condition [1,2].

Although prevalence of adult AD remains unclear, several studies have indicated that it has increased in recent decades, particularly in industrialized countries [3]. Current estimates place prevalence of AD at around 2–8% in adults, compared to 10–20% in children [4,5].

In AD, the breakdown of the skin barrier results in increased trans-epidermal water loss, reduced skin hydration, and improved antigen presentation by Langerhans cells, which initiate inflammation [6–8].

The largely activated mechanism is the T helper type 2 (Th2) and T helper type 22 (Th22) cascade, with consequent release of cytokines (e.g., interleukin (IL)4, IL13, IL2, IL10, IL17, IL22, Interferon gamma (IFNγ), Tumor necrosis factor alpha (TNFα), Granulocyte Colony-Stimulating Factor (G-CSF), and Transforming Growth Factor beta1 (TGFβ1) [9,10]. In active AD, Th2 inflammation and barrier disruption are characterized by reduced filaggrin and claudin 1 expression, resulting in further exacerbation of the barrier defect and enhanced risk of development of asthma and hay fever, as well as transcutaneous sensitization to a variety of food allergens (e.g., peanuts) [9,11,12].

Although all these immunologic features are well established in AD, the pathogenic model has profoundly changed in the last two decades, overcoming previous hypotheses based on the immune response mediated by immunoglobulin E (IgE) (type 1 hypersensitivity), the primary role of the epidermal barrier impairment ("outside-in" theory), and the primary role of the aberrant immune activation ("inside-out" theory). Previously [13,14], we demonstrated that skin-derived mesenchymal stem cells (MSCs)from AD patients showed strong differences compared to MSCs isolated from skin of healthy subjects. This finding suggests that MSCs may be involved in the pathogenesis of AD.

Dupilumab has entered the therapeutic armamentarium of AD in recent years, due to its selective action on IL4-IL13. Dupilumab is an IL4- receptor α-antagonist that inhibits IL4 and IL13 signaling by blocking the shared IL4 receptor α subunit. The blockade of IL4/13 is effective in reducing Th2 response.

In this study, the relative expression of selected Th1, Th2, and Th17 chemokines/cytokines has been analyzed in MSCs obtained from healthy subjects and adult AD patients, both before and after 16 weeks of treatment with Dupilumab. Recent studies on topical dermatitis, performed, for example, on skin biopsies or peripheral blood, have shown that the Th2 and Th17 responses are characteristic of the acute phase of disease, while the Treg and Th1 response are over-expressed in chronic phase of disease. Thus, it was our intention to evaluate the expression of these molecules in MSCs from patients with atopic dermatitis and compare it with that of MSCs from healthy patients, to delineate the immunophenotype of atopic stem cells in the different stages of disease [15].

2. Materials and Methods

2.1. Design of the Study

The study is a prospective case–control analysis approved by Polytechnic Marche University Ethical Commitee (Protocol 2016 0360 OR) and conducted according to the Declaration of Helsinki.

2.2. Patients' Population

The study group included 11 adult patients (6 males, 5 females, mean age 46.7 ± 12.4), suffering from chronic AD (mean duration of disease 27.3 ± 13.2 years), whereas the control group consisted of 11 adult Caucasian healthy subjects (6 males, 5 females, mean age 49.5 ± 11.7). Diagnosis of AD in adults was made by two independent trained dermatologists, according to Italian AD guidelines [16], and it was essentially based on the typical clinical signs and symptoms of the disease, as currently, no diagnostic markers are available. These include lichenified and minimally inflammatory features of the eczematous skin lesions, the lesions' sharp margins and symmetrical distribution, prevalent localization on

the backs of the hands and fingers and on the volar side of the wrists, the presence of severe pruritus, a chronic clinical course with temporary remissions during summer, association with atopic mucous manifestations, and a positive family history of atopy.

In all enrolled patients, the coexistence of allergic contact dermatitis has been excluded through standardized epicutaneous patch tests conducted according to the SIDAPA (Società Italiana di Dermatologia Allergologia Professionale e Ambientale) guidelines.

All atopic patients were asked to avoid sun exposure and the use of topical and systemic specific treatments (corticosteroids, antihistamines, UVA, PUVA, nb-UVB, cyclosporine, pimecrolimus, and tacrolimus) for at least 4 weeks.

The severity of adult AD for each subject was estimated according to the Eczema Area and Severity Index (EASI), SCORing AD (SCORAD), and Investigator Global Assessment (IGA).

EASI is a validated investigator-assessed scoring system that, by grading the physical signs of atopic dermatitis, determines the severity of the patient's eczema, according to a clinician's perspective [17]; its final score rages from 0 to 72.

SCORAD index is a mixed patient/clinicians' tool used to evaluate AD severity [18], and its final score ranges from 0 to 103.

The Physician Global Assessment (PGA), also referred to as IGA in clinical trials, assesses overall disease severity at a given timepoint on a 6-point severity scale, from clear to very severe disease [19]. Clinical characteristics of erythema, infiltration, papulation, oozing, and crusting are used as guidelines for the overall severity assessment.

2.3. Skin Samples

Atopic patients received a skin punch biopsy in lesional skin both before (T0) and after treatment with Dupilumab 600 mg (two 300 mg subcutaneous injections) once, and then 300 mg subcutaneous injection every other week for 16 weeks (T16).

Control healthy subjects, undergoing surgery for epidermal cysts, received one skin biopsy on healthy skin, after written informed consent was obtained. All punch biopsies were performed with a 5 mm sterile cutaneous skin punch biopsy device (Gima, medical devices, s.r.l. Rome, Italy) after local anesthesia with lidocain 2%. All specimens were obtained from the skin of the chest to standardize the skin specimens between patients and controls.

2.4. Isolation, Cell Culture and Characterization of MSCs

MSCs derived from skin samples were isolated, cultured as previously described [13,14,20], and characterized according to the criteria by Dominici [21] for the identification of MSCs. Briefly, after mincing, the samples were cultured with the Mesenchymal Stem Cell Growth Medium bullet kit (Euroclone, Milan, Italy). The morphology was assessed using phase contrast microscopy (Leica DM IL; Leica Microsystems GmbH, Wetzlar, Germany); the expression of cellular markers HLA-DR, CD14, CD19, CD34, CD45, CD73, CD90, and CD105 was measured using cytofluorimetric analysis to evaluate the immunophenotype, and osteogenic, chondrogenic, and adipogenic differentiation assays were performed as previously described [14]. Cells isolated from control healthy subjects and from AD patients were named C-MSCs and AD-MSCs, respectively.

2.5. IL4 and IL13 Expression by ICC and IIF

Considering the action of Dupilumab on IL4 and IL13, immunocytochemical and immunofluorescence analyzes for IL4 and IL13 were performed on C-MSC and AD-MSC at T0 and T16.

For Immuno-cytochemistry (ICC), 1.5×10^4 cells were incubated overnight with anti-IL4 (R & D Systems, Minneapolis, Canada) or anti-IL13 (Santa Cruz Biotechnology, Dallas, TX, USA) primary antibody. Then, cells were immune-stained using the streptavidin–biotin–peroxidase technique (Dako Cytomation, Milano, Italy) and incubated with 3,3-diaminobenzidine. Slides were counterstained with Mayer's hematoxylin.

For Indirect Immuno-Fluorescence (IIF), we incubated the same number of cells with the anti-IL4 or anti-IL13 antibody, followed by goat anti-mouse FITC-conjugated antibody. Nuclei were visualized using Hoechst 33342 (all from Sigma-Aldrich, St. Louis, MO, USA).

To quantify the expression of the proteins, the percentage of area occupied by the protein within the cells was calculated using Fiji-ImageJ software [22].

2.6. ELISA Test for Evaluation of Cytokines Levels in Supernatant

The levels of cytokines (IL2, IL4, IL5, IL6, IL10, IL12, IL13, IL17A, INFγ, TNFα, G-CSF, and TGFβ1) in the supernatant were determined using a commercial ELISA kit (Multi-Analyte ELISArray™ Kits, Qiagen Multi-Analyte ELISArray kit, Qiagen; ThermoFisher, Waltham, MA USA; Affymetric Ebioscences, Vienna, Switzerland) according to the manufacturer's instructions. Next, 2.5×10^5 cells were plated and cultured with 3 mL of medium; after 72 h, 50 µL of the supernatant was used for the ELISA test.

Briefly, samples (six C-MSCs and nine AD-MSCs at T0 and T16) were dispensed into a 96-well microtiter plate and incubated at room temperature for 2 h. Plates were then washed and reacted with avidin-HRP-conjugated antibody at room temperature for 30′.

After washing, captured cytokines were detected by addition of substrate solution. The OD at 450 and at 570 nm was determined using a microtiter plate reader (Multiskango microplate reader, Thermo Scientific). The concentration of cytokines was determined in pg/mL by comparing the absorbances with those of the antigen standards [23].

2.7. Statistical Analysis

All data were analyzed using Graph-Pad Prism (version 7.0, El Camino REAL, San Diego, CA, USA) and QuickCalcs software package. All data were expressed as means ± SD.

The distribution of continuous variables was verified with a Kolmogorov–Smirnov test. Since data did not assume Gaussian distribution, a nonparametric Kruskal–Wallis test was used for unpaired variables.

For correlation between variables, a computed nonparametric Spearman correlation was used. For all the analyses, a p-value less than 0.05 was statistically significant.

3. Results

3.1. Clinical Features

Treated patients were good responders to Dupilumab and achieved EASI 75 at T16 (Table 1).

Table 1. Absolute values of clinometric indexes in AD patients at baseline (T0) and after 16 weeks of treatment with Dupilumab (T16). (EASI: "Eczema Area and Severity Index"; PGA: "Physician Global Assessment"; SCORAD: "SCORing Atopic Dermatitis").

Patients (n = 11)	EASI T0	SCORAD T0	PGA T0	EASI T16	SCORAD T16	PGA T16
1	45	76	6	11	20	2
2	56	84	6	13	22	2
3	34	62	4	8	15	2
4	33	51	4	7	13	2
5	45	77	5	11	20	2
6	40	67	4	10	17	2
7	41	61	4	10	16	2
8	47	85	5	11	21	2
9	31	50	4	7	12	2
10	34	78	4	9	23	3
11	38	69	3	11	26	2

All median values for clinometrics showed a significant improvement from baseline values at T16 (Table 2).

Table 2. Clinometric changes after 16 weeks of treatment with Dupilumab. (EASI: "Eczema Area and Severity Index"; IGA: "Investigator's Global Assessmen"; SCORAD: "SCORing Atopic Dermatitis").

EASI T0 Mean Value ± SD	EASI T16 Mean Value ± SD	$p < 0.0001$
41.79 ± 2.38	9.81 ± 1.8	
SCORAD T0 Mean Value ± SD	**SCORAD T16 Mean Value ± SD**	$p < 0.0001$
69.09 ± 11.15	18.64 ± 1.32	
IGA T0 Mean Value ± SD	**IGA T16 Mean Value ± SD**	$p < 0.0001$
4.45 ± 0.93	2.09 ± 0.30	

3.2. Isolation, Culture and Characterization of MSCs

MSCs were isolated from the skin of AD patients and control healthy subjects (Figure 1).

Isolated cells met the criteria by Dominici for MSCs identification: they were plastic adherent with a fibroblastoid morphology (Figure 1A), able to differentiate into osteoblasts (Figure 1B), chondrocytes (Figure 1C) and adipocytes (Figure 1D), and with a stem-like immunophenotype (Figure 1E). No differences were found among C-MSCs, AD-MSCs T0, and AD-MSCs T16.

3.3. IL4 and IL13 Expression by ICC and IF in C-MSCs and AD-MSCs at T0 and T16

Since Dupilumab specifically modulates IL4 and IL13, as demonstrated by several previous studies [24,25] their expression was evaluated by ICC and IIF in C-MSC and AD-MSC before and after drug treatment, followed by quantification using Fiji-ImageJ software [22]. Both experimental approaches revealed a significant increase in IL13 expression in AD-MSC (T0) compared to control. After 16 weeks of drug treatment, IL13 expression was restored to levels closer to controls (Figure 2).

In contrast, IL4 expression was significantly reduced in AD-MSC (T0) compared to controls, and drug treatment did not produce any significant change in its expression (Figure 3).

3.4. Expression Profiles of Th1, Th2, Th17 Cytokines by ELISA in C-MSCs and AD-MSCs at T0 and T16

The secretion of 12 cytokines (IL2, IL4, IL5, IL6, IL10, IL12, IL13, IL17A, INFg, TNFa, G-CSF, TGFβ1) was measured by ELISA test in the supernatant of 6 samples of C-MSCs and 9 samples of AD-MSCs, both at baseline and after 16 weeks of treatment with Dupilumab.

IL6 and IL13 were more secreted by AD-MSCs than C-MSCs at T0, and their secretion was directly correlated with disease severity at baseline, according to clinometric indexes EASI, SCORAD and IGA (except for IL6 and IGA) (Figures 4 and 5). The secretion of IL6 and IL13 was significantly reduced after 16 weeks of treatment with Dupilumab, reaching levels closer to controls (Figures 4 and 5; Table 3).

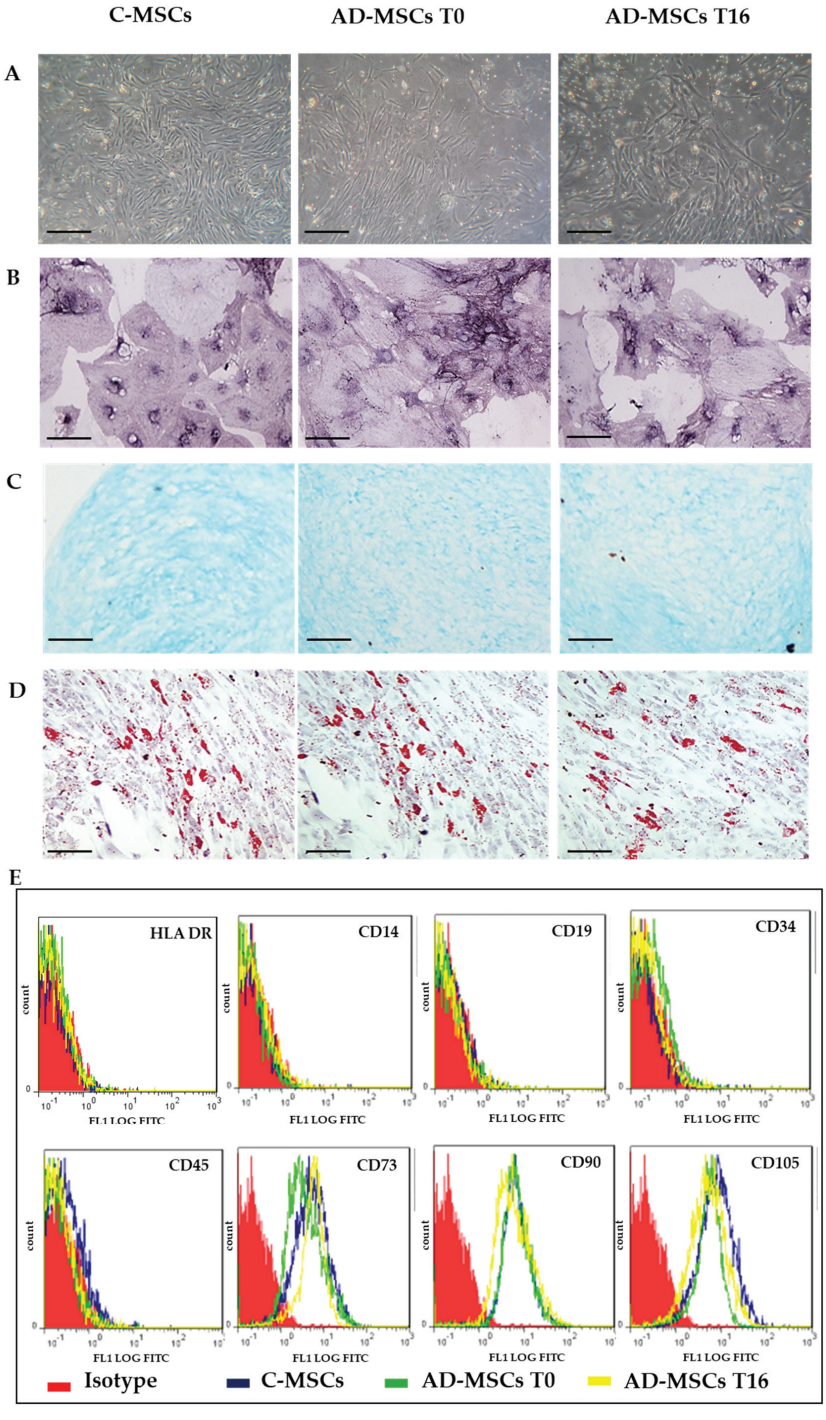


Figure 1. Representative images for Mesenchymal Stem Cells (MSCs) characterization. (**A**) Phase-contrast images of MSCs derived from skin of healthy control subject (C-MSC) and of patients with

atopic dermatitis (AD-MSC) before (T0) and after (T16) treatment with Dupilumab. Scale bar: 100 μm; (**B**) Osteogenic differentiation, ALP staining, Scale bar: 50 μm; (**C**) Chondrogenic differentiation, Alcian blue staining, Scale bar: 200 μm; (**D**) Adipogenic differentiation, Oil red staining, Scale bar: 200 μm; (**E**) Flow cytometry analyses of cell-surface antigen expression, as indicated. Red histograms refer to the negative control (IgG1 isotype control–fluorescein isothiocyanate (FITC) labeled). Blue histograms: C-MSCs; Green: AD-MSCs T0; Yellow: AD-MSCs T16.

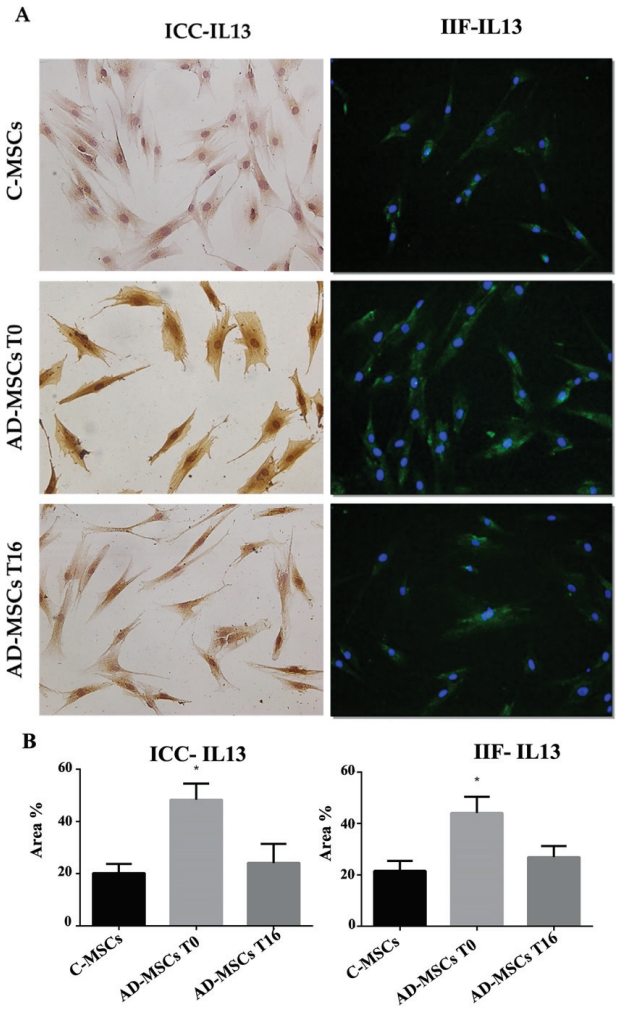


Figure 2. Analysis of the expression of IL13 by Immuno-cytochemistry (ICC), and indirect immune-fluorescence (IIF). (**A**) Representative images of IIF and ICC of IL13 on MSCs from healthy controls (C-MSCs), and MSCs from patients with AD (AD-MSCs) at T0 and at T16. For IIF, a secondary FITC-conjugated antibody was used after incubation with the primary antibodies. Nuclei were counterstained with Hoechst 33342. For ICC, slides were treated with 3,3-diaminobenzidine and counterstained with Mayer's hematoxylin. (Scale bar: 100 μm). (**B**) Quantification of proteins expression processed by Fiji-ImageJ. Protein expression is represented by the percentage of the area it occupies inside the cell. The * indicates significative differences of C-MSCs vs. AD-MSC (unpaired *t*-test; $p < 0.05$).

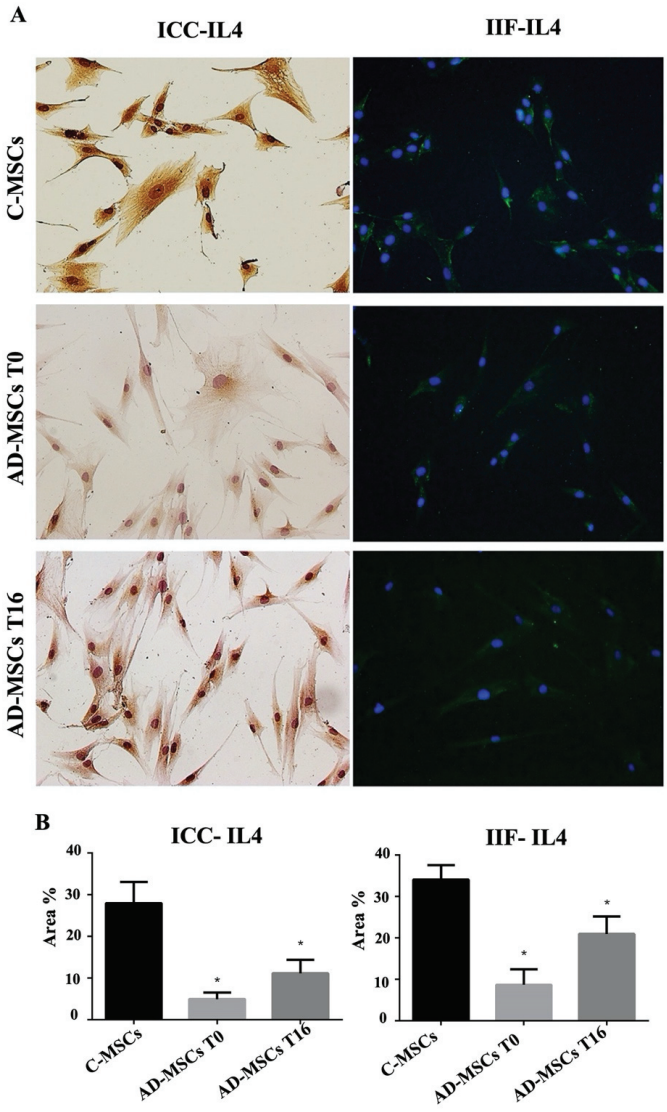
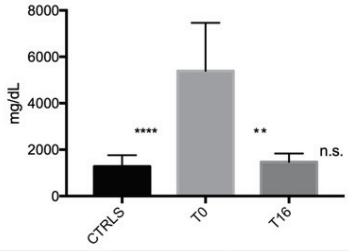


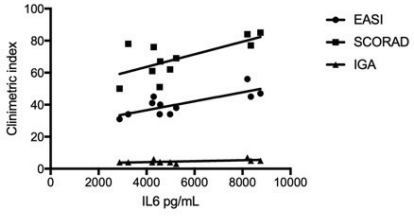
Figure 3. Analysis of the expression of IL4 by Immuno-cytochemistry (ICC) and indirect immune-fluorescence (IIF). (**A**) Representative images of IIF and ICC of IL4 on MSCs from healthy controls (C-MSCs), and MSCs from patients with AD (AD-MSCs)at T0 and at T16. For IIF, a secondary FITC-conjugated antibody was used after incubation with the primary antibodies. Nuclei were counterstained with Hoechst 33342. For ICC, slides were treated with 3,3-diaminobenzidine and counterstained with Mayer's hematoxylin. (Scale bar: 100 μm). (**B**) Quantification of proteins expression processed by Fiji-ImageJ. Protein expression is represented by the percentage of the area it occupies inside the cell. The * indicates significative differences of C-MSCs vs. AD-MSC (unpaired *t*-test; $p < 0.05$).

A



Dunn's multiple comparisons test	Mean rank diff.	Significant?	Summary	Adjusted P Value
T0 vs. CTRLS	18.91	Yes	****	<0.0001
T16 vs. CTRLS	4.818	No	ns	0.7277
T16 vs. T0	-14.09	Yes	**	0.0019

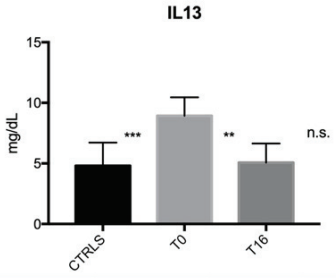
B



	IL6 T0 vs. EASI	IL6 T0 vs. SCORAD	IL6 T0 vs. IGA
R squared	0.6131	0.4508	0.2962
P value			
P (two-tailed)	0.0044	0.0237	0.0835
P value summary	**	*	ns
Significant? (alpha = 0.05)	Yes	Yes	No

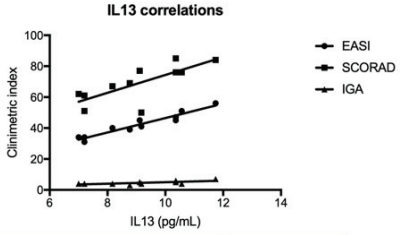
Figure 4. IL6 secretion by C- and AD-MSCs at T0 and T16 and its correlation with AD severity at baseline. (**A**) The histogram depicts the level of secreted IL6 in C-MSCs and in AD-MSCs at T0 and at T16. Secreted protein, evaluated in triplicates by ELISA test, has been reported as pg/mL. ****: $p < 0.0001$, AD-MSCs T0 vs. C-MSCs; **: $p = 0.0019$, AD-MSCs T0 vs. AD-MSCs T16. (**B**) Correlation between IL6 secreted by MSCs at baseline and AD severity according to EASI, SCORAD, and IGA. ** $p = 0.0044$; * $p = 0.0237$.

A



Dunn's multiple comparisons test	Mean rank diff.	Significant?	Summary	Adjusted P Value
T0 vs. CTRLS	15.59	Yes	***	0.0004
T16 vs. CTRLS	1.182	No	ns	>0.9999
T16 vs. T0	-14.41	Yes	**	0.0014

B



	IL13T0 vs. EASI	IL13T0 vs. SCORAD	IL13T0 vs. IGA
R squared	0.9274	0.5729	0.477
P value			
P (two-tailed)	<0.0001	0.0070	0.0186
P value summary	****	**	*
Significant? (alpha = 0.05)	Yes	Yes	Yes

Figure 5. IL13 secretion by C-and AD-MSCs at T0 and T16 and its correlation with AD severity at baseline. (**A**) The histogram depicts the level of secreted IL13 in C-MSCs and in AD-MSCs at T0 and at T16. Secreted protein, evaluated in triplicates by ELISA test, has been reported as pg/mL. ***: $p < = 0.0004$, AD-MSCs T0 vs. C-MSCs; **: $p = 0.0014$, AD-MSCs T0 vs. AD-MSCs T16. (**B**) Correlation between IL13 secreted by MSCs at baseline and AD severity according to EASI, SCORAD, and IGA. **** $p < 0.0001$; ** $p = 0.0070$; * $p = 0.0186$. Conversely, IL4 was secreted at lower levels by AD-MSCs than C-MSCs both at T0 and T16 (Table 3). Finally, IL2, IL5, IL10, IL12, IL17A, TNFα, IFNgamma, G-CSF, TGFβ1 were similarly secreted by C- and AD-MSCs (Table 3).

Table 3. Th1, Th2, Th17 cytokines expression by ELISA in MSCs from healthy controls (C-MSCs) and MSCs from AD patients (AD-MSCs) at T0 and T16. C-MSCs.

IL	MSCs-AD T0 Mean Value ± SD	MSCs-AD T16 Mean Value ± SD	C-MSCs Mean Value ± SD	*p*
IL2	48.98 ± 6.73	65.75 ± 13.83	35.56 ± 8.54	T0 vs. Controls $p = 0.0523$ T0 vs. T16 $p = 0.1299$
IL4	7.14 ± 0.89	9.53 ± 1.31	20.71 ± 1.92	T0 vs. Controls $p < 0.0001$ T0 vs. T16 $p = 0.0777$
IL5	4.40 ± 0.81	3.65 ± 0.55	3.64 ± 0.38	T0 vs. Controls $p = 0.1088$ T0 vs. T16 $p = 0.1213$
IL6	5387.03 ± 2073	1469.01 ± 364.9	1277.02 ± 483.1	T0 vs. Controls $p < 0.0001$ T0 vs. T16 $p = 0.0019$
IL10	29.80 ± 5.82	31.55 ± 6.45	34.66 ± 6.11	T0 vs. Controls $p = 0.1399$ T0 vs. T16 $p > 0.9999$
IL12	3.32 ± 3.12	6.17 ± 3.48	26.38 ± 3.48	T0 vs. Controls $p = 0.1399$ T0 vs. T16 $p > 0.9999$
IL13	8.937 ± 1.51	5.0 ± 1.5	4.807 ± 1.91	T0 vs. Controls $p = 0.0004$ T0 vs. T16 $p = 0.0014$
IL17A	5.90 ± 0.86	6.13 ± 1.19	5.31 ± 0.89	T0 vs. Controls $p = 0.3628$ T0 vs. T16 $p > 0.9999$
IFNγ	6.19 ± 0.92	5.13 ± 0.88	4.81 ± 1.44	T0 vs. Controls $p = 0.1170$ T0 vs. T16 $p = 0.0808$
TNFα	81.46 ± 19.48	66.60 ± 6.55	62.89 ± 11.52	T0 vs. Controls $p = 0.0639$ T0 vs. T16 $p = 0.4878$
G-CSF	94.07 ± 80.53	68.53 ± 38.44	30.93 ± 3.25	T0 vs. Controls $p = 0.6532$ T0 vs. T16 $p > 0.9999$
TGFβ	843.09 ± 170.40	641.10 ± 155.70	759.5 ± 302.70	T0 vs. Controls $p = 0.0807$ T0 vs. T16 $p > 0.9999$

Cytokines' concentration is expressed in pg/mL. Experiments were performed in triplicates.

4. Discussion

Adult AD is a systemic, immune-allergic inflammatory skin disease; the inflammatory pathway most involved in development of AD is the type (Th) 2 immune response, although it has recently been shown that type 1 immune cells contribute to the chronic phase of AD [25].

The course of AD is characterized by biphasic inflammation, the Th2 and Th22 inflammatory pathways predominate at the onset and in the acute phases of disease, with an increase in tissue and serum levels of several cytokines, including IL4, IL5, IL13, IL22, IL31, and TSLP [26,27]. In chronic skin lesions, the prevalence of Th1/Th0 pathways has been described with increased production of IFNγ, TNFa, IL6, IL12, IL17A, and G-CSF [28–31].

The reservoir of MSCs in the skin has been extensively studied in order to understand their possible role in the pathogenesis of several skin diseases, since MSCs are known to modulate the innate and adaptive immune systems [32–37]. However, it has largely been demonstrated that MSCs are strongly influenced by the microenvironment of the so-called "stem cell niche", which is able to drive the physiological MSCs phenotype towards an inflammatory profile, making them a source of pro-inflammatory cytokines capable of amplifying the inflammation according to a vicious circle [13].

In 2017, Orciani et al. [13] demonstrated that MSCs derived from skin samples of adult patients with chronic AD contribute to disease pathogenesis.

The results of the present study support our previous data, confirming the overexpression of IL6 and IL13 in AD-MSCs compared to C-MSCs. As extensively described in the

literature, the lesional skin levels of IL13 and IL6 directly correlate with AD severity in adults [38–42].

In our case series, this trend is also confirmed in MSCs isolated from inflamed skin of adult AD patients, suggesting that the role of IL6 and IL13 can be backdated to MSCs.

The growing evidence of the involvement of IL4 and IL13 in the onset and development of AD has suggested that MSCs may be considered targets for molecular therapy.

The concept that AD may be an IL4-driven disorder emerges from the evidence that it is crucial in the regulation of the IgE synthesis, and several reports have emphasized the high response to and production of IL4 by appropriately activated lymphocytes isolated from AD lesional skin and in vivo overstimulation of the IL4/IL4R pathway [43]. However, despite this common perception that AD is an IL4-driven disease, data have consistently shown that while the expression of IL13 is always detected at high levels regardless of the methodology used, the level of expression of IL4 is changing, from high to low up to undetectable and strictly related to the experimental approach used [43]. These observations may explain our results: the expression of IL4 has been found to be faint, whereas the expression of IL13 was highly detected in all samples. According to our results, it has already been demonstrated that in subacute and chronic AD, IL13 mRNA is more expressed than IL4 mRNA [44,45]. The same trend observed for gene expression was also found at protein level in lesional skin, confirming high levels of IL13 in all skin samples, whereas IL4 expression was low or nondetectable in most patients [46]. Taken together, all these results focus on the idea that IL13 plays a central role in the skin manifestation related to AD, whereas IL4 drives the Th2 response, i.e., for activation in the lymph nodes [47].

All enrolled patients have been treated with Dupilumab for 16 weeks; all of them were good responders, reaching EASI75 (Table 1), and all clinometric (EASI, PGA, and SCORAD) significantly decreased over time (Table 2). Dupilumab is a totally human monoclonal antibody of the IgG4 subclass, a competitive antagonist of the α subunit of IL4Rα, shared by both the IL4 and IL13 receptors, which therefore inhibits intracellular signaling of both interleukins. Until the 2000s, the IL4 was considered the key player of AD, and the choice of Dupilumab in the treatment of AD was initially driven by its ability to block IL4. Nowadays, considering the swinging expression of IL4 compared with the strong and constant IL13 in AD, it is reasonable to suggest that the real target of Dupilumab in AD treatment is IL13. This hypothesis is supported by our results, which indicate that after treatment, both secretion and expression of IL13 (and IL6) by MSCs were normalized, whereas no significant changes have been detected for the other investigated cytokines.

The dual inhibition of IL4 and IL13 has proven to be effective and with a synergic result, but taken individually, IL13 is certainly the major target in treatment strategy for AD. The pathogenetic role of IL-6 is also of interest; like IL-13, it is higher in AD-MSCs than in C-MSCs, and is reduced after Dupilumab administration.

It has already been demonstrated that Dupilumab Suppresses the Activation of Th2 and Th17/Th22, but its action on Th1 Immune Pathways is still unclear. Th1 cells release predominantly IL2, INF-gamma, and IL6 and Th2 cells release IL4, IL5, not INF-gamma. The increased IL6 production by atopic T cells may also result from the activation of a Th2 sub-set, which may represent the target of Dupilumab. However, it is also possible that Dupilumab also acts indirectly on the atopic Th1 subset, which our study results suggest, thereby reducing IL-6 production, but this will need to be investigated in further studies looking at cellular and molecular targets of the drug [48,49].

IL-6 is a pro-inflammatory cytokine produced by macrophages, dendritic cells (DC), and B cells that stimulates the acute-phase response, B-cell maturation, and macrophage differentiation [50]. In AD, IL-6 promotes Th2 differentiation, simultaneously inhibits Th1 polarization, and is involved in the transition from acute to chronic AD [51,52].

The inhibitory effect of Dupilumab on the expression of IL13 and IL6 in MSCs is of particular interest, since IL-6 and IL-13 are key molecules in the pathogenesis of atopic dermatitis, with different levels of involvement according to the clinical phase of disease, the former in the acute, and the latter in the chronic phase.

The evidence of IL6 and IL13 inhibition at the mesenchymal level configures an upstream therapeutic intervention, able to potentially modify the natural history of the disease, both in the acute and chronic phase of disease.

The value of IL-6 as therapeutic target in several inflammatory and immune mediated has been postulated on psoriasis [53] and on atopic dermatitis, as demonstrated by data on Tocilizumab, a humanized monoclonal antibody directed against the IL-6 receptor, which in three case series demonstrated efficacy on pruritus and EASI index, in patients refractory to topical corticosteroid therapy and, in two cases, to cyclosporine [54].

The role of IL6 in psoriasis comorbidities (e.g., depression) has been established [55,56], whereas its involvement in atopic dermatitis comorbidities is far from being proven, although data from literature seem to indicate that the prevalence of depression among atopic patients is higher than in the general population [57], and the role of IL6 need to be investigated in more detail. In this regard, in 2020, He et al. emphasized the potential utility of tape-strip proteomic profiling for tracking biomarkers of therapeutic response in real-life settings, as well as clinical trials and longitudinal studies of AD [48].

However, several emerging items need to be clarified through further studies. It is unclear which pathway dupilumab takes in exerting inhibitory action on IL6 expression, and comprehension of this method of action could have pathogenetic and perhaps therapeutic relevance. Moreover, given the clinical efficacy of Tocilizumab, it might be interesting to evaluate modification of MSCs immunophenotype under the effect of an IL-6 inhibitor. In conclusion, our studies highlight the efficacy of Dupilumab at clinical and subclinical level, focusing on its effect on AD-MSCs immunophenotypic profile.

Further studies focusing on the changes in inflammatory immunophenotype of MSCs obtained from nonresponding patients could be of interest. However, in accordance with the protocol approved by our local ethics committee, we were not allowed to perform skin biopsies to evaluate skin changes in nonresponding patients.

An interesting implication of our results could also be to investigate whether the effect of target molecular therapy on MSCs might be able to restore the typical anti-inflammatory profile of naive MSCs. This could clarify the therapeutic potential of regenerative medicine with the use of autologous MSCs in treating AD [58].

To the best of our knowledge, only one clinical trial (phase I/IIa) has been conducted on human-umbilical-cord-derived mesenchymal stem cells (hUCB-MSCs) to treat adult patients with AD. This trial has demonstrated the potential efficacy of hUCB-MSCs with 50% reduction in EASI in 6 out of 11 subjects, with no reported side effects [59]. Therefore, in future therapeutic strategies for AD, both an increase in knowledge of AD-MSCs and the mechanisms of action exerted on them by the target molecular therapies are crucial to integrate the current pharmacological approach with regenerative medicine, whose preliminary results look promising.

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References

1. Fölster-Holst, R. Management of atopic dermatitis: Are there differences between children and adults? *J. Eur. Acad. Dermatol. Venereol.* **2014**, *28*, 5–8. [CrossRef] [PubMed]
2. Mortz, C.G.; Andersen, K.E.; Dellgren, C.D.; Barington, T.; Bindslev-Jensen, C. Atopic dermatitis from adolescence to adulthood in the TOACS cohort: Prevalence, persistence and comorbidities. *Allergy* **2015**, *70*, 836–845. [CrossRef]
3. Bannister, M.J.; Freeman, S. Adult-onset atopic dermatitis. *Australas. J. Dermatol.* **2000**, *41*, 225–228. [CrossRef]
4. Shaw, T.E.; Currie, G.P.; Koudelka, C.W.; Simpson, E.L. Eczema prevalence in the United States: Data from the 2003 national Survey of Children's Health. *J. Investig. Dermatol.* **2011**, *131*, 67–73. [CrossRef] [PubMed]
5. Barbarot, S.; Auziere, S.; Gadkari, A.; Girolomoni, G.; Puig, L.; Simpson, E.L.; Margolis, D.J.; De Bruin-Weller, M.; Eckert, L. Epidemiology of atopic dermatitis in adults: Results from an international survey. *Allergy* **2018**, *73*, 1284–1293. [CrossRef]
6. De Benedetto, A.; Kubo, A.; Beck, L.A. Skin barrier disruption: A requirement for allergen sensitization? *J. Investig. Dermatol.* **2012**, *132*, 949–963. [CrossRef] [PubMed]
7. Visscher, M.O.; Adam, R.; Brink, S.; Odio, M. Newborn infant skin: Physiology, development, and care. *Clin. Dermatol.* **2015**, *33*, 271–280. [CrossRef] [PubMed]
8. Hanifin, J.M. Evolving concepts of pathogenesis in atopic dermatitis and other eczemas. *J. Investig. Dermatol.* **2009**, *129*, 320–322. [CrossRef]
9. Kondo, H.; Ichikawa, Y.; Imokawa, G. Percutaneous sensiti- zation with allergens through barrier-disrupted skin elicits a Th2-dominant cytokine response. *Eur. J. Immunol.* **1998**, *28*, 769–779. [CrossRef]
10. Correa da Rosa, J.; Malajian, D.; Shemer, A.; Rozenblit, M.; Dhingra, N.; Czarnowicki, T.; Khattri, S.; Ungar, B.; Finney, R.; Xu, H.; et al. Patients with atopic dermatitis have attenuated and distinct con- tact hypersensitivity responses to common allergens in skin. *J. Allergy Clin. Immunol.* **2015**, *135*, 712–720. [CrossRef] [PubMed]
11. Paller, A.S. Latest approaches to treating atopic dermatitis. *Chem. Immunol. Allergy* **2012**, *96*, 132–140. [PubMed]
12. Batista, D.I.S.; Perez, L.; Orfali, R.L.; Zaniboni, M.C.; Samorano, L.P.; Pereira, N.V.; Sotto, M.N.; Ishizaki, A.S.; Oliveira, L.M.S.; Sato, M.N.; et al. Profile of skin barrier proteins (filaggrin, claudins 1 and 4) and Th1/Th2/Th17 cytokines in adults with atopic dermatitis. *J. Eur. Acad. Dermatol. Venereol.* **2015**, *29*, 1091–1095. [CrossRef] [PubMed]
13. Orciani, M.; Campanati, A.; Salvolini, E.; Lucarini, G.; Di Benedetto, G.; Offidani, A.; Di Primio, R. The mesenchymal stem cell profile in psoriasis. *Br. J. Dermatol.* **2011**, *165*, 585–592. [CrossRef]
14. Orciani, M.; Campanati, A.; Caffarini, M.; Ganzetti, G.; Consales, V.; Lucarini, G.; Offidani, A.; Di Primio, R. T helper (Th)1, Th17 and Th2 imbalance in mesenchymal stem cells of adult patients with atopic dermatitis: At the origin of the problem. *Br. J. Dermatol.* **2017**, *176*, 1569–1576. [CrossRef] [PubMed]
15. Su, C.; Yang, T.; Wu, Z.; Zhong, J.; Huang, Y.; Huang, T.; Zheng, E. Differentiation of T-helper cells in distinct phases of atopic dermatitis involves Th1/Th2 and Th17/Treg. *Eur. J. Inflamm.* **2017**, *15*, 46–52. [CrossRef]
16. Patruno, C.; Amerio, P.; Chiricozzi, A.; Costanzo, A.; Cristaudo, A.; Cusano, F.; Foti, C.; Girolomoni, G.; Guarneri, F.; Naldi, L.; et al. Optimizing a clinical guidance for diagnosis of atopic dermatitis in adults: Joint recommendations of the Italian Society of Dermatology and Venereology (SIDeMaST), Italian Association of Hospital Dermatologists (ADOI), and Italian Society of Allergological, Occupational and Environmental Dermatology (SIDAPA). *G. Ital. Dermatol. Venereol.* **2020**, *155*, 1–7.
17. Hanifin, J.M.; Thurston, M.; Omoto, M.; Cherill, R.; Tofte, S.J.; Graeber, M. The eczema area and severity index (EASI): Assessment of reliability in atopic dermatitis. Easi Evaluator Group. *Exp. Dermatol.* **2001**, *10*, 11–18. [CrossRef]
18. Kunz, B.; Oranje, A.; Labrèze, L.; Stalder, J.-F.; Ring, J.; Taïeb, A. Clinical validation and guidelines for the SCORAD index: Consensus report of the European Task Force on Atopic Dermatitis. *Dermatology* **1997**, *195*, 10–19. [CrossRef]
19. Rehal, B.; Armstrong, A.W. Health outcome measures in atopic dermatitis: A systematic review of trends in disease severity and quality-of-life instruments 1985–2010. *PLoS ONE* **2011**, *6*, e17520. [CrossRef]
20. Campanati, A.; Orciani, M.; Consales, V.; Lazzarini, R.; Ganzetti, G.; di Benedetto, G.; di Primio, R.; Offidani, A. Characterization and profiling of immunomodulatory genes in resident mesenchymal stem cells reflect the Th1-Th17/Th2 imbalance of psoriasis. *Arch. Dermatol. Res.* **2014**, *306*, 915–920. [CrossRef]
21. Dominici, M.; Le Blanc, K.; Mueller, I.; Slaper-Cortenbach, I.; Marini, F.C.; Krause, D.S.; Deans, R.J.; Keating, A.; Prockop, D.J.; Horwitz, E.M. Minimal criteria for defining multipotent mesenchymal stromal cells. The International Society for Cellular Therapy position statement. *Cytotherapy* **2006**, *8*, 315–317. [CrossRef] [PubMed]
22. Rueden, C.T.; Schindelin, J.; Hiner, M.C.; Dezonia, B.E.; Walter, A.E.; Arena, E.T.; Eliceiri, K.W. ImageJ2: ImageJ for the next generation of scientific image data. *BMC Bioinform.* **2017**, *18*, 529. [CrossRef] [PubMed]
23. Campanati, A.; Orciani, M.; Lazzarini, R.; Ganzetti, G.; Consales, V.; Sorgentoni, G.; Di Primio, R.; Offidani, A. TNF-α inhibitors reduce the pathological Th1–Th17/Th2 imbalance in cutaneous mesenchymal stem cells of psoriasis patients. *Exp. Dermatol.* **2017**, *26*, 319–324. [CrossRef] [PubMed]
24. Hamilton, J.D.; Suárez-Fariñas, M.; Dhingra, N.; Cardinale, I.; Li, X.; Kostic, A.; Ming, J.E.; Radin, A.R.; Krueger, J.G.; Graham, N.; et al. Dupilumab improves the molecular signature in skin of patients with moderate-to-severe atopic dermatitis. *J. Allergy Clin. Immunol.* **2014**, *134*, 1293–1300. [CrossRef]
25. Ariens, L.F.; Bakker, D.S.; Van Der Schaft, J.; Garritsen, F.M.; Thijs, J.L.; De Bruin-Weller, M.S. Dupilumab in atopic dermatitis: Rationale, latest evidence and place in therapy. *Ther. Adv. Chronic Dis.* **2018**, *9*, 159–170. [CrossRef]

26. Klonowska, J.; Gleń, J.; Nowicki, R.J.; Trzeciak, M. New cytokines in the pathogenesis of atopic dermatitis—New therapeutic targets. *Int. J. Mol. Sci.* **2018**, *19*, 3086. [CrossRef]
27. Gittler, J.K.; Shemer, A.; Suárez-Fariñas, M.; Fuentes-Duculan, J.; Gulewicz, K.J.; Wang, C.Q.F.; Mitsui, H.; Cardinale, I.; de Guzman Strong, C.; Krueger, J.G.; et al. Progressive activation of T(H)2/T(H)22 cytokines and selective epidermal proteins characterizes acute and chronic atopic dermatitis. *J. Allergy Clin. Immunol.* **2012**, *130*, 1344–1354. [CrossRef]
28. Chaudhary, S.K.; Singh, S.K.; Kumari, P.; Kanwal, S.; Soman, S.P.; Choudhury, S.; Garg, S.K. Alterations in circulating concentrations of IL-17, IL-31 and total IgE in dogs with atopic dermatitis. *Vet. Dermatol.* **2019**, *30*, 383-e114. [CrossRef]
29. Nutten, S. Atopic demartitis: Global epidemiology and risk factors. *Ann. Nutr. Metab.* **2015**, *66* (Suppl. 1), 8–18. [CrossRef]
30. Miossec, P.; Kolls, J.K. Targeting IL-17 and TH17 cells in chronic Inflammation. *Nat. Rev. Drug Discov.* **2012**, *11*, 763–776. [CrossRef]
31. Sidburry, R.; Khorsand, K. Evolving concepts in atopic dermatitis. *Curr. Allergy Asthma Rep.* **2017**, *17*, 42. [CrossRef] [PubMed]
32. Campanati, A.; Orciani, M.; Sorgentoni, G.; Consales, V.; Offidani, A.; Di Primio, R. Pathogenetic Characteristics of Mesenchymal Stem Cells in Hidradenitis Suppurativa. *JAMA Dermatol.* **2018**, *154*, 1184–1190. [CrossRef] [PubMed]
33. Campanati, A.; Consales, V.; Orciani, M.; Giuliodori, K.; Ganzetti, G.; Bobyr, I.; Sorgentoni, G.; Di Primio, R.; Offidani, A. Role of mesenchymal stem cells in the pathogenesis of psoriasis: Current perspectives. *Psoriasis* **2017**, *7*, 73–85. [CrossRef] [PubMed]
34. Campanati, A.; Orciani, M.; Sorgentoni, G.; Consales, V.; Mattioli Belmonte, M.; Di Primio, R.; Offidani, A. Indirect co-cultures of healthy mesenchymal stem cells restore the physiological phenotypical profile of psoriatic mesenchymal stem cells. *Clin. Exp. Immunol.* **2018**, *193*, 234–240. [CrossRef]
35. Campanati, A.; Orciani, M.; Ganzetti, G.; Consales, V.; Di Primio, R.; Offidani, A. The effect of etanercept on vascular endothelial growth factor production by cutaneous mesenchymal stem cells from patients with psoriasis. *J. Int. Med. Res.* **2016**, *44* (Suppl. 1), 6–9. [CrossRef]
36. Campanati, A.; Orciani, M.; Gorbi, S.; Regoli, F.; Di Primio, R.; Offidani, A. Effect of biologic therapies targeting tumour necrosis factor-α on cutaneous mesenchymal stem cells in psoriasis. *Br. J. Dermatol.* **2012**, *167*, 68–76. [CrossRef]
37. Campanati, A.; Caffarini, M.; Diotallevi, F.; Radi, G.; Lucarini, G.; Di Vincenzo, M.; Orciani, M.; Offidani, A. The efficacy of in vivo administration of Apremilast on mesenchymal stem cells derived from psoriatic patients. *Inflamm. Res.* **2020**, *70*, 79–87. [CrossRef]
38. Campione, E.; Lanna, C.; Diluvio, L.; Cannizzaro, M.V.; Grelli, S.; Galluzzo, M.; Talamonti, M.; Annicchiarico-Petruzzelli, M.; Mancini, M.; Melino, G.; et al. Skin immunity and its dysregulation in vitiligo, atopic dermatitis, hidradenitis supurativa and vitiligo. *Cell Cycle* **2020**, *19*, 257–267. [CrossRef]
39. Gharagozlou, M.; Farhadi, E.; Khaledi, M.; Behniafard, N.; Sotoudeh, S.; Salari, R.; Darabi, B.; Fathi, S.M.; Mahmoudi, M.; Aghamohammadi, A.; et al. Association between the interleukin 6 genotype at position-174 and atopic dermatitis. *J. Investig. Allergol. Clin. Immunol.* **2013**, *23*, 89–93.
40. Raap, U.; Weißmantel, S.; Gehring, M.; Eisenberg, A.M.; Kapp, A.; Fölster-Holst, R. IL-31 significantly correlates with disease activity and Th2 cytokine levels in children with atopic dermatitis. *Pediatr. Allergy Immunol.* **2012**, *23*, 285–288. [CrossRef]
41. Ungar, B.; Garcet, S.; Gonzalez, J.; Dhingra, N.; Da Rosa, J.C.; Shemer, A.; Krueger, J.G.; Suarez-Farinas, M.; Guttman-Yassky, E. An integrated model of atopic dermatitis biomarkers highlights the systemic nature of the disease. *J. Investig. Dermatol.* **2017**, *137*, 603–613. [CrossRef] [PubMed]
42. Szegedi, K.; Lutter, R.; Bos, J.D.; Luiten, R.M.; Kezic, S.; Middelkamp-Hup, M.A. Cytokine profiles in intersti- tial fluid from chronic atopic dermatitis skin. *J. Eur. Acad. Dermatol. Venereol.* **2015**, *29*, 2136–2144. [CrossRef] [PubMed]
43. Bieber, T. Interleukin-13: Targeting an underestimated cytokine in atopic dermatitis. *Allergy* **2020**, *75*, 54–62. [CrossRef] [PubMed]
44. Tazawa, T.; Sugiura, H.; Sugiura, Y.; Uehara, M. Relative importance of IL-4 and IL-13 in lesional skin of atopic dermatitis. *Arch. Dermatol. Res.* **2004**, *295*, 459–464. [CrossRef]
45. Tsoi, L.C.; Rodriguez, E.; Degenhardt, F.; Baurecht, H.; Wehkamp, U.; Volks, N.; Szymczak, S.; Swindell, W.R.; Sarkar, M.K.; Raja, K.; et al. Atopic dermatitis is an IL-13–dominant disease with greater molecular heterogeneity compared to psoriasis. *J. Investig. Dermatol.* **2019**, *139*, 1480–1489. [CrossRef]
46. Koppes, S.A.; Brans, R.; Ljubojevic Hadzavdic, S.; Frings-Dresen, M.H.; Rustemeyer, T.; Kezic, S. Stratum Corneum Tape Stripping: Monitoring of Inflammatory Mediators in Atopic Dermatitis Patients Using Topical Therapy. *Int. Arch. Allergy Immunol.* **2016**, *170*, 187–193. [CrossRef] [PubMed]
47. Bao, K.; Reinhardt, R.L. The differential expression of IL-4 and IL-13 and its impact on type-2 immunity. *Cytokine* **2015**, *75*, 25–37. [CrossRef] [PubMed]
48. He, H.; Olesen, C.M.; Pavel, A.B.; Clausen, M.-L.; Wu, J.; Estrada, Y.; Zhang, N.; Agner, T.; Guttman-Yassky, E. Tape-Strip Proteomic Profiling of Atopic Dermatitis on Dupilumab Identifies Minimally Invasive Biomarkers. *Front. Immunol.* **2020**, *11*, 1768. [CrossRef] [PubMed]
49. Toshitani, A.; Ansel, J.C.; Chan, S.C.; Li, S.-H.; Hanifin, J.M. Increased interleukin 6 production by T cells derived from patients with atopic dermatitis. *J. Investig. Dermatol.* **1993**, *100*, 299–304. [CrossRef]
50. Diehl, S.; Rincon, M. The two faces of IL-6 on Th1/Th2 differentiation. *Mol. Immunol.* **2002**, *39*, 531–536. [CrossRef]
51. Howell, M.D.; Parker, M.L.; Mustelin, T.; Ranade, K. Past, present, and future for biologic intervention in atopic dermatitis. *Allergy* **2015**, *70*, 887–896. [CrossRef]

52. Saeki, H.; Kabashima, K.; Tokura, Y.; Murata, Y.; Shiraishi, A.; Tamamura, R.; Randazzo, B.; Imanaka, K. Efficacy and safety of Ustekinumab in Japanese patients with severe atopic dermatitis: A randomised, double-blind, placebo-controlled, phase 2 study. *Br. J. Dermatol.* **2017**, *177*, 419–427. [CrossRef] [PubMed]
53. Sobolev, V.V.; Denisova, E.V.; Chebysheva, S.N.; Geppe, N.A.; Korsunskaya, I.M. IL-6 Gene Expression as a Marker of Pathological State in Psoriasis and Psoriatic Arthritis. *Bull. Exp. Biol. Med.* **2022**, *173*, 77–80. [CrossRef]
54. Navarini, A.; French, L.; Hofbauer, G. Interrupting IL-6–receptor signaling improves atopic dermatitis but associates with bacterial superinfection. *J. Allergy Clin. Immunol.* **2011**, *128*, 1128–1130. [CrossRef] [PubMed]
55. Chatrath, S.; Lei, D.; Yousaf, M.; Chavda, R.; Gabriel, S.; Silverberg, J.I. Longitudinal course and predictors of depressive symptoms in atopic dermatitis. *J. Am. Acad. Dermatol.* 2022; *in press*. [CrossRef]
56. Bardazzi, F.; Magnano, M.; Campanati, A.; Loconsole, F.; Carpentieri, A.; Potenza, C.; Bernardini, N.; Lernia, V.; Carrera, C.; Raone, B.; et al. Biologic Therapies in HIV-infected Patients with Psoriasis: An Italian Experience. *Acta Derm. Venereol.* **2017**, *97*, 989–990. [CrossRef]
57. Marek-Józefowicz, L.; Jaracz, M.; Borkowska, A. Affective temperament, depressive symptoms and interleukins in patients with psoriasis. *Postepy Dermatol. Alergol.* **2021**, *38*, 137–143. [CrossRef] [PubMed]
58. Daltro, S.R.T.; Meira, C.S.; Santos, I.P.; Dos Santos, R.R.; Soares, M.B.P. Mesenchymal Stem Cells and Atopic Dermatitis: A Review. *Front. Cell Dev. Biol.* **2020**, *8*, 326. [CrossRef] [PubMed]
59. Kim, H.-S.; Lee, J.H.; Roh, K.-H.; Jun, H.J.; Kang, K.-S.; Kim, T.-Y. Clinical trial of human umbilical cord blood-derived stem cells for the treatment of moderate-to-severe atopic dermatitis: Phase I/IIa studies. *Stem Cells* **2017**, *35*, 248–255. [CrossRef] [PubMed]

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Review

New and Upcoming Topical Treatments for Atopic Dermatitis: A Review of the Literature

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Abstract: Atopic dermatitis (AD) is a chronic inflammatory dermatosis with periods of exacerbation and remissions. AD is characterized by intense, persistent pruritus and heterogeneity in clinical symptomatology and severity. Therapeutic goals include the amelioration of cutaneous eruptions, diminishing relapses and eventually the disease burden. To date, topical corticosteroids (TCS) and calcineurin inhibitors (TCI) have yet been deemed the mainstay of topical treatments in AD management. Nevertheless, despite their indisputable efficiency, TCS and TCI are not indicated for continuous long-term use given their safety profile. While research in AD has concentrated predominantly on systemic therapies, more than 30 novel topical compounds are under development. The existing data appear encouraging, with some regimens that are already FDA-approved (ruxolitinib was the most recent in September 2021) and several pharmaceutical pipeline products for mild-to-moderate AD that are in an advanced stage of development, such as tapinarof, difamilast and roflumilast. Larger, long-term studies are still required to evaluate the efficacy and safety of these novel compounds in the long run and weigh their advantages over present treatments. In this review, we aim to provide an overview of the latest knowledge about AD topical treatments, echoing upcoming research trends.

Keywords: atopic dermatitis; topical treatment; JAK inhibitors; PDE-4 inhibitors

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1. Introduction

Atopic dermatitis (AD) is a common inflammatory skin disease affecting as much as 25% of children and up to 10% of adults [1].

Prevalence depends mainly on genetic and socio-economic factors, with developed countries being more affected, while environmental factors, such as latitude and UV exposure, also play a role [2]. As the child grows, the disease improves or completely resolves in more than 50% of the patients over 6 years old. In some cases, nevertheless, AD persists or even starts in adulthood [3]. The main clinical characteristics of the disease are pruritus, eczematous lesions usually in age-specific body sites, dry skin and chronic courses with relapses and remissions.

AD has a multifactorial etiology, including immune system dysfunction, an impaired skin barrier and genetic and environmental contributing factors [4,5]. Although the interaction among those factors is not fully understood, it is clear that their synergy leads to a defective skin barrier that is unable to preserve moisture. Skin becomes dry, irritated, erythematous, exudative and prone to infection. Some lesions, after extensive scratching, become lichenified.

The goal of AD management is to treat the skin-barrier defect and inflammation and to restore the microbiome, thus obtaining prolonged patient remission. Topical therapies

are key players in achieving those goals. They provide targeted anti-inflammatory activity and improve skin pathology with lower costs and increased safety compared to systemic treatments.

For more than 50 years, the cornerstone of topical AD treatment has been emollients and topical corticosteroids (TCS) [6]. In 2000, topical calcineurin inhibitors (TCI) were introduced, and no significant progress has been made ever since. Until recently, molecular targeting therapies began to emerge, and a revolutionary era started for medicine. Dermatology is one of the most privileged specialties in this field, given the plethora of biologics, Janus kinase and phosphodiesterase-4 inhibitors and other new molecules available for the treatment of various chronic diseases. In this review, we present the latest topical treatments for AD, including not only those that are already approved but also those in the pipeline. To find the data presented here, we conducted an extensive search in Medline, ScienceDirect and Google Scholar with various combinations of the search terms, including "Atopic Dermatitis", "Eczema", "treatment", "development", "therapy" and "new", "emerging" and "upcoming". We also used the terms "JAK inhibitors", "PDE-4 inhibitors", "Aryl Hydrocarbon Receptor Agonists" and specific drug names found in the literature. The "Reference" section of relevant manuscripts was hand-searched to maximize the sensitivity of our search. We also searched the archives of major recent dermatology conferences and contacted some of the developers for information that we could not find elsewhere. Two authors (N.S. and E.P.) searched clinicaltrials.gov and clinicaltrialsregister.eu with Atopic Dermatitis as the only term. Several hundred studies were identified and searched one by one to find those concerning topical treatments. Apart from the data contained in the two registries, all identified drug names were used as search terms both in Medline and in websites for the general public/search engines to identify more medical literature and other information as press releases.

As this review includes many types of evidence, the risk of bias of included sources was not examined.

2. Janus Kinase Inhibitors

Janus kinases (JAKs) is a family of intracellular tyrosine kinases comprising four members (JAK1, 2, 3 and tyrosine kinase 2 [TYK2]). The JAKs, along with Signal Transducer and Activator of Transcription (STAT) proteins (STAT-1,-2,-3,-4,-5a,-5b and -6) and transmembrane receptors are the three main parts of the JAK-STAT pathway [7].

A vast array of hormones, interferons, colony-stimulating factors and interleukins exert their actions through the JAK-STAT pathway [8]. Receptors for those factors rely on JAKs for downstream responses and subsequent modulation of gene expression. Pairs of JAKs, varying by receptor, bind to them with different results for each dimeric cytokine receptor–JAK pair. The now activated JAKs phosphorylate the receptors, forming a docking site for STATs. Those are then also phosphorylated and migrate to nucleus, affecting gene expression [9–11].

The vital role of JAKs in human physiology was evident upon their discovery. Involved in both hematopoiesis and immunity, JAKs became a treatment target for numerous diseases across various medical specialties [12,13].

The role of JAKs in the pathogenesis of AD is complex. They exaggerate Th2 cell response, activate eosinophils, suppress regulatory T cells (Tregs), upregulate epidermal chemokines, pro-inflammatory cytokines, etc. [14]. Building on this concept, their inhibition has been considered a promising treatment option.

2.1. Delgocitinib

On 23 January 2020, in Japan, delgocitinib became the first topical JAK inhibitor approved for the treatment of AD. Delgocitinib is a first-generation pan-JAK inhibitor blocking the activation of inflammatory cells (T and B), monocytes and mast cells and improving skin-barrier dysfunction [15]. Three major randomized phase-3 trials (RCTs) of

topical delgocitinib for AD have been held: (i) JapicCTI-173554, (ii) JapicCTI-173555 and (iii) JapicCTI-184064 (www.clinicaltrials.jp, accessed on 1 June 2022).

In JapicCTI-173554, Japanese patients over 16 years of age with modified Eczema Area and Severity Index (mEASI) score > 10, Investigator's Global Assessment (IGA) score of 3 or 4 and body surface area (BSA) involvement of 10% to 30% were eligible for enrollment. Part 1 was a 4-week, randomized, double-blind, vehicle-controlled study. After completion of part 1, patients could enter part 2, a 24-week, open-label extension study. Patients with worsening AD during part 1 could discontinue treatment or enter part 2 at the investigators' discretion. In part 2, all patients received delgocitinib 0.5% ointment.

In part 1, the least-squares mean percent changes from baseline in mEASI score were −44.3% in the delgocitinib group and 1.7% in the vehicle group at the end of treatment. Reduction in mEASI score in the delgocitinib group started from week 1 and continued to week 4. IGA and pruritus Numerical Rating Scale (NRS) scores were also improved in the delgocitinib group compared with the vehicle group (p = 0.32 for the overall IGA score, 0.05 for the IGA face/neck score, 0.01 for NRS).

The proportion of patients with a mEASI-50 was 51.9% in the delgocitinib group and 11.5% in the vehicle group. The proportion of patients with a mEASI-75 was 26.4%in the delgocitinib group and 5.8% in the vehicle group.

Improvements in all AD parameters (mEASI, IGA, pruritus NRS scores and the percentage of patients with mEASI-50 and mEASI-75) persisted at the second part of the study. At week 24, the mean percent change from baseline in the mEASI score was −56.3%, and the proportions of patients with mEASI-50 and mEASI-75 were 69.3% and 35.8%, respectively [16].

JapicCTI −173,555 was an open-label, multicenter, phase 3 study, with Japanese patients over 16 years old with mild to severe AD (IGA 2 to 4, BSA 5–30%) and longer treatment durations (52 weeks). Improvement in all AD scores with delgocitinib 0.5% ointment b.i.d persisted for the study duration. The proportion of patients with mEASI-50 was 31.5% at week 4, 42.3% at week 24 and 51.9% at week 52. The proportion of patients with mEASI-75 was 10.9% at week 4, 22.7% at week 24 and 27.5% at week 52 [17].

The last study, JapicCTI-184064, evaluated the safety and efficacy of topical delgocitinib in pediatric patients aged 2 to 15 years old. Part 1 was a 4-week double-blind period in which patients were randomized in a 1:1 ratio to delgocitinib 0.25% ointment or vehicle. Part 2 was a 52-week open-label extension period. Eligible patients who received the vehicle treatment in part 1 were also treated with delgocitinib 0.25% or 0.5% ointment in part 2, according to the investigator's judgment. In the first four weeks, 50.7% of patients in the delgocitinib group compared with 17.6% of patients in the vehicle group achieved a mEASI-50. mEASI-75 was achieved by 37.7% of patients in the delgocitinib group compared with 4.4% of patients in the vehicle group. In part 2, improvements in mEASI, IGA and pruritus scores persisted through week 56. Around 73.6% and 52.8% of the patients who received the drug in part 1, achieved mEASI-50 and mEASI-75, respectively. In patients who received vehicle in part 1, the percentage who achieved mEASI-50 and mEASI-75 at week 52 was 70.5% and 52.5%, respectively [18].

In all studies, adverse events (AEs) related to the drug were mostly mild. The most common AE was nasopharyngitis, followed by application site folliculitis and acne, influenza, Kaposi's varicelliform eruption, herpes simplex, impetigo, fungal infections, molluscum contagiosum, etc.

Recently, a small, retrospective, 4-week Japanese study with 25 patients was published, in which delgocitinib was compared to topical corticosteroids as twice-weekly maintenance therapy in adults with AD [19].

AD parameters did not differ significantly between the two groups. Mean changes in NRS score and erythema index were slightly better in the TCS treated areas. However, the stratum corneum hydration in the delgocitinib group was maintained, while that of the TCS group worsened. The authors conclude that topical delgocitinib may be an effective maintenance therapy of AD in dry skin patients, sparing long-term corticosteroids usage.

2.2. Ruxolitinib

Ruxolitinib is a first-generation potent JAK1/2 inhibitor. Its oral form revolutionized the treatment of JAK2-driven myeloproliferative disorders and is currently FDA approved for polycythemia vera, myelofibrosis and steroid-refractory acute graft-versus-host disease [20].

The topical form was approved by the FDA on September 2021 for the short-term and non-continuous chronic treatment of mild to moderate AD in non-immunocompromised patients over 12 years of age. Topical Ruxolitinib Evaluation in Atopic Dermatitis (TRuE-AD) 1 and 2 were the two major, identical, phase 3 trials that confirmed the antipruritic and anti-inflammatory effects of the drug in patients with AD.

A total of 631 patients in TRuE-AD 1 and 618 in TRuE-AD 2 were randomized 2:2:1 to twice-daily 0.75% cream, 1.5% cream or vehicle cream for the first 8 weeks of the studies. Key inclusion criteria included age $\geq$ 12 years, diagnosis of AD for $\geq$2 years, IGA score 2–3 and BSA involvement of 3–20%. After the 8-week period, eligible patients continued treatment for an additional 44 weeks with 0.75% or 1.5% ruxolitinib cream. Patients initially randomized to the drug remained on their treatment and those randomized to vehicle were re-randomized to either cream regimen.

The primary endpoint of IGA 0–1 at week 8 was achieved by 50% and 39% of patients in TRuE-AD1 and TRuE-AD2, respectively, on 0.75% cream. For 1.5% cream, the percentage of patients achieving IGA 0–1 was 53.8% and 51.3%, while 15.1% and 7.6% ($p < 0.0001$) were achieved for vehicle only. EASI-75 at week 8 was achieved by 56.0% and 51.5% of patients on 0.75% cream, 62.1% and 61.8% on 1.5% cream and 24.6% and 14.4% on vehicle, respectively. Improvements in EASI-90 and NRS score were also significant ($p < 0.05$). It is worth mentioning that a reduction in itching started just 12 **h** after treatment initiation with 1.5% ruxolitinib cream [21].

The most common treatment-related AE was application-site burning sensation, mainly observed with vehicle (4.4%) than with 0.75% (0.6%) or 1.5% (0.8%) cream. None of the reported AEs were serious.

In a long-term safety period, up to week 52, patients were to treat skin areas with active AD only and stop treatment 3 days after clearance of lesions. They were to restart at the first sign of recurrence.

The proportion of patients with no/minimal AD lesions (IGA score 0/1) with ruxolitinib 0.75% and 1.5% cream ranged from 62.4% to 76.9% and from 66.5% to 77.3%, respectively, in TRuE-AD1 and from 59.6% to 76.7% and from 72.0% to 80.1% in TRuE-AD2 from weeks 12 to 52. In both studies, the mean measured total affected BSA was <3% throughout the long-term safety period in the ruxolitinib 1.5% cream and most of the period in the ruxolitinib 0.75% cream. Treatment-emergent AEs were reported in 60.1% and 53.8% patients who applied ruxolitinib 0.75% and 1.5%, respectively. Treatment-related AEs were reported in 20 patients (4.7%) who applied ruxolitinib 0.75% cream and in 13 patients (2.9%) who applied ruxolitinib 1.5% cream: none were serious, with upper respiratory tract infection, nasopharyngitis and influenza being the most common.

Treatment-emergent AEs resulted in discontinuations in nine patients (2.1%) in the ruxolitinib 0.75% cream group and no patients in the ruxolitinib 1.5% cream group. In summary, approximately 70% of patients maintained no or minimal lesions (IGA score 0/1) and the extent of AD lesions remained low during the 44-week extension period of TRuE-AD1 and 2. Ruxolitinib cream was well tolerated in the long-term setting, with no serious treatment-related AEs [22].

TRuE-AD3, a phase 3 randomized trial to assess the efficacy and safety of topical ruxolitinib in children aged 2 to 12 years old, is currently underway (NCT04921969).

2.3. Tofacitinib

Topical tofacitinib, a JAK 1/3 inhibitor, was evaluated in a 4-week, phase 2a, randomized study (NCT02001181). Sixty-nine adults with mild-to-moderate AD were randomized 1:1 to 2% tofacitinib or vehicle ointment b.i.d.

Percentage change from baseline (CFB) in EASI score at week 4 was the primary endpoint. Secondary endpoints included percentage CFB in body surface area (BSA), CFB in EASI Clinical Signs Severity Sum Score, the proportion of patients with Physician's Global Assessment (PGA) response and CFB in patient-reported pruritus. Safety, local tolerability and pharmacokinetics were monitored.

The mean percentage change from baseline at week 4 in EASI score was −81.7% for tofacitinib and −29.9% for vehicle ($p < 0.001$). Similarly, all other efficacy endpoints were significantly improved with tofacitinib compared to vehicle ($p < 0.001$). Safety was comparable between regimens [23].

2.4. Brepocitinib

Brepocitinib, a JAK1/TYK2 inhibitor, was also evaluated in a phase 2 study (NCT03903822) with 240 adolescents and adults with mild-to-moderate AD. Patients were randomized to 6 weeks of treatment in one of eight study arms: once-daily topical brepocitinib at 0.1%, 0.3%, 1% or 3% concentration, twice-daily brepocitinib at 1% or 3% concentration or once or twice-daily vehicle cream.

The primary endpoint was a percentage change in EASI score from baseline to week 6. Brepocitinib 1% and 3% once daily and 1% twice daily achieved EASI score reductions of 70.1%, 67.9% and 75%, respectively, while the decrease was 44.4% and 47.6% among those in the once-daily and twice-daily vehicle control groups.

In the once-daily regimens, 29.7–44.4% of the patients achieved an IGA score of 0/1, compared to the 10.8% of the patients on once-daily and 13.9% on the twice-daily vehicle [24]. No phase 3 trial of brepocitinib in AD is currently active.

2.5. Other JAK Inhibitors

Another new agent is ATI-1777, a "soft" JAK1/JAK3 inhibitor. "Soft" JAK inhibitors are designed to provide JAK inhibition at the site of application and be rapidly metabolized in the systemic circulation. A phase 2 randomized study (NCT04598269) has been completed, but results have not been published yet. The developer has reported positive results in a press release [25], namely a 74.4% reduction in mEASI score from baseline at week 4 in the drug arm, compared to a 41.4% reduction in patients applying vehicle. No serious adverse events were reported

Ifidancitinib (ATI-502) is another JAK1/JAK3 inhibitor evaluated in a phase 2, open-label safety study (NCT03585296). Patients over 18 years old with moderate to severe AD and 2–20% BSA involvement were enrolled. They applied ATI-502 to affected areas b.i.d for 4 weeks. Seven out of twenty-two subjects reported 16 AEs, all unrelated to the drug. One person discontinued due to an unrelated bilateral lower extremity cellulitis, outside of the site of cream application.

Proportions of subjects with PGA of near clear with $\geq$2 grade improvement from baseline were 10.5%, 23.5% and 41.2% at weeks 1, 2 and 4. The percentage change from baseline in EASI was 18%, 35% and 40% at weeks 1, 2 and 4, respectively [26].

Studies for another two JAK inhibitors are currently active. NCT04435392 (phase 1/2) for jaktinib, a pan-JAK inhibitor, and NCT04717310 (phase 2/3) for ivarmacitinib (SHR 0302), a JAK1/STAT3 inhibitor. JAK inhibitors for the topical treatment of AD are summarized in Table 1.

Table 1. JAK inhibitors for the topical treatment of AD.

Name	Selectivity	Phase	Age/Severity	Regimens
Delgocitinib	pan-JAK	approved (Japan)	approved for children 2–15, adults >16, moderate-to-severe	0.25% children 0.5% adults, b.i.d
Ruxolitinib	JAK1, 2	approved (USA)	approved for >12, on trial for 2–12, mild-to-moderate	1.5% b.i.d
Tofacitinib	JAK1, 3	II	18–60, mild-to-moderate	2% b.i.d for 4 weeks
Brepocitinib	JAK1, TYK2	II	12–75, mild-to-moderate	0.1%, 0.3%, 1%, 3% q.d, 0.3%, 1% b.i.d for 6 weeks
ATI-1777	JAK1, 3	II	18–65, moderate-to-severe	2% b.i.d for 4 weeks
Ifidancitinib	JAK1, 3	II	>18, moderate-to-severe	0.46% b.i.d for 4 weeks
Jaktinib	pan-JAK	II	18–65, mild-to-moderate	0.5%, 1.5%, 2.5% b.i.d, 2.5% q.d
Ivarmacitinib	JAK1	III	>12, mild-to-moderate	0.5%, 1%, 2% b.i.d

3. Phosphodiesterase-4 Inhibitors

Over the last few years, PDE4 inhibitors have been identified as promising therapeutic agents for AD treatment [27,28]. The role of PDE4 activity in the circulating leukocytes in AD pathology is through the degradation of cyclic adenosine monophosphate (cAMP). The inhibition of PDE4 leads to increased levels of cAMP, which is involved in controlling the production of key inflammatory cytokines such as IL-4, IL-5, IL-10, IL-13 and prostaglandin E2 [29]. Moreover, the inhibition of PDE4 in monocytes in vitro enhances the cellular control of inflammation by downstream regulation of the nuclear factor-kB and nuclear factor of activated T-cell signaling pathways [30].

3.1. Crisaborole

Crisaborole 2% ointment, the first topical PDE4 inhibitor, was licensed by the FDA in December 2016 for the treatment of mild-to-moderate AD in children 2 years and older and in March 2020 for infants 3 months of age and older. Two 28-day, randomized, double-blind, vehicle-controlled trials (AD-301: NCT02118766, AD-302: NCT02118792) assessed the efficacy and safety for patients aged 2 years or older with mild-to-moderate AD. The primary endpoint was clear (0) or almost clear: (1) Investigator Static Global Assessment score (ISGA) with a greater than or equal to 2 grade improvement on day 29. Twice-daily applications led to a 32.8% (AD-301) and 31.4% (AD-302) reduction in ISGA compared to 25.4% and 18% for vehicle-treated subjects ($p = 0.038$, $p < 0.001$), respectively [31]. Moreover, in a phase 4 open-label study of 137 infants 3 to less than 24 months with mild-to-moderate AD, 30.2% of the patients achieved the primary endpoint of the ISGA with a safety profile equal to that in older children [32].

More common AEs related to crisaborole 2% ointment were local pain, burning, and stinging [31,32]. The mechanism that causes pain is still unknown. In a real-world retrospective study, more patients reported pain than in phase 3 clinical trials (31.7% versus 4.4%). Fifty percent of them applied crisaborole exclusively to the face ($p = 0.048$) [33]. An open-label 48-week safety extension study demonstrated that crisaborole had a favorable long-term safety profile without increasing the risk for treatment-related adverse events [34]. In the same study, 77.8% of patients did not require rescue therapy, defined as the need for the concomitant, nonconcurrent use of low-to mid-potency TCS or TCIs. Other important points regarding the treatment of AD with crisaborole include the rapid improvement in pruritus (as early as 24 h after the first application) and the normalization of epidermal pathology toward nonlesional skin [35].

3.2. Difamilast (OPA-15406)

PDE4 inhibitor OPA-15406 is a novel topical treatment for AD with high selectivity for phosphodiesterase-4-B. The effectiveness and tolerability were evaluated in a phase 2 study. Patients who met eligibility criteria were randomized into three groups to receive OPA-15406 0.3%, OPA-15406 1% or vehicle. OPA-15406 1% cream achieved the best results (IGA 0 or 1) at week 4 at a rate of 20.9% versus 14.6% for 0.3% cream and 2.7% for vehicle [36]. Recently, results from a phase 3 study have been published [37]. Patients aged 15 to 70 years received difamilast 1% ointment or vehicle twice daily for 4 weeks. The primary endpoint of IGA 0–1 with ≥2-grade improvement at week 4 was achieved by 38.46% of patients in the ointment group, compared to 12.64% in the vehicle. No serious adverse events were reported.

3.3. E6005 (RVT-501)

For the PDE4 inhibitor E6005, also known as RVT-501 or lotamilast, results are available from phase 1/2 trials in Japan. In early phase 1 trials in healthy adults and patients with AD, this compound was well tolerated with minimal systemic exposure [38]. In phase 2 dose-finding studies, 40 adult male patients randomized to receive E6005 ointment (0.01%, 0.03%, 0.1% or 0.2%). The targeted lesion severity scores significant decreased in the 0.2% group [39]. Subsequently, in a vehicle-controlled trial, 78 adult patients were randomized to receive either the 0.2% E6005 ointment or vehicle. The reduction in EASI and SCORAD scores from baseline after 12 weeks was significant in the E6005 group [40].

3.4. Other PDE-4 Inhibitors

In a randomized, vehicle-controlled, phase 2a trial, topical roflumilast (ARQ-151) 0.5% demonstrated no significant improvements in EASI score; however, there was a significant decrease in pruritus as measured by the NRS score (NCT01856764) [41,42]. Currently, roflumilast 0.15% cream is trialed (NCT04156191) for children and adults with AD. Furthermore, roflumilast 0.3% has shown very promising results in trials for psoriasis. Phase 2 studies have been completed for other topical PDE4 inhibitors, DRM02, Hemay808 and PF-07038124, but results have not been published to date. Phase 1 studies have been completed for LEO-39652 and LEO-32731, also known as orismilast, but results are not available yet. PDE-4 inhibitors for the topical treatment of AD are summarized in Table 2.

Table 2. PDE-4 inhibitors for the topical treatment of AD.

Name	Phase	Age/Severity	Regimens
Crisaborole	approved	>3 months, mild-to-moderate	2% b.i.d
Difamilast (OPA-15406)	III	15–70, mild-to-moderate	1% b.i.d for 4 weeks
Lotamilast (E6005, RVT-501)	II	20–64, all	0.2% b.i.d for 12 weeks
Roflumilast	II	18–65, moderate. 3 months—17 years currently on trial	0.5% b.i.d for 15 days, 0.05% and 0.15% for 4 weeks currently on trial
DRM02	II	18–70	0.25% b.i.d for 6 weeks
Hemay808	II	18–65, mild-to-moderate	1%, 3%, 7% for 29 days
PF-07038124	II	18–70, mild-to-moderate	0.01% q.d for 6 weeks
LEO-39652	I	>18, mild-to-moderate	3 weeks
Orismilast (LEO-32731)	I	>18, mild-to-moderate	3 weeks

4. Aryl Hydrocarbon Receptor Agonists

The aryl hydrocarbon receptor (AhR) is a member of the Pern-Arnt-Sim (PAS) superfamily of transcription factors. They detect and respond sense diverse endogenous and exogenous molecules and impact multiple biological activities [43]. Initially, AhR was

recognized as the mediator of the toxic effects of dioxins, but consequently, other ligands were identified. In response to activation by a ligand, AhR translocates from the cytoplasm to the nucleus, where it controls the transcription of a wide variety of target genes [44].

AhR is broadly expressed in the skin, and when activated, it upregulates the gene expression of filaggrin, loricrin and involucrin, accelerating epidermal terminal differentiation [45]. Consequently, AhR plays a vital role in developing and maintaining the skin barrier, epidermal homeostasis, pigmentation and the response to external signals, such as UVB, phytochemicals, environmental toxins or microbial products [46,47].

Although the mechanism of action was unknown, humans used AhR agonists empirically for thousands of years in numerous dermatologic conditions, the most classic example being coal tar [48]. Today, targeting the AhR system is an up-and-coming field for developing new drugs in dermatology and in many other specialties. Recently (23 May 2022), tapinarof was approved by the FDA for the treatment of plaque psoriasis in adults [49].

Tapinarof

Tapinarof is a naturally derived small molecule produced by bacterial symbionts (Photorhabdus luminescens) of entomopathogenic nematodes of the genus Heterorhabditis. It regulates skin-barrier protein expression, has antioxidant activity and suppresses IL-17 and -22 [47].

A randomized, multicenter, phase 2b, double-blind, vehicle-controlled study (NCT02564055) is the most important one that has been completed for AD. In this study, 247 adults and adolescent patients (12 to 65 years old, BSA between 5% and 35%, and IGA score $\geq$ 3) were randomized to receive tapinarof cream (0.5% or 1%) or a vehicle control, either once daily or twice daily for 12 weeks with a 4-week follow-up period.

At week 12, IGA response rates (IGA 0 or 1) were 53% (1% twice daily), 46% (1% once daily), 37% (0.5% twice daily) and 34% (0.5% once daily) versus 24% (vehicle twice daily) and 28% (vehicle once daily). This improvement was maintained for the 4-week follow-up period. Overall, among patients treated with tapinarof cream, 1% showed higher response rates than the 0.5% groups. EASI75 was also significantly higher in the tapinarof-treated groups at week 12: 60% (1% twice daily), 51% (1% once daily), 51% (0.5% twice daily) and 39% (0.5% once daily) versus 26% (vehicle twice daily) and 25% (vehicle once daily). The same was also true for EASI90 at week 12: 43% (1% twice daily), 27% (1% once daily), 28% (0.5% twice daily), and 22% (0.5% once daily) versus 14% (vehicle twice daily) and 5% (vehicle once daily). BSA, pruritus NRS, subject impressions of AD and Patient-Oriented Eczema Measure (POEM) scores were also significantly improved in tapinarof groups [50].

Treatment-emergent AEs (TEAEs) were reported in 51% of patients, and the majority were mild to moderate in intensity. The most frequently reported TEAE was nasopharyngitis. The other TEAEs reported in at least 5% of patients in any arm or in total were folliculitis, worsening or flare of AD, upper respiratory tract infection, headache, acne and impetigo. Overall, 32 patients (13%) had TEAEs that were considered treatment-related.

More patients in the vehicle groups (6 of 82 [7%]) than in the groups treated with tapinarof (7 of 165 [4%]) discontinued the study because of TEAEs. Worsening or flare of AD was the most frequent TEAE that led to a discontinuation of the study treatment. ECG findings were observed in 21% of patients in the groups treated with tapinarof and 17% in the vehicle groups. They were not considered significant, resolved over time and never led to treatment discontinuation. Elevations in liver enzyme levels (at least twice the upper limit of the normal range) were observed in six patients treated with tapinarof. They all resolved during treatment, and none of those patients discontinued treatment [51].

Three large (estimated enrollment: 961, 400 and 400 patients) phase 3 studies (NCT05142774, NCT05014568 and NCT05032859) are currently active.

It is worth mentioning here a common misconception about tapinarof synonyms. At the beginning of its development, the drug was known as GSK-2894512, WBI-1001 and DMVT-505. A common error by many authors is that benvitimod is considered the same as tapinarof. Indeed, the active ingredient is the same (3,5-dihydroxy-4- isopropyl-trans-

stilbene) and is isolated from the bacterium Photorhabdus luminescens [52]. However, tapinarof cream 1% comprises a novel vehicle with specific excipients to enhance efficacy, drug delivery and patient acceptability. Benvitimod 1%, uses a petrolatum-based vehicle for delivery and requires twice-daily dosing. Clinical trials for the two formulations are also separate with essential differences. Benvitimod is currently trialed in China for AD (NCT05326672) and is approved for psoriasis [53].

5. Transient Receptor Potential Vanilloid 1 Antagonists

The transient receptor potential channels (TRPs) are a superfamily of ion channels consisting of six members (TRPC, TRPV, TRPP, TRPM, TRPA and TRPML). TRP proteins are remarkable channels because of the diversity of their activation mechanisms, cation selectivity and biological function [54].

Member 1 of the TRPV family (TRPV1) is a nonselective cation channel with high permeability to calcium. It is activated by heat, low pH, capsaicin and endogenous inflammatory mediators [55]. It is expressed, among others, in keratinocytes, mast cells and cutaneous sensory nerves, indicating its important role in cutaneous physiology and disease [56]. TRPV1 has shown to play a role in pruritus, epidermal barrier function and inflammation [57]. In AD lesions, TRPV1 is overexpressed and its activation results in the production of molecules that promote itch and inflammation [58].

Asivatrep

Since preclinical experiments in rats and mice, asivatrep (then known as PAC-14028) demonstrated promising results, positively affecting many aspects of AD. Its use suppressed serum IgE, mast cell degranulation, the expression of IL-4 and IL-13, itch and inflammatory cell infiltration. Skin-barrier recovery was accelerated, and possible carcinogenicity was ruled out [55,59–61].

Later, a phase 2b study (NCT02757729) was conducted on 194 adults (19–70 years old) with mild to moderate AD. Patients were randomized in asivatrep 0.1%, 0.3%, 1.0% or vehicle, twice daily for 8 weeks. The primary efficacy variable was the IGA success rate (IGA score of 0 or 1 with at least a two-grade improvement from baseline) at week 8. Secondary efficacy variables included SCORAD and EASI scores, pruritus VAS and sleep disturbance scores.

The IGA success rates were 14.6% for vehicle cream, 42.6% for 0.1% cream, 38.3% for 0.3% cream and 57.5% for 1.0% cream. All secondary variables were also improved, although the improvement was not statistically significant. The small sample and adult age of patients may have been important factors affecting statistical insignificance. The safety profile and the overall incidence of AEs were similar in the drug and vehicle groups [62].

Recently, results of a phase 3 study were published [57]. In it (CAPTAIN-AD and NCT02965118), 240 patients 12 to 70 years old were randomized 2:1 to 1.0% asivatrep or vehicle cream for 8 weeks.

At week 8, the proportion of patients with an IGA score of 0 or 1 was 36.0% in the asivatrep group and 12.8% in the vehicle group ($p < 0.001$). Improvements of at least 2 points on the IGA from baseline score was 20.3% versus 7.7% ($p = 0.01$). At week 8, the reduction in EASI score was 44.3% versus 21.4%, respectively ($p < 0.001$). Additionally, significantly more patients in the asivatrep group experienced an improvement of at least 50% (EASI-50), 75% (EASI-75) and 90% (EASI-90) (50.3%, 23.5% and 9.8% versus 28.2%, 11.5% and 2.6%). Pruritus and sleep disturbance were significantly reduced in the drug group compared to the vehicle ($p = 0.02$).

Asivatrep cream was well-tolerated and was not associated with clinically significant application site reactions. Overall, the incidence of TEAEs was reported in 14.7% of patients treated with asivatrep and 6.3% treated with vehicle cream. The most common TEAEs were nasopharyngitis (2.6%), urticaria (1.3%), burning sensation (1.3%) and rhinorrhea (1.3%), which were similar in the vehicle group. No patient discontinued treatment due to adverse events, and serious adverse events were not reported.

Overall, in clinical trials, asivatrep cream resulted in evident and enduring positive results in treating AD, along with an acceptable safety profile.

6. Skin Microbiome Modulators

In the 1970s, it was shown that Staphylococcus aureus (S.aureus) is overrepresented on the skin of AD patients [63]. Later, cutaneous dysbiosis was identified as a contributing factor to AD pathogenesis [64]. Cutaneous dysbiosis is characterized by an increased colonization of S.aureus and reduced colonization by the abundant bacterial genera of healthy skin.

More recently, patients colonized with S.aureus have been described as a unique AD phenotype. Patients in this category have more severe disease, reduced barrier function, increased allergen sensitization and elevated IgE, eosinophils, lactate dehydrogenase and various Th2 biomarkers such as TARC, periostin and CCL26 [65,66].

S.aureus toxins induce mast cell degranulation, promoting innate and adaptive immune responses, and induce IL-1b production from monocytes. In the dermis, through the defective skin barrier, S.aureus interacts with immune cells and triggers cytokine production including IL-4, IL-13, IL-22 and TSLP [67].

Those observations led to various strategies to try and modulate the skin microbiome of AD patients, either by decreasing S.aureus or increasing normal microbiota [68].

In healthy individuals, Roseomonas mucosa (R.mucosa) is the most representative Gram-negative bacteria [69]. An open-label trial (NCT03018275) with topical application of R.mucosa twice-weekly for 6 weeks in both adults and children (9–14 years old) with AD found that the commensal bacterium was associated with improvements in SCORAD and pruritus and a reduction in TCS use, with no significant AEs [70]. In the same trial, it was noted that non-responders had a family history of AD persisting into adulthood for at least 3 generations, suggesting that heritable factors may influence responses to R. mucosa therapy.

Another topical formulation, FB-401 with three strains of R.mucosa, showed promising results in a phase 1/2 trial. Sixty percent of adult patients showed a 50% reduction in SCORAD, while 90% of the pediatric patients achieved EASI50 and 30% achieved EASI90 [68]. Unfortunately, in a later, more extensive trial, FB-401 failed to meet the primary goal of EASI50 (58% in the FB-401 arm versus 60% in the placebo arm), and development will not continue [71].

Staphylococcus hominis A9 (ShA9) is another healthy human skin microbiome bacterium that has been trialed as a topical therapy for AD. ShA9 killed S.aureus on the skin of mice and inhibited the expression of the toxin psmα that promotes inflammation. Then, in a phase 1, randomized, 1-week trial (NCT03151148), topical ShA9 or vehicle was applied on the forearm skin of 54 adults with S. aureus-positive AD. The primary endpoint of safety was met, and a small improvement of AD lesions was also induced, rendering ShA9 a safe and potentially beneficial future treatment [72].

Nitrosomonas eutropha is an ammonia-oxidizing Gram-negative bacterium able to produce nitric oxide, which is an important mediator with beneficial metabolic and potential anti-inflammatory properties [67]. Results of three phase 1 and 2 trials (NCT04490109, NCT03775434 and NCT03235024) are not yet available, although pruritus and AD appearance were significantly improved in adults and children, according to a press release [73].

One of the reasons AD patients have a predisposition for cutaneous and systemic infections is the decreased antimicrobial peptides production [74]. Omiganan is a synthetic indolicidin analogue. Indolicidin is an antimicrobial peptide isolated from the neutrophils of cows. It is active against Gram-positive and -negative bacteria but has also been shown to kill fungi and even HIV [75]. Cationic peptides, such as omiganan, are also suggested to have immunomodulatory roles in both pro- and anti-inflammatory pathways.

Because of those properties, omiganan gel is investigated as a possible treatment for various infectious and inflammatory disorders, among them some cutaneous ones,

including acne, rosacea, condylomata acuminata and vulvar intraepithelial neoplasia (VIN) [74].

For AD, a phase II trial randomized 36 patients with mild to moderate disease 1:1:1 to omiganan gel 1%, 2.5% and vehicle, once daily for 4 weeks. Small but significant results in BSA, SCORAD and pruritus were observed only in the 2.5% arm. Skin microbiota shifted from lesional to non-lesional [76]. In a later trial by the same group, omiganan gel 2.5% twice daily led to a recovery of dysbiosis but without clinical improvement [77]. The authors concluded that dysbiosis does not seem a viable monotherapy drug target in mild-to-moderate AD.

Omiganan development in the future may focus on diseases where S.aureus plays a more central role, e.g., in superinfected AD, reducing the need for oral antibiotics, or eradicating multi-drug resistant S. aureus strains in long-term carriers.

The niclosamide ATx201 can also achieve the decolonization of S. aureus. In a phase 2 trial (NCT03304470), 31 patients with mild-to-severe AD received ATx201 cream 2% and a matching vehicle once daily for 3 weeks. Treatment was generally safe and the histological and transcriptional profiling analysis on day 22 demonstrated that treatment significantly increased the expression of biomarkers related to the skin-barrier function and decreased expression levels of markers related to inflammation [78].

Primary and major secondary endpoints of the most important trials for the above-mentioned therapeutic targets are summarized in Table 3.

Table 3. Primary and major secondary endpoints of the most important trials for the drugs mentioned in Sections 2–6.

Drug	Primary End-Point	Other End-Points
Delgocitinib	JapicCTI-173554: Mean percent change in mEASI at week 4: −44.3% in the drug group vs. 1.7% for vehicle. ($p < 0.001$). JapicCTI-173555: Safety: AEs in 69% of patients. 15.4% considered treatment-related. 1.4% considered serious (Kaposi's varicelliform eruption)	JapicCTI-173554: mEASI-50 at week 4: 51.9% for drug vs. 11.5% for vehicle ($p < 0.001$). mEASI-75 at week 4: 26.4% vs. 5.8% respectively ($p < 0.01$). IGA response rates at week 4: $p = 0.32$ for overall score, $p < 0.05$ for face/neck score. NRS: lower in drug group. All results maintained at week 24. JapicCTI-173555: mEASI-50 at week 4, 24, 52: 31.5%, 42.3% and 51.9%. mEASI-75 at week 4, 24, 52: 10.9%, 22.7% and 27.5% IGA and NRS: improved at weeks 4, 24 and 52
Ruxolitinib	IGA 0–1 at week 8: 53.8%(TRuE-AD1) and 51.3% (TRuE-AD2) in the 1.5% cream groups vs. 15.1% and 7.6% for vehicle ($p < 0.0001$)	EASI-75 at week 8: 62.1% and 61.8% in the 1.5% cream groups vs. 24.6% and 14.4% for vehicle ($p < 0.0001$). EASI90 at week 8: ($p < 0.0001$) vs. vehicle. Reduction in NRS: ($p < 0.05$) vs. vehicle
Tofacitinib	EASI score change at week 4: 81.7% vs. 29.9% for vehicle.	EASI 50, 75 and 90: Significantly higher for drug vs. vehicle ($p < 0.05$) at weeks 2 and 4. Change in BSA: −76% for drug vs. −31% for vehicle, significantly greater ($p < 0.001$) at week 4. ISI scores: significantly greater for drug vs. vehicle at weeks 2 and 4 ($p < 0.001$).
Brepocitinib	EASI score change at week 6: 70.1%, 67.9%, and 75%, for the 1%, 3% q.d and 1% b.i.d groups respectively. 44.4% and 47.6% in the q.d and b.i.d vehicle groups.	IGA score of 0/1 at week 6: 27.8–44.4% of patients on q.d drug vs. 10.8% for q.d vehicle. EASI 90 at week 6: 27.8–41.7% of patients on 0.3%, 1%, and 3% q.d cream, vs. 10.8% for q.d vehicle, 27% of patients on 1% b.i.d cream, vs. 8.3% b.i.d vehicle. Improvement of at least 4 points on the PP-NRS at week 6: 45.2% of patients on 1% cream q.d, 50% on 3% q.d, and 40.7% on 1% b.i.d, vs. 17% for vehicle.
ATI-1777	Reduction in mEASI score at week 4: 74.4% in the drug arm, vs. 41.4% for vehicle	not yet available
Ifidancitinib	PGA of near clear with ≥2 grade improvement: 10.5%, 23.5%, 41.2% of patients at weeks 1, 2, and 4.	Change in EASI: 18%, 35%, 40% at weeks 1, 2, and 4. Percent change in SPA: 35%, 46% and 31% at weeks 1, 2, and 4.
Jaktinib	PGA 0/1 or a decrease of ≥2, 7 days after the last dose: not yet available	PGA 0/1 at 8 and 16 weeks: not yet available
Ivarmacitinib	Change in EASI at Week 8: not yet available	not yet available
Crisaborole	ISGA score 0/1 with ≥2 grade improvement at day 29: 32.8% (AD-301) and 31.4% (AD-302) reduction vs. 25.4% ($p = 0.038$) and 18% ($p < 0.001$) for vehicle.	ISGA score 0/1 at day 29: 51.7% vs. 40.6% ($p < 0.005$) and 48.5% vs. 29.7% ($p < 0.001$) respectively. Time to ISGA success: 14.7% for drug vs. 5.4% for vehicle at day 8. Median time to improvement in pruritus: 4 days for drug vs. 9 days for vehicle. Mean change in DLQI at day 29: −5.2 for drug vs. −3.5 for vehicle.

Table 3. *Cont.*

Drug	Primary End-Point	Other End-Points
Difamilast (OPA-15406)	IGA 0–1 with ≥2 grade improvement at week 4: 38.46% of patients in the ointment group vs. 12.64% for vehicle ($p < 0.0001$)	EASI 50, 75, 90 at week 4: 58.24%, 42.86% and 24.73 of patients in drug group vs. 25.82%, 13.19% and 5.49% for vehicle. Mean percent change in EASI score at week 1: −32.6% vs. −10.4% for drug and vehicle respectively ($p < 0.0001$). POEM, affected BSA, pruritus VRS, Skindex-16: all significantly improved vs. vehicle ($p < 0.0001$) at week 4
Lotamilast (E6005, RVT-501)	Long-term safety and tolerance: Neither death nor serious TEAEs were encountered in the entire study period. In the randomization phase, the incidence of TEAEs was 50.0% in the drug group vs. 38.5% for vehicle group. The incidence of TEAEs leading to study withdrawal was 9.6% in the drug group and 15.4% for vehicle group.	Scores reduction at week 12: significantly reduced: EASI, $p = 0.030$; SCORAD-objective, $p < 0.001$; SCORAD-C, $p = 0.038$) Not significantly reduced: Itch Behavioral Rating Scale, ($p = 0.462$)
Roflumilast	Change in Modified Local SCORAD at day 15: Not significant reduction vs. vehicle ($p = 0.276$)	Change in PAP at day 15: Significantly reduced ($p < 0.013$)
DRM02	not yet available	not yet available
Hemay808	not yet available	not yet available
PF-07038124	not yet available	not yet available
LEO-39652	not yet available	not yet available
Orismilast (LEO-32731)	not yet available	not yet available
Tapinarof	IGA response rates at week 12: 53% (1% b.i.d; $p = 0.008$), 46% (1% q.d; $p = 0.084$), 37% (0.5% b.i.d; $p = 0.240$), and 34% (0.5% q.d; $p = 0.535$) vs. 24% (vehicle b.i.d) and 28% (vehicle q.d).	EASI75 at week 12: significantly higher in the tapinarof groups, except the 0.5% q.d, vs. vehicle groups. EASI90 at week 12: significantly higher in the tapinarof groups, except the 0.5% b.i.d, vs. vehicle groups. Mean percent change in EASI at week 12: significantly higher in all tapinarof groups vs. vehicle groups. Mean percent change in BSA at week 12: significantly greater in the tapinarof groups, except the 0.5% b.i.d, vs. vehicle groups.
Asivatrep	IGA score of 0 or 1 at week 8: 36.0% in the drug group vs. 12.8% for vehicle.	Improvement ≥2 points on IGA score at week 8: 20.3% for drug vs. 7.7% for vehicle. EASI reduction at week 8: 44.3% vs. 21.4% respectively. EASI-50, 75, and 90 at week 8: 50.3%, 23.5%, and 9.8% of patients on drug vs. 28.2%, 11.5%, and 2.6% on vehicle. Statistical significance achieved in all secondary end-points, as also in pruritus and sleep disturbance reduction.
R.mucosa	50% improvement in SCORAD: 66.7% of patients	75% improvement in SCORAD: 40% of patients. Subjective pruritus: significantly decreased.
FB-401	EASI50: 58% in drug arm vs. 60% in placebo arm	-
ShA9	Safety through day 8 compared to vehicle: Significantly fewer AEs in participants treated with ShA9 ($p = 0.044$)	EASI and SCORAD: no significant difference Decrease in S. aureus and increased ShA9 DNA: endpoints met
Nitrosomonas eutropha	Not yet available, positive results in pruritus and AD appearance reported in press release	-
Omiganan gel	*S.aureus* reduction at day 28: Statistically significant in the omiganan 1% ($p = 0.03$) and 2.5% ($p = 0.01$) vs. vehicle.	Clinical improvement evaluated by EASI, SCORAD, IGA, POEM, DLQI and NRS: no improvement
ATx201	Safety: safe and well tolerated	Expression of biomarkers related to skin-barrier function: Significantly increased ($p < 0.05$). Histological responders: 51.7% of those receiving 2% cream vs. 31.0% for vehicle.

ISI: Itch Severity Item, PP-NRS: Peak Pruritus Numerical Rating Scale, SPA: Subject's Pruritus Assessment, DLQI: Dermatology Life Quality Index, VRS: verbal rating scale, PAP: Participants' Assessment of Pruritus.

7. Newer Emolients

Emollients and moisturizers are the cornerstone of basic disease management of AD [79,80]. Fourteen independent publications from Europe, North America, Asia, the Asia-Pacific region and Australia, including AD management guidelines between 2007 and 2022, were reviewed and displayed that daily moisturization is an integral part of recommendations [81,82].

These products contain vehicle-type substances such as humectants (urea or glycerol) and occludents (petrolatum) and act as an occlusive layer on the skin, promoting stratum corneum hydration and reducing transepidermal water loss [79,83,84]. Data from Cochrane review with emollient trials display a favorable impact on AD management with no superiority among them [85].

In recent years, there has been a rapid increase in non-medicated emollients containing active ingredients termed "emollient plus" or AD therapeutic moisturizers that improve

skin barrier with antipruritic, anti-inflammatory and antioxidant effects. Active ingredients are, for example, ceramides, saponins, colloidal oatmeal and nonpathogenic bacterial lysates from Aquaphilus dolomiae or Vitreoscilla filiformis with possible molecular targets as it emerges from in vitro and clinical research data [86–88]. A prospective, double-blind, placebo-controlled clinical study demonstrated that patients with mild AD who received cream with 5% Vitreoscilla filiformis decreased SCORAD levels and pruritus significantly compared with those who received placebo [89]. These lysates might influence the skin microbiome due to the reduction in Staphylococcus aureus colonization and display immunomodulatory effects locally on the skin. Thus, they could be a therapeutic approach targeting prevention of relapses and stabilization of AD skin [90].

8. Other

Many other molecules are under investigation for the topical treatment of AD. Those with trials that have not started yet are ongoing, or those with results that are not yet available are presented in Table 4.

Table 4. Agents for the topical treatment of AD for which trials have not started yet, are ongoing or results are not available in ClinicalTrials.gov or clinicaltrialsregister.eu.

Agent	Mechanicm of Action	NCT ID
ALX-101 Gel 1.5% (Rovazolac)	LXR agonists	NCT03175354
AM1030-CREAM	5-HT2BR antagonist	NCT02379910
AMTX-100 CF	Nuclear transport modifier (NTM)	NCT04313400
ASN008 (*1)	Targets small afferent sodium channels/Antipruritic	NCT03798561
Atuzabrutinib (SAR 444727 or PRN 473)	BTK inhibitor	NCT04992546
Aurstat Hydrogel	Emolient/Antipruritic	NCT01905631
BEN2293	TRK inhibitor	NCT04737304
BioLexa	Antibacterial	NCT04544943
BMX-010	Antioxidant	NCT03381625
BPR 277	Kallikrein-related peptidase	NCT01428297
BX005-A (*2)	Phage cocktail targeting S.aureus	NCT05240300
CD 5024/Ivermectin (Soolantra)	Chloride channel agonists	NCT03250624
CYCLATOP(Cyclosporine 5% solution) (*3)	Calcineurin inhibitor	NCT02865356
DBI-001 (*4)	Antibacterial	NCT05253755
DMT210 Topical Gel	G protein-coupled receptor agonist	NCT02949960
DS 107/DGLA (*5)	Bioactive lipid (dihomo-γ-linolenic acid) inhibiting the expression of CD40	NCT02925793 NCT03676036 NCT03676933
Ectoin Dermatitis Cream 7% (EHK02)	Emolient	NCT04097327
FMX114 (tofacitinib and fingolimod) (*6)	Jak inhibitor and sphingosine-1-phosphate receptor modulator	NCT04927572
GM-XANTHO	Botanical drug balm	NCT04369846
HAT01	Botanical complex	NCT03089229
HL-009 Liposomal Gel(Cobamamide)	Vitamin B12 analogues, Nitric oxide inhibitor.	NCT01568489
HY209 Gel/Taurodeoxycholic acid	G Protein Coupled Receptor 19(GPCR19) agonist	NCT04530643
IDP-124	Undefined mechanism	NCT03058783 NCT03002571
Isopentenyltheophylline 0.44% + Glycerin 4.56%	Undefined mechanism	NCT05057351
Jaungo (Shiunko in Chinese) (*7)	Herbal ointment	NCT02900131
Lactibiane Topic AD	Emolient/Cosmetic product	NCT04728269
Lactobacillus reuteri (ADreuteri)	Probiotic	NCT04265716

Table 4. *Cont.*

Agent	Mechanicm of Action	NCT ID
Levagen+/ Palmitoylethanolamide (PEA)	Endocannabinoid-like lipid mediator	NCT05003453
Menthoxypropanediol	Anti-TRPM8/Antipruritic	NCT03610386
MH004	Unknown	NCT04815148
NLAC (Natural Lactic Acid-enriched Cream)	Emolient	NCT05092464
PR022 (Hypochlorous acid)	Antiseptic	NCT03351777
Q301(Zileuton) (*8)	leukotriene inhibitor	NCT03571620 NCT02426359
RelizemaTM cream	Antioxidant/ Antipruritus	NCT05259774
SB414 (Berdazimer sodium) (*9)	Nitric oxide donors	NCT03431610
SB011	GATA3 transcription factor inhibitor	NCT02079688
SNG100	Unknown	NCT04615962
TER-101	Unknown	NCT04753034
Topialyse Baume Barrière (TOPIA)	Emolient	NCT05006300
ZEP-3Na	synthetic analogue of a compound of rattle snake venom	NCT04307862
ZK245186 (Mapracorat)	Selective glucocorticoid receptor agonists (SEGRAs).	NCT01228513 NCT00944632 NCT01359787
0.5% Cannabidiol and 1% Hemp Oil (Celosia)	Emolient	NCT04045314
2.5% and 5% Cis-urocanic Acid	Emolient	NCT01320579

LXR: liver X receptor, 5-HT2BR: serotonin receptor 2B, TRPM8: Transient receptor potential cation channel subfamily M(melastatin) member 8, HAT01: herbal anti-inflammatory treatment; S.aureus: Staphylococcus aureus; TRK: tropomyosin receptor kinases; BTK: Bruton's tyrosine kinase. Results from various sources (press releases, conference posters or the literature) are available online for agents marked with * in Table 4, as follows (All accessed on 17 June 2022): *1 https://www.asanabiosciences.com/_files/ugd/d170b0_f8d4c69d2e374ce99f41e2d734cb78dc.pdf; *2 https://www.biomx.com/wp-content/uploads/2022/01/a11y-RAD-2021-Poster_June-2021F.pdf; *3 https://www.mdedge.com/pediatrics/article/175673/atopic-dermatitis/topical-cyclosporine-safely-tamed-atopic-dermatitis-4; *4 https://www.dermbiont.com/in-the-news/2021/1/8/dermbiont-announces-positive-results-in-phase-2a-clinical-trial-in-atopic-dermatitis-with-a-topical-live-biotherapeutic; *5 https://www.dsbiopharma.com/2018/10/03/ds-biopharma-announces-positive-top-line-phase-2b-trial-results-for-ds107-as-a-topical-treatment-for-mild-to-moderate-atopic-dermatitis/; *6 https://vynetherapeutics.com/pipeline-overview/fmx114/; *7 https://pubmed.ncbi.nlm.nih.gov/30219454/; *8 http://www.qurient.com/bbs/content.php?co_id=q301; *9 https://novan.com/novan-to-present-data-from-sb414-phase-1b-atopic-dermatitis-clinical-trial-at-3rd-inflammatory-skin-disease-summit/.

9. Discussion

AD is the most common inflammatory skin disease with a considerable impact on the lives of patients and their families. It imposes a substantial physical, psychological and social burden. Pruritus and the accompanying sleep disturbance are distressing and increase the risk for psychiatric conditions such as ADHD, depression, suicidal ideation and autism. Multiple other complications and comorbidities have been reported, including, but not limited to, growth delay, bacterial and viral infections, ocular abnormalities, aortic stiffness and other allergic, metabolic and autoimmune conditions [1,2]

Nevertheless, for decades and until recently, the only options for the topical treatment of the disease were TCS and TCIs in combination with emollients. Although effective, significant concerns about their long-term usage bother clinicians and patients. TCS may cause skin atrophy, telangiectasia and numerous other adverse effects. Systemic absorption is also a concern, especially in younger children with a greater surface-to-weight ratio than adults. TCIs are less effective than TCS in controlling exacerbations of the disease. Additionally, the FDA black box about the theoretical increased risk of malignancy distressed many parents of children with AD. The multifaceted and, to an extent, still unknown pathophysiology; inconsistent clinical manifestations depending on age, body sites affected and other factors; and the chronic course with relapses and remissions added to the difficulty of managing the disease.

This unpleasant situation is rapidly changing for the better. The pathophysiologies of AD, inflammatory pathways and physiology, in general, are better understood due to advances in basic science research. At the same time, research fields regarding drug development are also advancing swiftly. AD or dermatology, as well as medicine in general, is experiencing a revolution in therapeutics, which began with biologics. Since then, progress seems to have accelerated, and the future is eagerly awaited. New drug categories that we mentioned in this review and many others in other specialties are or will be available in the times ahead.

Subsequently, other important steps are advancements in safety, more effective production and, therefore, lower cost, discovering biomarkers and an improved understanding of the various phenotypes and/or behaviors of the disease in different patients. Ultimately, clinicians will not only have many treatment options but also the knowledge to use them in a non-blind manner by targeting specific aspects of the disease in each patient. The result will be improved, including the individualized treatment of AD and other chronic, complex diseases.

Finally, concerning AD, another urgent necessity in the coming years is the comparison of efficacy and safety between all new and upcoming treatments. Although TCS and TCIs were the only available topical treatments for decades, they are indeed effective and inexpensive. The adverse events mentioned above are not frequent if properly used. Therefore, it will be difficult for many of the upcoming treatments to outplace the old ones until their efficacy, safety, tolerability, adherence and cost-effectiveness are proven by comparison studies [91]. Until now, in most studies of new treatments, the comparator is usually the vehicle. Since the industry can generally obtain approval for new drugs without head-to-head studies, real-life data comparing new and old topical treatments will be valuable in the following years. Until then, a level of confusion and uninformed choices by clinicians and patients is expected. Network meta-analyses can, to a certain degree, fill this knowledge gap. For example, a recent meta-analysis from China comparing JAK and PDE-4 inhibitors concluded that tofacitinib 2% b.i.d, ruxolitinib 1.5% b.i.d and delgocitinib 3% b.i.d showed superior efficacy over other JAK and PDE4 inhibitors [92]. Future real-life data will also specify another important aspect of the new treatments, namely if any of them is better at targeting specific phenotypes of the disease. In most clinical trials and those presented here, patients are recruited just by having the required conditions for the trial severity of the disease. More information, such as IgE levels, filaggrin mutation status, other atopic comorbidities, etc., are usually unmentioned. Regardless, progress in the topical treatment of AD is of major importance since even the safest systemic agent has significantly more safety concerns than topical agents, especially in children.

In summary, in this review, we aimed to make a substantial amount of information about an exciting and fast-growing field of dermatology easily accessible to anyone interested, including emerging topical therapies for AD. We focused on the most important information, major therapeutic targets and principal aspects of trial results, namely safety and efficacy. It is evident that the landscape will change dramatically soon enough, albeit many important questions remain to be answered. The upcoming approval of the presented topical therapies (and many more systemic ones) will result in a wide range of available treatment options and will enable personalized decision making depending on patient characteristics, making clinicians' and patients' lives easier.

It is still impossible to predict the role that each one of all the presented agents will have in the treatment of AD in future. It seems unlikely that only one agent will replace TCS in being the gold standard for the majority of patients. Responses in trials are varying, and the heterogeneity of the disease is now better understood. This knowledge and all other aspects of the disease, such as age, age of onset, comorbidities, etc., together with new discoveries in the stratification of AD cases (biomarkers and artificial intelligence) will lead to the best possible treatment for each patient.

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References

1. Weidinger, S.; Beck, L.A.; Bieber, T.; Kabashima, K.; Irvine, A.D. Atopic dermatitis. *Nat. Rev. Dis. Prim.* **2018**, *4*, 18003. [CrossRef] [PubMed]
2. Cascella, R.; Strafella, C.; Germani, C.; Manzo, L.; Marsella, L.T.; Borgiani, P.; Sobhy, N.; Abdelmaksood, R.; Gerou, S.; Ioannides, D.; et al. FLG (filaggrin) null mutations and sunlight exposure: Evidence of a correlation. *J. Am. Acad. Dermatol.* **2015**, *73*, 528–529. [CrossRef] [PubMed]
3. Sideris, N.; Vakirlis, E.; Tsentemeidou, A.; Kourouklidou, A.; Ioannides, D.; Sotiriou, E. Under Development JAK Inhibitors for Dermatologic Diseases. *Mediterr. J. Rheumatol.* **2020**, *31*, 137–144. [CrossRef] [PubMed]
4. Kusari, A.; Han, A.M.; Schairer, D.; Eichenfield, L.F. Atopic Dermatitis. *Dermatol. Clin.* **2019**, *37*, 11–20. [CrossRef]
5. Otsuka, A.; Nomura, T.; Rerknimitr, P.; Seidel, J.; Honda, T.; Kabashima, K. The interplay between genetic and environmental factors in the pathogenesis of atopic dermatitis. *Immunol. Rev.* **2017**, *278*, 246–262. [CrossRef]
6. Nygaard, U.; Deleuran, M.; Vestergaard, C. Emerging Treatment Options in Atopic Dermatitis: Topical Therapies. *Dermatology* **2017**, *233*, 333–343. [CrossRef]
7. Aaronson, D.S.; Horvath, C.M. A Road Map for Those Who Don't Know JAK-STAT. *Science* **2002**, *296*, 1653–1655. [CrossRef]
8. Aittomäki, S.; Pesu, M. Therapeutic Targeting of the JAK/STAT Pathway. *Basic Clin. Pharmacol. Toxicol.* **2014**, *114*, 18–23. [CrossRef]
9. Darnell, J.E., Jr.; Kerr, I.M.; Stark, G.R. Jak-STAT pathways and transcriptional activation in response to IFNs and other extracellular signaling proteins. *Science* **1994**, *264*, 1415–1421. [CrossRef]
10. Virtanen, A.T.; Haikarainen, T.; Raivola, J.; Silvennoinen, O. Selective JAKinibs: Prospects in Inflammatory and Autoimmune Diseases. *BioDrugs* **2019**, *33*, 15–32. [CrossRef]
11. Lin, C.M.; Cooles, F.A.; Isaacs, J.D. Basic Mechanisms of JAK Inhibition. *Mediterr. J. Rheumatol.* **2020**, *31*, 100–104. [CrossRef] [PubMed]
12. Walker, J.G.; Ahern, M.J.; Coleman, M.; Weedon, H.; Papangelis, V.; Beroukas, D.; Roberts-Thomson, P.J.; Smith, M.D. Characterisation of a dendritic cell subset in synovial tissue which strongly expresses Jak/STAT transcription factors from patients with rheumatoid arthritis. *Ann. Rheum. Dis.* **2007**, *66*, 992–999. [CrossRef] [PubMed]
13. Baxter, E.J.; Scott, L.M.; Campbell, P.J.; East, C.; Fourouclas, N.; Swanton, S.; Vassiliou, G.S.; Bench, A.J.; Boyd, E.M.; Curtin, N.; et al. Acquired mutation of the tyrosine kinase JAK2 in human myeloproliferative disorders. *Lancet* **2005**, *365*, 1054–1061. [CrossRef]
14. Bao, L.; Zhang, H.; Chan, L.S. The involvement of the JAK-STAT signaling pathway in chronic inflammatory skin disease atopic dermatitis. *JAK-STAT* **2013**, *2*, e24137. [CrossRef] [PubMed]
15. Amano, W.; Nakajima, S.; Kunugi, H.; Numata, Y.; Kitoh, A.; Egawa, G.; Dainichi, T.; Honda, T.; Otsuka, A.; Kimoto, Y.; et al. The Janus kinase inhibitor JTE-052 improves skin barrier function through suppressing signal transducer and activator of transcription 3 signaling. *J. Allergy Clin. Immunol.* **2015**, *136*, 667–677.e7. [CrossRef]
16. Nakagawa, H.; Nemoto, O.; Igarashi, A.; Saeki, H.; Kaino, H.; Nagata, T. Delgocitinib ointment, a topical Janus kinase inhibitor, in adult patients with moderate to severe atopic dermatitis: A phase 3, randomized, double-blind, vehicle-controlled study and an open-label, long-term extension study. *J. Am. Acad. Dermatol.* **2020**, *82*, 823–831. [CrossRef]
17. Nakagawa, H.; Nemoto, O.; Igarashi, A.; Saeki, H.; Murata, R.; Kaino, H.; Nagata, T. Long-term safety and efficacy of delgocitinib ointment, a topical Janus kinase inhibitor, in adult patients with atopic dermatitis. *J. Dermatol.* **2020**, *47*, 114–120. [CrossRef]
18. Nakagawa, H.; Nemoto, O.; Igarashi, A.; Saeki, H.; Kabashima, K.; Oda, M.; Nagata, T. Delgocitinib ointment in pediatric patients with atopic dermatitis: A phase 3, randomized, double-blind, vehicle-controlled study and a subsequent open-label, long-term study. *J. Am. Acad. Dermatol.* **2021**, *85*, 854–862. [CrossRef]
19. Suehiro, M.; Numata, T.; Murakami, E.; Takahashi, M.; Saito, R.; Morioke, S.; Kamegashira, A.; Takahagi, S.; Hide, M.; Tanaka, A. Real-world efficacy of proactive maintenance treatment with delgocitinib ointment twice weekly in adult patients with atopic dermatitis. *Dermatol. Ther.* **2022**, *35*, e15526. [CrossRef]
20. McLornan, D.P.; E Pope, J.; Gotlib, J.; Harrison, C.N. Current and future status of JAK inhibitors. *Lancet* **2021**, *398*, 803–816. [CrossRef]
21. Papp, K.; Szepietowski, J.C.; Kircik, L.; Toth, D.; Eichenfield, L.F.; Leung, D.Y.; Forman, S.B.; Venturanza, M.E.; Sun, K.; Kuligowski, M.E.; et al. Efficacy and safety of ruxolitinib cream for the treatment of atopic dermatitis: Results from 2 phase 3, randomized, double-blind studies. *J. Am. Acad. Dermatol.* **2021**, *85*, 863–872. [CrossRef] [PubMed]

22. Papp, K.; Szepietowski, J.C.; Kircik, L.; Toth, D.; Eichenfield, L.F.; Forman, S.B.; Kuligowski, M.E.; Venturanza, M.E.; Sun, K.; Simpson, E.L. Long-term safety and disease control with ruxolitinib cream in atopic dermatitis: Results from two phase III studies. Revolutionizing Atopic Dermatitis. *Br. J. Dermatol.* **2021**, *185*, e109–e146. [CrossRef]
23. Bissonnette, R.; Papp, K.A.; Poulin, Y.; Gooderham, M.; Raman, M.; Mallbris, L.; Wang, C.; Purohit, V.; Mamolo, C.; Papacharalambous, J.; et al. Topical tofacitinib for atopic dermatitis: A phase II a randomized trial. *Br. J. Dermatol.* **2016**, *175*, 902–911. [CrossRef]
24. Dose Ranging Study to Assess Efficacy, Safety, Tolerability and Pharmacokinetics of PF-06700841 Topical Cream in Participants With Mild or Moderate Atopic Dermatitis. Available online: https://clinicaltrials.gov/ct2/show/results/NCT03903822?term=PF-06700841&cond=Atopic+Dermatitis&draw=2&rank=1&view=results (accessed on 2 June 2022).
25. Aclaris Therapeutics Announces Positive Preliminary Topline Data from Phase 2a Trial of ATI-1777 for Moderate to Severe Atopic Dermatitis. Available online: https://www.globenewswire.com/news-release/2021/06/08/2243460/37216/en/Aclaris-Therapeutics-Announces-Positive-Preliminary-Topline-Data-from-Phase-2a-Trial-of-ATI-1777-for-Moderate-to-Severe-Atopic-Dermatitis.html (accessed on 4 June 2022).
26. Smith, S.; Bhatia, N.; Shanler, S.D.; Demoor, R.; Schnyder, J. 16089 Safety of ATI-502, a novel topical JAK1/3 inhibitor, in adults with moderate to severe atopic dermatitis: Results from a phase 2a open-label trial. *J. Am. Acad. Dermatol.* **2020**, *83*, AB170. [CrossRef]
27. Dastidar, S.G.; Rajagopal, D.; Ray, A. Therapeutic benefit of PDE4 inhibitors in inflammatory diseases. *Curr. Opin. Investig. Drugs.* **2007**, *8*, 364–372. [PubMed]
28. Bäumer, W.; Hoppmann, J.; Rundfeldt, C.; Kietzmann, M. Highly selective phosphodiesterase 4 inhibitors for the treatment of allergic skin diseases and psoriasis. *Inflamm. Allergy-Drug Targets* **2007**, *6*, 17–26. [CrossRef]
29. Hanifin, J.M.; Chan, S.C.; Cheng, J.B.; Tofte, S.J.; Henderson, W.R.; Kirby, D.S.; Weiner, E.S. Type 4 Phosphodiesterase Inhibitors Have Clinical and In Vitro Anti-inflammatory Effects in Atopic Dermatitis. *J. Investig. Dermatol.* **1996**, *107*, 51–56. [CrossRef]
30. Jimenez, J.L.; Punzon, C.; Navarro, J.; Muñoz-Fernández, M.A.; Fresno, M. Phosphodiesterase 4 inhibitors prevent cytokine secretion by T lymphocytes by inhibiting nuclear factor-kappaB and nuclear factor of activated T cells activation. *J. Pharmacol. Exp. Ther.* **2001**, *299*, 753–759.
31. Paller, A.S.; Tom, W.L.; Lebwohl, M.G.; Blumenthal, R.L.; Boguniewicz, M.; Call, R.S.; Eichenfield, L.F.; Forsha, D.W.; Rees, W.C.; Simpson, E.L.; et al. Efficacy and safety of crisaborole ointment, a novel, nonsteroidal phosphodiesterase 4 (PDE4) inhibitor for the topical treatment of atopic dermatitis (AD) in children and adults. *J. Am. Acad. Dermatol.* **2016**, *75*, 494–503.e6. [CrossRef]
32. Schlessinger, J.; Shepard, J.S.; Gower, R.; Su, J.C.; Lynde, C.; Cha, A.; Ports, W.C.; Purohit, V.; Takiya, L.; Werth, J.L.; et al. Safety, Effectiveness, and Pharmacokinetics of Crisaborole in Infants Aged 3 to <24 Months with Mild-to-Moderate Atopic Dermatitis: A Phase IV Open-Label Study (CrisADe CARE 1). *Am. J. Clin. Dermatol.* **2020**, *21*, 275–284. [CrossRef]
33. Lin, C.P.-L.; Gordon, S.; Her, M.J.; Rosmarin, D. A retrospective study: Application site pain with the use of crisaborole, a topical phosphodiesterase 4 inhibitor. *J. Am. Acad. Dermatol.* **2019**, *80*, 1451–1453. [CrossRef]
34. Eichenfield, L.F.; Call, R.S.; Forsha, D.W.; Fowler, J., Jr.; Hebert, A.A.; Spellman, M.; Stein Gold, L.F.; Van Syoc, M.; Zane, L.T.; Tschen, E. Long-term safety of crisaborole ointment 2% in children and adults with mild to moderate atopic dermatitis. *J. Am. Acad. Dermatol.* **2017**, *77*, 641–649.e5. [CrossRef] [PubMed]
35. Bissonnette, R.; Pavel, A.B.; Diaz, A.; Werth, J.L.; Zang, C.; Vranic, I.; Purohit, V.S.; Zielinski, M.A.; Vlahos, B.; Estrada, Y.D.; et al. Crisaborole and atopic dermatitis skin biomarkers: An intrapatient randomized trial. *J. Allergy Clin. Immunol.* **2019**, *144*, 1274–1289. [CrossRef] [PubMed]
36. Hanifin, J.M.; Ellis, C.N.; Frieden, I.J.; Fölster-Holst, R.; Stein Gold, L.F.; Secci, A.; Smith, A.J.; Zhao, C.; Kornyeyeva, E.; Eichenfield, L.F. OPA-15406, a novel, topical, nonsteroidal, selective phosphodiesterase-4 (PDE4) inhibitor, in the treatment of adult and adolescent patients with mild to moderate atopic dermatitis (AD): A phase-II randomized, double-blind, placebo-controlled study. *J. Am. Acad. Dermatol.* **2016**, *75*, 297–305. [CrossRef] [PubMed]
37. Saeki, H.; Ito, K.; Yokota, D.; Tsubouchi, H. Difamilast ointment in adult patients with atopic dermatitis: A phase 3 randomized, double-blind, vehicle-controlled trial. *J. Am. Acad. Dermatol.* **2022**, *86*, 607–614. [CrossRef] [PubMed]
38. Ohba, F.; Nomoto, M.; Hojo, S.; Akama, H. Safety, tolerability and pharmacokinetics of a novel phosphodiesterase inhibitor, E6005 ointment, in healthy volunteers and in patients with atopic dermatitis. *J. Dermatol. Treat.* **2016**, *27*, 241–246. [CrossRef] [PubMed]
39. Ohba, F.; Matsuki, S.; Imayama, S.; Matsuguma, K.; Hojo, S.; Nomoto, M.; Akama, H. Efficacy of a novel phosphodiesterase inhibitor, E6005, in patients with atopic dermatitis: An investigator-blinded, vehicle-controlled study. *J. Dermatol. Treat.* **2016**, *27*, 467–472. [CrossRef]
40. Furue, M.; Kitahara, Y.; Akama, H.; Hojo, S.; Hayashi, N.; Nakagawa, H. The JAPANESE E6005 Study Investigators Safety and efficacy of topical E6005, a phosphodiesterase 4 inhibitor, in Japanese adult patients with atopic dermatitis: Results of a randomized, vehicle-controlled, multicenter clinical trial. *J. Dermatol.* **2014**, *41*, 577–585. [CrossRef]
41. Nguyen, H.L.; Anderson, K.R.; Tollefson, M.M. New and Emerging Therapies for Pediatric Atopic Dermatitis. *Pediatr. Drugs* **2019**, *21*, 239–260. [CrossRef]
42. Ilyas, M.; Logas, C.M.; Glick, B.P.; Del Rosso, J.Q. Advancements in topical therapy. *Dermatol. Rev.* **2022**, *3*, 9–13. [CrossRef]
43. Denison, M.S.; Nagy, S.R. Activation of the Aryl Hydrocarbon Receptor by Structurally Diverse Exogenous and Endogenous Chemicals. *Annu. Rev. Pharmacol. Toxicol.* **2003**, *43*, 309–334. [CrossRef] [PubMed]

44. Gutiérrez-Vázquez, C.; Quintana, F.J. Regulation of the Immune Response by the Aryl Hydrocarbon Receptor. *Immunity* **2018**, *48*, 19–33. [CrossRef] [PubMed]
45. Furue, M.; Nakahara, T. Revival of AHR Agonist for the Treatment of Atopic Dermatitis: Tapinarof. *Curr. Treat. Options Allergy* **2020**, *7*, 414–421.e3. [CrossRef]
46. Furue, M.; Tsuji, G.; Mitoma, C.; Nakahara, T.; Chiba, T.; Morino-Koga, S.; Uchi, H. Gene regulation of filaggrin and other skin barrier proteins via aryl hydrocarbon receptor. *J. Dermatol. Sci.* **2015**, *80*, 83–88. [CrossRef] [PubMed]
47. Bissonnette, R.; Gold, L.S.; Rubenstein, D.S.; Tallman, A.M.; Armstrong, A. Tapinarof in the treatment of psoriasis: A review of the unique mechanism of action of a novel therapeutic aryl hydrocarbon receptor–modulating agent. *J. Am. Acad. Dermatol.* **2021**, *84*, 1059–1067. [CrossRef]
48. Paghdal, K.V.; Schwartz, R.A. Topical tar: Back to the future. *J. Am. Acad. Dermatol.* **2009**, *61*, 294–302. [CrossRef]
49. FDA Approves Dermavant's VTAMA® (Tapinarof) Cream, 1% for the Treatment of Plaque Psoriasis in Adults: First Topical Novel Chemical Entity Launched for Psoriasis in the U.S. in 25 Years. Available online: https://www.globenewswire.com/news-release/2022/05/24/2449068/34323/en/FDA-Approves-Dermavant-s-VTAMA-tapinarof-cream-1-for-the-Treatment-of-Plaque-Psoriasis-in-Adults-First-Topical-Novel-Chemical-Entity-Launched-for-Psoriasis-in-the-U-S-in-25-Years.html (accessed on 13 June 2022).
50. Paller, A.S.; Gold, L.S.; Soung, J.; Tallman, A.M.; Rubenstein, D.S.; Gooderham, M. Efficacy and patient-reported outcomes from a phase 2b, randomized clinical trial of tapinarof cream for the treatment of adolescents and adults with atopic dermatitis. *J. Am. Acad. Dermatol.* **2021**, *84*, 632–638. [CrossRef] [PubMed]
51. Peppers, J.; Paller, A.S.; Maeda-Chubachi, T.; Wu, S.; Robbins, K.; Gallagher, K.; Kraus, J.E. A phase 2, randomized dose-finding study of tapinarof (GSK2894512 cream) for the treatment of atopic dermatitis. *J. Am. Acad. Dermatol.* **2019**, *80*, 89–98.e3. [CrossRef]
52. Hwang, J.; Newton, E.M.; Hsiao, J.; Shi, V.Y. Aryl hydrocarbon receptor/nuclear factor E2-related factor 2 (AHR/NRF2) signalling: A novel therapeutic target for atopic dermatitis. *Exp. Dermatol.* **2022**, *31*, 485–497. [CrossRef]
53. Gold, L.S.; Rubenstein, D.S.; Peist, K.; Jain, P.; Tallman, A.M. Tapinarof cream 1% once daily and benvitimod 1% twice daily are 2 distinct topical medications. *J. Am. Acad. Dermatol.* **2021**, *85*, e201–e202. [CrossRef]
54. Messeguer, A.; Planells-Cases, R.; Ferrer-Montiel, A. Physiology and Pharmacology of the Vanilloid Receptor. *Curr. Neuropharmacol.* **2006**, *4*, 1–15. [CrossRef] [PubMed]
55. Yun, J.-W.; Seo, J.A.; Jeong, Y.S.; Bae, I.-H.; Jang, W.-H.; Lee, J.; Kim, S.-Y.; Shin, S.-S.; Woo, B.-Y.; Lee, K.-W. TRPV1 antagonist can suppress the atopic dermatitis-like symptoms by accelerating skin barrier recovery. *J. Dermatol. Sci.* **2011**, *62*, 8–15. [CrossRef]
56. Stander, S.; Moormann, C.; Schumacher, M.; Buddenkotte, J.; Artuc, M.; Shpacovitch, V.; Brzoska, T.; Lippert, U.; Henz, B.M.; Luger, T.A.; et al. Expression of vanilloid receptor subtype 1 in cutaneous sensory nerve fibers, mast cells, and epithelial cells of appendage structures. *Exp. Dermatol.* **2004**, *13*, 129–139. [CrossRef] [PubMed]
57. Song, P.; Armstrong, C. Novel therapeutic approach with PAC -14028 cream, a TRPV 1 antagonist, for patients with mild-to-moderate atopic dermatitis. *Br. J. Dermatol.* **2019**, *180*, 971–972. [CrossRef] [PubMed]
58. Park, C.W.; Kim, B.J.; Lee, Y.W.; Won, C.; Park, C.O.; Chung, B.Y.; Lee, D.H.; Jung, K.; Nam, H.-J.; Choi, G.; et al. Asivatrep, a TRPV1 antagonist, for the topical treatment of atopic dermatitis: Phase 3, randomized, vehicle-controlled study (CAPTAIN-AD). *J. Allergy Clin. Immunol.* **2022**, *149*, 1340–1347.e4. [CrossRef]
59. Lee, J.H.; Choi, C.S.; Bae, I.H.; Choi, J.K.; Park, Y.H.; Park, M. A novel, topical, nonsteroidal, TRPV1 antagonist, PAC-14028 cream improves skin barrier function and exerts anti-inflammatory action through modulating epidermal differentiation markers and suppressing Th2 cytokines in atopic dermatitis. *J. Dermatol. Sci.* **2018**, *30*, 184–194. [CrossRef]
60. Choi, J.K.; Cho, W.; Lee, J.-H.; Choi, G.; Park, M. A TRPV1 antagonist, PAC-14028 does not increase the risk of tumorigenesis in chemically induced mouse skin carcinogenesis. *Regul. Toxicol. Pharmacol.* **2020**, *112*, 104613. [CrossRef]
61. Park, M.; Naidoo, A.A.; Burns, A.; Choi, J.K.; Gatfield, K.M.; Vidgeon-Hart, M.; Bae, I.-H.; Lee, C.S.; Choi, G.; Powell, A.; et al. Do TRPV1 antagonists increase the risk for skin tumourigenesis? A collaborative in vitro and in vivo assessment. *Cell Biol. Toxicol.* **2018**, *34*, 143–162. [CrossRef]
62. Lee, Y.; Won, C.-H.; Jung, K.; Nam, H.-J.; Choi, G.; Park, Y.-H.; Park, M.; Kim, B. Efficacy and safety of PAC -14028 Cream—A novel, topical, nonsteroidal, selective TRPV 1 antagonist in patients with mild-to-moderate atopic dermatitis: A phase II b randomized trial. *Br. J. Dermatol.* **2019**, *180*, 1030–1038. [CrossRef]
63. Ederveen, T.H.A.; Smits, J.P.H.; Boekhorst, J.; Schalkwijk, J.; Bogaard, E.H.V.D.; Zeeuwen, P.L.J.M. Skin microbiota in health and disease: From sequencing to biology. *J. Dermatol.* **2020**, *47*, 1110–1118. [CrossRef]
64. Kong, H.H.; Oh, J.; Deming, C.; Conlan, S.; Grice, E.A.; Beatson, M.A.; Nomicos, E.; Polley, E.C.; Komarow, H.D.; Murray, P.R.; et al. Temporal shifts in the skin microbiome associated with disease flares and treatment in children with atopic dermatitis. *Genome Res.* **2012**, *22*, 850–859. [CrossRef] [PubMed]
65. Simpson, E.L.; Villarreal, M.; Jepson, B.; Rafaels, N.; David, G.; Hanifin, J.; Taylor, P.; Boguniewicz, M.; Yoshida, T.; De Benedetto, A.; et al. Patients with Atopic Dermatitis Colonized with Staphylococcus aureus Have a Distinct Phenotype and Endotype. *J. Investig. Dermatol.* **2018**, *138*, 2224–2233. [CrossRef] [PubMed]
66. Na, C.; Baghoomian, W.; Simpson, E. A Therapeutic Renaissance—Emerging Treatments for Atopic Dermatitis. *Acta Derm. Venereol.* **2020**, *100*, adv00165. [CrossRef] [PubMed]
67. Lunjani, N.; Hlela, C.; O'Mahony, L. Microbiome and skin biology. *Curr. Opin. Allergy Clin. Immunol.* **2019**, *19*, 328–333. [CrossRef] [PubMed]

68. Bieber, T. Atopic dermatitis: An expanding therapeutic pipeline for a complex disease. *Nat. Rev. Drug Discov.* **2022**, *21*, 21–40. [CrossRef]
69. Paller, A.S.; Kong, H.H.; Seed, P.; Naik, S.; Scharschmidt, T.C.; Gallo, R.L.; Luger, T.; Irvine, A.D. The microbiome in patients with atopic dermatitis. *J. Allergy Clin. Immunol.* **2019**, *143*, 26–35. [CrossRef]
70. Myles, I.A.; Earland, N.J.; Anderson, E.D.; Moore, I.N.; Kieh, M.D.; Williams, K.W.; Saleem, A.; Fontecilla, N.M.; Welch, P.A.; Darnell, D.A.; et al. First-in-human topical microbiome transplantation with Roseomonas mucosa for atopic dermatitis. *JCI Insight* **2018**, *3*, e120608. [CrossRef]
71. Clinical Trial of FB-401 For the Treatment of Atopic Dermatitis Fails to Meet Statistical Significance. Available online: https://www.fortebiorx.com/investor-relations/news/news-details/2021/Clinical-Trial-of-FB-401-For-the-Treatment-of-Atopic-Dermatitis-Fails-to-Meet-Statistical-Significance/default.aspx (accessed on 14 June 2022).
72. Nakatsuji, T.; Hata, T.R.; Tong, Y.; Cheng, J.Y.; Shafiq, F.; Butcher, A.M.; Salem, S.S.; Brinton, S.L.; Spergel, A.K.R.; Johnson, K.; et al. Development of a human skin commensal microbe for bacteriotherapy of atopic dermatitis and use in a phase 1 randomized clinical trial. *Nat. Med.* **2021**, *27*, 700–709. [CrossRef]
73. Todd Krueger Discusses AOBiome's Positive Results from its Phase 2b Trial for Both Pruritus (Itch) and Appearance of Atopic Dermatitis (Eczema). Available online: https://www.aobiome.com/pressreleases/aobiomes-ceo-president-todd-krueger-featured-in-ceo-cfo-magazine/#primary (accessed on 14 June 2022).
74. Vakharia, P.P.; Silverberg, J.I. New therapies for atopic dermatitis: Additional treatment classes. *J. Am. Acad. Dermatol.* **2018**, *78*, S76–S83. [CrossRef]
75. Hancock, R.E.W.; Scott, M.G. The role of antimicrobial peptides in animal defenses. *Proc. Natl. Acad. Sci. USA* **2000**, *97*, 8856–8861. [CrossRef]
76. Der Kolk, T.N.; Van Der Wall, H.; Hogendoorn, G.K.; Rijneveld, R.; Luijten, S.; Van Alewijk, D.C.; Munckhof, E.H.V.D.; De Kam, M.L.; Feiss, G.L.; Prens, E.P.; et al. Pharmacodynamic Effects of Topical Omiganan in Patients With Mild to Moderate Atopic Dermatitis in a Randomized, Placebo-Controlled, Phase II Trial. *Clin. Transl. Sci.* **2020**, *13*, 994–1003. [CrossRef]
77. Niemeyer-van der Kolk, T.; Buters, T.P.; Krouwels, L.; Boltjes, J.; de Kam, M.L.; van der Wall, H.; van Alewijk, D.C.J.G.; van den Munckhof, E.H.A.; Becker, M.J.; Feiss, G.; et al. Topical antimicrobial peptide omiganan recovers cutaneous dysbiosis but does not improve clinical symptoms in patients with mild to moderate atopic dermatitis in a phase 2 randomized controlled trial. *J. Am. Acad. Dermatol.* **2022**, *86*, 854–862. [CrossRef] [PubMed]
78. Guttman-Yassky, E.; Bissonnette, R.; Pavel, A.; Proulx, E.S.-C.; Prokocimer, P.; Toft-Kehler, R.; Sommer, M. 563 ATx201 modulates biomarkers of skin barrier function and cutaneous inflammation in patients with moderate atopic dermatitis. *J. Investig. Dermatol.* **2020**, *140*, S77. [CrossRef]
79. Wollenberg, A.; Barbarot, S.; Bieber, T.; Christen-Zaech, S.; Deleuran, M.; Fink-Wagner, A.; Gieler, U.; Girolomoni, G.; Lau, S.; Muraro, A.; et al. Consensus-based European guidelines for treatment of atopic eczema (atopic dermatitis) in adults and children: Part I. *J. Eur. Acad. Dermatol. Venereol.* **2018**, *32*, 657–682. [CrossRef] [PubMed]
80. Eichenfield, L.F.; Tom, W.L.; Berger, T.G.; Krol, A.; Paller, A.S.; Schwarzenberger, K.; Bergman, J.N.; Chamlin, S.L.; Cohen, D.E.; Cooper, K.D.; et al. Guidelines of care for the management of atopic dermatitis. *J. Am. Acad. Dermatol.* **2014**, *71*, 116–132. [CrossRef]
81. Bs, L.M.L.; Lee, D.E.; Shi, V.Y. A comparison of international management guidelines for atopic dermatitis. *Pediatr. Dermatol.* **2019**, *36*, 36–65. [CrossRef]
82. Wollenberg, A.; Kinberger, M.; Arents, B.; Aszodi, N.; Avila Valle, G.; Barbarot, S.; Bieber, T.; Brough, H.A.; Calzavara Pinton, P.; Christen-Zäch, S.; et al. Eurogуiderm Guideline On Atopic Eczema, Version 1.0, June 2022, European Dermatology Forum. Available online: https://www.edf.one/dam/jcr:0a8bb62f-7abe-4f86-84d9-bc0a0b370efb/0_Atopic_Eczema_GL_full_version_Jun_2022_.pdf (accessed on 27 July 2022).
83. Boralevi, F.; Aroman, M.S.; Delarue, A.; Raudsepp, H.; Kaszuba, A.; Bylaite, M.; Tiplica, G.S. Long-term emollient therapy improves xerosis in children with atopic dermatitis. *J. Eur. Acad. Dermatol. Venereol.* **2014**, *28*, 1456–1462. [CrossRef]
84. Correa, M.C.M.; Nebus, J. Management of Patients with Atopic Dermatitis: The Role of Emollient Therapy. *Dermatol. Res. Pract.* **2012**, *2012*, 836931. [CrossRef]
85. Van Zuuren, E.J.; Fedorowicz, Z.; Arents, B.W.M. Performance and Tolerability of the Moisturizers Cetaphil® and Excipial® in Atopic Dermatitis: What is the Evidence Based on Randomized Trials? *Dermatol. Ther.* **2017**, *7*, 331–347. [CrossRef]
86. Aries, M.F.; Hernandez-Pigeon, H.; Vaissière, C.; Delga, H.; Caruana, A.; Lévêque, M.; Bourrain, M.; Ravard Helffer, K.; Chol, B.; Nguyen, T.; et al. Anti-inflammatory and immunomodulatory effects of Aquaphilus dolomiae extract on in vitro models. *Clin. Cosmet. Investig. Dermatol.* **2016**, *9*, 421–434. [CrossRef]
87. Mahe, Y.F.; Perez, M.J.; Tacheau, C.; Fanchon, C.; Martin, R.; Rousset, F.; Seite, S. A new Vitreoscilla filiformis extract grown on spa water-enriched medium activates endogenous cutaneous antioxidant and antimicrobial defenses through a potential Toll-like receptor 2/protein kinase C, zeta transduction pathway. *Clin. Cosmet. Investig. Dermatol.* **2013**, *6*, 191–196. [CrossRef]
88. Fostini, A.C.; Georgescu, V.; DeCoster, C.J.; Girolomoni, G. A cream based on Aquaphilus dolomiae extracts alleviates non-histaminergic pruritus in humans. *Eur. J. Dermatol.* **2017**, *27*, 317–318. [CrossRef] [PubMed]
89. Gueniche, A.; Knaudt, B.; Schuck, E.; Volz, T.; Bastien, P.; Martin, R.; Röcken, M.; Breton, L.; Biedermann, T. Effects of nonpathogenic gram-negative bacterium *Vitreoscilla filiformis* lysate on atopic dermatitis: A prospective, randomized, double-blind, placebo-controlled clinical study. *Br. J. Dermatol.* **2008**, *159*, 1357–1363. [CrossRef] [PubMed]

90. Bianchi, P.; Theunis, J.; Casas, C.; Villeneuve, C.; Patrizi, A.; Phulpin, C.; Bacquey, A.; Redoulès, D.; Mengeaud, V.; Schmitt, A. Effects of a New Emollient-Based Treatment on Skin Microflora Balance and Barrier Function in Children with Mild Atopic Dermatitis. *Pediatr. Dermatol.* **2016**, *33*, 165–171. [CrossRef] [PubMed]
91. Williams, H.C. New topical treatments for atopic dermatitis: Active comparators are needed. *J. Am. Acad. Dermatol.* **2021**, *85*, 1065–1066. [CrossRef]
92. Zhang, L.; Du, D.; Wang, L.; Guo, L.; Jiang, X. Efficacy and safety of topical Janus kinase and phosphodiesterase inhibitor-4 inhibitors for the treatment of atopic dermatitis: A network meta-analysis. *J. Dermatol.* **2021**, *48*, 1877–1883. [CrossRef] [PubMed]

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National Information Campaign Revealed Disease Characteristic and Burden in Adult Patients Suffering from Atopic Dermatitis

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Abstract: Atopic dermatitis (AD) is a common inflammatory skin disease often associated with a significant impairment in the quality of life of affected patients. The Italian Society of Dermatology and Venereology (SIDeMaST) planned a national information campaign, providing direct access to 27 dermatologic centers dedicated to the management of AD. The aim of this study aimed was to outline critical aspects related to AD in the general population. Overall, 643 adult subjects were included in this study, and in 44.2% (284/643) of cases, a diagnosis of AD was confirmed, whereas about 55% of subjects were affected by other pruritic cutaneous diseases. Higher intensity of pruritus and sleep disturbance, as well as an increased interference in sport, work, and social confidence

was reported in the AD group compared to the non-AD group. In the AD subgroup, the mean duration of disease was of 15.3 years, with a mean eczema area and severity index (EASI) score of 11.2, and investigator global assessment (IGA) score of 1.9 and an itch numeric rating scale (NRS) of 6.9. Almost 32% of patients were untreated, either with topical or systemic agents, whereas 44.3% used routine topical compounds (topical corticosteroids and calcineurin inhibitors), and only 7.0% of patients were systemically treated. Only 2.8% of patients reported complete satisfaction with the treatment received for AD to date. This study reveals a profound unmet need in AD, showing a poorly managed and undertreated patient population despite a high reported burden of disease. This suggests the usefulness of information campaigns with the goal of improving patient awareness regarding AD and facilitating early diagnosis and access to dedicated healthcare institutions.

Keywords: atopic dermatitis; information campaign; early diagnosis

1. Introduction

Atopic dermatitis (AD) is the most common chronic inflammatory skin disease, affecting nearly 230 million people worldwide, with a prevalence, in developed countries, ranging between 10% and 25% in children and 7–10% in adults [1]. AD is clinically characterized by intense itch, dry skin, and eczematous lesions, with the preferential involvement of flexures, head, neck, and hands in adulthood [1]. It is frequently associated with a personal and/or family history of atopic extracutaneous manifestations, such as allergic rhinitis, conjunctivitis, and asthma [1]. Several non-atopic diseases, including inflammatory, autoimmune, and mental health disorders, might be also observed in AD patients [2]. In addition, attention disturbances and poor sleep quality, likely related to itch, may occur, affecting school and work performance [3]. Treatment of mild AD is essentially based on the use of topical corticosteroids (TCSs) and calcineurin inhibitors (TCIs), whereas moderate-to-severe AD, accounting for as many as one-third of cases, is commonly treated with phototherapy and/or systemic therapies, including traditional immunosuppressants and novel immune-targeted therapies [4,5]. In addition, to improve skin dryness and reduce itch, moisturizers are usually applied daily, implying a significant economic burden for patients [6]. Notably, a recent cross-sectional study including nine European countries reported a mean annual personal extra out-of-pocket expense of EUR 927.12 for patients with AD [6].

Although they were conceived in the 1980s, the Hanifin and Rajka criteria are still the most used tool to diagnose AD in both clinical practice and research settings, whereas, considering the lack of specific diagnostic markers, diagnosis of AD is essentially based on the accurate evaluation of clinical signs, symptoms, and medical history by skilled physicians [7]. Diagnosis of AD is relatively easy in children but often challenging in adults, especially in late-onset forms, due to a broader clinical variability [7,8]. For this reason, adult AD is thought to be underdiagnosed, reflecting the highly variable prevalence of disease reported in the literature, with an estimated range varying from 0.3% to 14.3% [8–10]. To reduce the proportion of undiagnosed and undertreated cases of AD, it might be helpful to promote awareness of AD through multiple strategies, including the diffusion of disease information websites and the organization of screening campaigns. Despite the existence of numerous scientific societies and patient associations providing educational websites for AD patients, the first national information and screening campaign was organized in September 2020 by the Italian Society of Dermatology and Venereology (SIDeMaST), which provided direct access to several dermatologic centers dedicated to the management of AD, with the aim of improving patient awareness about AD and facilitating early diagnosis and access to optimal treatment management.

2. Materials and Methods

In this study, we considered data obtained from subjects referred to 27 dermatology centers homogeneously distributed in northern, central, and southern Italy for the first national AD screening program supported by the Italian Society of Dermatology (SIDeMaST). The purpose of the program was to promote the knowledge of AD among adult subjects with established disease or suspected symptoms.

In September 2020, information regarding the national AD screening program was posted on the patient-oriented AD website (www.dallapartedellatuapelle.it). In particular, the web site provided general information regarding pathogenesis and clinical presentation of AD in adults, as well as email and telephone contacts to join the AD screening program. Patients with suspected or diagnosed AD were screened by non-physician personnel on the phone or by e-mail through a brief questionnaire. As a screening program, it was not necessary to apply for ethics committee approval because patients did not furnish any sensitive data to the centers.

Inclusion criteria for prescreened patients were limited to comprehension of written Italian language and consent to compile a printed survey. In each dermatological center, patients were required to complete a 21-item questionnaire about demographic and clinical data, including age, sex, weight, height, job, disease duration, type of medical specialists previously consulted, personal and family history of AD and comorbidities, interference of AD with physical activities and work tasks, therapeutic management of disease, economic burden for supplying topical and systemic drugs, and consultations for AD. Individuals were subdivided according to working profession as white collar (intellectual jobs, including doctors, lawyers, teachers, office workers, managers, and civil servants) or blue collar (manual jobs, including craftsmen, farmers, specialized workmen, drivers of industrial machines/vehicles, armed services, and unqualified professions) [11].

Patients were evaluated by dermatologists with experience in inflammatory skin diseases to assess the diagnosis of AD and suggest the most appropriate therapeutic approach. Disease severity in patients with a confirmed diagnosis of AD was assessed using (a) the eczema area severity index (EASI), with scored ranging from 0 to 72; (b) the investigator global assessment (IGA), with scores ranging from 0 to 4; (c) the itch numeric rating scale (NRS), ranging from 0 to 10, assessing itch intensity (itch-NRS); (d) a 0–10 NRS scale rating sleeplessness (sleep-NRS); (e) a 0–10 NRS evaluating disease-induced embarrassment (e-NRS); (f) a 0–10 NRS evaluating the influence of disease on work tasks (w-NRS); and (g) a 0–10 NRS assessing the impact of disease on sporting activity (s-NRS).

Statistical Analyses

Categorical variables were analyzed as frequencies and percentages. Continuous variables were analyzed as mean and standard deviation (SD) or medians and interquartile ranges (IQRs). Variable normality was assessed by the Shapiro–Wilk W test. We compared questionnaire-obtained personal and clinical data between AD and non-AD groups, using a T test for comparison of means or Mann–Whitney test for comparison of medians, and by chi-square (or Fisher's exact test) for categorical variables. In the subgroup with confirmed AD diagnosis, clinical data were described in terms of frequencies and percentages, whereas EASI and IGA scores were analyzed as mean and SD. All statistical tests were two-tailed, and a p-value less than 0.05 was considered statistically significant. Analysis was performed using STATA 17 software (StataCorp, College Station, TX, USA).

3. Results

3.1. Characteristic of the General Population

Overall, 641 adult subjects were referred to the 27 outpatient dermatology centers during the open day in September 2020. Demographic and clinical data are summarized in Table 1.

Table 1. Characteristics of general population, and separately for atopic dermatitis (AD) and non-AD.

	General Population	AD Population	Non-AD Population	*p*-Value *
Patients N tot	641	284	357	
Males n (%)	246 (38.4)	96 (33.8)	150 (42.0)	0.033
Age (mean ± SD)	46.2 ± 19.4	36.9 ± 16.4	53.7 ± 17.9	<0.0001
BMI (mean ± SD)	24.3 ± 4.4	23.5 ± 4.4	25.1 ± 0.23	<0.0001
Previous diagnosis of AD n (%)	362 (56.5)	219 (77.1)	126 (35.3)	<0.0001
Disease duration (mean ± SD)	10.7 ± 11.8	15.3 ± 12.6	7.0 ± 9.7	<0.0001
Family history of AD n (%)	115 (17.9)	66 (23.2)	47 (13.1)	<0.0001
Family history of other atopic comorbidities n (%)	183 (28.5)	109 (38.4)	72 (20.2)	<0.0001
Job Title				
Intellectual jobs (white collars) n (%)	260 (40.6)	124 (43.7)	136 (38.1)	0.176
Manual jobs (blue collars) n (%)	154 (24.0)	78 (27.5)	76 (21.3)	0.069
Physicians Previously Consulted				
Dermatologist n (%)	542 (84.5)	264 (93.0)	278 (77.9)	<0.0001
Allergologist n (%)	219 (34.2)	139 (48.9)	80 (22.4)	<0.0001
General practitioner n (%)	39 (6.1)	6 (2.1)	33 (9.2)	<0.0001
Patient's Related Outcomes				
Itch intensity (mean, SD)	6.5 ± 2.5	6.9 ± 2.4	6.2 ± 2.6	0.0003
Interference with sleep (mean ± SD)	4.4 ± 3.4	5.0 ± 3.4	3.9 ± 3.4	<0.0001
Interference with work (mean ± SD)	3.7 ± 3.4	4.3 ± 3.5	3.4 ± 3.4	0.002
Interference with sport (mean ± SD)	3.4 ± 3.4	4.1 ± 3.4	2.8 ± 3.2	<0.0001
Disease induced embarassment (mean ± SD)	5.7 ± 3.2	6.4 ± 2.9	5.1 ± 3.3	<0.0001
Treatments Routinely Used				
Topical compounds n (%)	259 (40.4)	126 (44.3)	133 (37.2)	<0.0001
Moisturizers n (%)	294 (45.8)	160 (56.3)	134 (37.5)	<0.0001
Systemic therapies n (%)	41 (6.4)	20 (7.0)	21 (5.9)	0.07
No treatments n (%)	225 (35.1)	90 (31.7)	135 (37.9)	<0.0001

Legend. Categorical data expressed as n (%); continuous data expressed as mean ± standard deviation * *p* value refers to the comparison between AD and non-AD population.

Work activity was classified as intellectual jobs (white collars), accounting for 40.6% of participants (260/641); or manual jobs (blue collars), accounting for 24.0% of participants (154/641); whereas 18.4% of participants (118/641) were students and 21.5% (138/641) were retired professionals. Proportions of 84.9% (544/641) and 34.5% (221/641) of participants reported to have visited a dermatologist an allergist, respectively, at least one time for their skin problems, whereas only 6.4% (41/641) were managed by their general practitioner.

The mean duration of skin manifestations was 10.7 ± 11.8 years. Mean itch-NRS and sleep-NRS values at the time of evaluation were 6.5 ± 2.5 and 4.4 ± 3.4, respectively. Approximately 55% (352/641) of patients reported itch as the major cause of discomfort, whereas another 17.3% (111/641) of subjects identified the presence of eczematous lesions as the main burdening factor, with 26.5% (170/641) reporting being equally disturbed by both manifestations.

3.2. Clinical Features of the AD Subpopulation Differ from Those of the Non-AD Population

A diagnosis of AD was confirmed in 44.3% (284/641) of the screened population.

On the other hand, 55.7% of subjects were included in the non-AD group, as they reported being affected by other pruritic cutaneous disease, including seborrheic dermatitis, psoriasis, allergic contact dermatitis, scabies, prurigo nodularis, dermatitis herpetiformis, bullous pemphigoid, and pruritus.

Demographic and clinical data of each subpopulation are summarized in Table 1.

Approximately 77% (219/284) of AD patients had previously received a diagnosis of AD. In 35.3% (126/357) of non-AD patients, an erroneous diagnosis of AD had been previously made. Most AD patients (93.0%; 264/284) visited a dermatologist at least once, compared to 77.9% (278/357) of the non-AD population, whereas 48.9% (139/284) of AD compared to 20.4% (80/357) of non-AD patients had visited their skin disease evaluated by an allergologist and 2.1% (6/284) of AD versus 9.2% (33/357) of non-AD patients by a general practitioner. Mean duration of skin disease was 15.3 ± 12.6 years in the AD group and 7.0 ± 9.7 in the non-AD group ($p < 0.0001$). A family history of AD, rhino-conjunctivitis, and asthma was significantly more frequent in AD patients compared to non-AD patients ($p < 0.0001$). A higher intensity of pruritus and sleep deterioration were detected in the AD group, with mean values of itch- and sleep-NRS of 6.9 ± 2.4 and 5.0 ± 3.4, respectively, compared to 6.2 ± 2.6 and 3.9 ± 3.4 in the non-AD cohort (Table 1). Moreover, a higher grade of disease-related embarrassment and disease interference with sport and work was reported in the AD group compared with the non-AD population (Table 1). The regular use of TCS or TCI was reported in 44.3% (126/284) of AD patients compared to 37.2% (133/357) of non-AD patients ($p < 0.0001$). In addition, AD patients reported a greater use of moisturizers compared to non-AD patients (56.3% (160/284) versus 37.5% (134/357), $p < 0.0001$). On the contrary, no significant differences between the two populations were detected in terms of the use of systemic therapies. Only 2.8% (8/284) of AD patients and 4.5% (16/357) of the non-AD group reported complete satisfaction with therapy received, whereas 34.5% (98/284) of AD subjects and 18.8% (67/357) of non-AD sub-cohort reported only partial satisfaction, and 57.7% (164/284) of AD patients and 60.2% (215/357) of non-AD patients reported no satisfaction ($p < 0.0001$).

No significant difference was detected in terms of the average monthly expense for topicals, systemic drugs, and visits between the AD and non-AD groups (Table 2).

Table 2. Predictors of average monthly expense > 20 euros for topical therapies, systemic therapies, and visits.

	Monthly Expense for Topical Therapies > 20 Euros OR (95%CI)	Monthly Expense for Systemic Therapies > 20 Euros OR (95%CI)	Monthly Expense for Visits > 20 Euros OR (95%CI)
EASI *	**1.03 (1.01–1.06)**	0.99 (0.96–1.02)	1.01 (0.99–1.03)
IGA scoring *	**1.56 (1.18–2.05)**	1.26 (0.89–1.78)	0.96 (0.72–1.28)
Pruritus (0–10) **	**1.12 (1.01–1.25)**	1.11 (0.94–1.31)	1.02 (0.90–1.15)
Embarassment (0–10) **	**1.19 (1.09–1.31)**	**1.21 (1.04–1.42)**	**1.14 (1.02–1.27)**
Interference with work (0–10) **	**1.15 (1.06–1.26)**	1.10 (0.99–1.24)	1.07 (0.98–1.17)
Interference with sport (0–10) **	**1.19 (1.07–1.32)**	1.09 (0.95–1.24)	1.09 (0.98–1.21)
Interference with sleep (0–10) **	**1.13 (1.04–1.23)**	1.04 (0.93–1.18)	1.06 (0.97–1.17)
Atopic comorbidities **	**1.35 (0.71–2.57)**	1.76 (0.65–4.78)	1.42 (0.69–2.95)
Job (ref: white collar) **			
Blue collar	0.98 (0.49–1.97)	1.88 (0.75–4.72)	1.49 (0.73–3.08)
Retired/unemployed	1.31 (0.47–3.6)	2.15 (0.56–8.28)	0.85 (0.26–2.77)

Legend. * Model adjusted for: age, gender ** Model adjusted for age, gender, EASI. Statistically significant results are highlighted in bold.

3.3. Physician-Oriented Assessment of AD Patients

In individuals with a confirmed diagnosis of AD (284 patients), mean EASI and mean IGA scores were 11.2 ± 12.0 and 1.9 ± 2.0, respectively. Moderate-to-severe AD, defined by an EASI score ≥16 and an IGA score ≥3, was reported in 21.5% (61/284) and 25.3% (72/284), respectively. The upper limbs were the body site more frequently affected by skin lesions (72.2%), followed by head and neck (49.6%), lower limbs (44,0%), trunk (28.2%), and back (25.7%). History of atopic comorbidities was reported in 48.2% of patients; in particular, rhinitis was described in 36.3% (103/284) of patients, conjunctivitis in 20.4% (58/284), and allergic asthma in 21.8% (62/284). In this patient population, the increase in disease severity scores was directly associated with a monthly expense for topical treatments of more than EUR 20, whereas no significant correlation between disease severity and monthly expense was observed for systemic drugs and visits (Table 2).

4. Discussion

AD is a chronic inflammatory skin disease associated with a significant deterioration of patients' quality of life [1,3]. Although it is the most common inflammatory skin disease, the current lack of specific diagnostic markers and criteria makes the identification of adult AD challenging, particularly in the adult-onset subtype [7,12].

The latest national and international guidelines suggest that diagnosis of AD in adulthood is essentially clinical, based on evaluation of morphology and distribution of lesions and the exclusion of possible differential diagnoses, including allergic contact dermatitis, scabies, dermatitis herpetiformis, and cutaneous lymphomas [7,13]. The lack of experienced general practitioners and territorial dermatologists in recognizing adult AD could result in an underestimation of disease prevalence and burden.

In this study, 44.3% (284/641) of the whole population received a clinical diagnosis of AD, and in 22.9% (65/284) of these cases, a different diagnosis was proposed during previous visits, most of which had been performed by dermatologists.

Notably, more that 35% (126/357) of patients who resulted not affected by AD in this study had received an incorrect diagnosis of AD during previous visits, thus revealing not only a low sensitivity but also a low specificity in the diagnosis of adult AD with possible overestimation of disease prevalence in some cases. This significant number of misdiagnosedcases of AD in the studied population suggests the importance of information campaigns dedicated to the general population and the relevance of scientific activities with respect to increasing knowledge and awareness of AD among physicians.

Notably, serological markers currently used by physicians to support the diagnosis of AD are limited to total and/or allergen-specific serum IgE levels and peripheral eosinophil counts, which are characterized by low sensibility and specificity [14]. The recent discovery of a new subset of T-cell cytokines and chemokines has resulted in the introduction of multiple potential biomarkers, including serum levels of CD30; macrophage-derived chemoattractant (MDC); interleukins (IL)-12, -16, -18, and -31; and thymus and activation-regulated chemokine (TARC) [14–16]. Although none of these novel biomarkers have proven reliable for the diagnosis of AD in clinical practice to date, we consider further research necessary in this field with the aim of simplifying the diagnosis and management of the disease [14].

In the AD subgroup, the mean duration of disease was of 15.3 years, with a mean EASI score of 11.2, IGA score of 1.9, itch-NRS of 6.9 and sleep-NRS of 5.0. Interestingly, AD patients showed the highest values of all patient-reported outcomes in comparison with non-AD patients. Statistically higher severity in sleep disturbances was observed in the AD group vs. 3.9 in non-AD group ($p < 0.0001$). Notably, sleep disturbances are a well-known manifestation associated with AD, with a prevalence ranging between 33% and 81.7% in adults, not exclusively related to itch but also to immunological and neuroendocrine imbalance [17]. A few studies suggested a correlation between poor sleep quality and AD, regardless of disease status, suggesting that repeated flares of AD over time can lead to behavior-related sleep disorders persisting despite disease remission [18]. Thus, high

prevalence of sleep disturbances detected in our AD patients could be also explained by the significantly longer duration of disease reported in the AD subgroup (15.6 years) compared to non-AD individuals (6.9 years) ($p < 0.0001$).

Furthermore, chronic sleep disorders have been identified as one of the most important risk factors for the development of several non-atopic comorbidities in AD, including mental health disorders (e.g., anxiety, depression) and cardiovascular diseases (e.g., coronary artery disease and hypertension) [1,2,19]. All these comorbidities may in turn adversely affect sleep quality and increase the disease burden of AD patients [19].

A recent survey including Irish adult patients affected by AD revealed a negative influence of disease on social and relational life, with 70% of patients reporting social anxiety, 65% avoiding sport and physical activities, 52% avoiding social activities, and 52% avoiding sexual intimacy [20]. Similarly, we detected significantly higher social embarrassment and interference with sport activities and job tasks in the AD subgroup compared to the non-AD subgroup, confirming AD as a severely debilitating cutaneous disease with multiple effects on patients' overall quality of life. The effect of AD on adults and children can currently be determined by different quality of life questionnaires, the most used of which are the Dermatology Quality of Life (DLQI), Children's Dermatology Quality of Life, and Infants Dermatology Quality of Life questionnaires; it is important to consider that all these tools are not specific for AD [21–23].

Considering the extensive and multimodal burden of AD, the development and evaluation of new specific questionnaires to evaluate the multiple domains influenced by AD would be very useful.

Almost one-third of patients did not use any compound, either topical or systemic, whereas only 7.0% of patients were treated with a systemic therapeutic agent. Notably, the economic burden of topical therapies, which are not covered by the national health care system, might negatively impact treatment access. In this study, we found a positive correlation between the monthly expense for topicals and both patient- and physician-assessed disease severity, suggesting that poorly controlled AD requires an increased use of topical agents. This increased use of topical agents could be due to undertreatment, which does not include systemic agents, which are only prescribed in a small percentage of patients (7.0%), notwithstanding the consistent number of subjects suffering from moderate-to-severe AD (EASI $\geq$ 16 and IGA $\geq$ 3 reported in 21.5% and 25.3% of patients, respectively).

Importantly, only 2.8% of patients reported complete satisfaction with treatments received to date, showing profound unmet therapeutic needs among adult patients affected by AD [24]. Notwithstanding the recent introduction of novel targeted therapies approved for AD, which can be prescribed by tertiary healthcare centers only, more than 80% of patients reported lack of awareness about the existence of these therapeutic opportunities [25–28]. This indicates the necessity of creating a proactive network connecting territorial dermatologists to secondary and tertiary centers with the aim of enhancing the therapeutic management of AD patients.

In conclusion, this study underlines the utility of organizing information campaigns on AD to enhance awareness regarding disease features and management and to facilitate early diagnosis with a subsequent reduction in the burden of disease.

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References

1. Weidinger, S.; Beck, L.A.; Bieber, T.; Kabashima, K.; Irvine, A.D. Atopic dermatitis. *Nat. Rev. Dis. Primers* **2018**, *4*, 1. [CrossRef] [PubMed]
2. Hester, J.; Kaleci, S.; Liberati, S.; Alicandro, T.; Rivi, M.; Bonzano, L.; Guanti, M.; Andreone, P.; Pellacani, G. Atopic dermatitis associated with autoimmune, cardiovascular and mental health comorbidities: A systematic review and meta-analysis. *Eur. J. Dermatol.* **2021**, *32*, 34–48. [CrossRef] [PubMed]
3. Birdi, G.; Cooke, R.; Knibb, R.C. Impact of atopic dermatitis on quality of life in adults: A systematic review and meta-analysis. *Int. J. Dermatol.* **2020**, *59*, e75–e91. [CrossRef] [PubMed]
4. Wollenberg, A.; Barbarot, S.; Bieber, T.; Christen-Zaech, S.; Deleuran, M.; Fink-Wagner, A.; Gieler, U.; Girolomoni, G.; Lau, S.; Muraro, A.; et al. Consensus-based European guidelines for treatment of atopic eczema (atopic dermatitis) in adults and children: Part I. *J. Eur. Acad. Dermatol. Venereol.* **2018**, *32*, 657–682, Published correction appears in *J. Eur. Acad. Dermatol. Venereol.* **2019**, *33*, 1436. [CrossRef]
5. Wollenberg, A.; Barbarot, S.; Bieber, T.; Christen-Zaech, S.; Deleuran, M.; Fink-Wagner, A.; Gieler, U.; Girolomoni, G.; Lau, S.; Muraro, A.; et al. Consensus-based European guidelines for treatment of atopic eczema (atopic dermatitis) in adults and children: Part II. *J. Eur. Acad. Dermatol. Venereol.* **2018**, *32*, 850–878. [CrossRef]
6. Zink, A.G.S.; Arents, B.; Fink-Wagner, A.; Seitz, I.A.; Mensing, U.; Wettemann, N.; de Carlo, G.; Ring, J. Out-of-pocket Costs for Individuals with Atopic Eczema: A Cross-sectional Study in Nine European Countries. *Acta Derm. Venereol.* **2019**, *99*, 263–267. [CrossRef]
7. Silvestre Salvador, J.F.; Romero-Pérez, D.; Encabo-Durán, B. Atopic Dermatitis in Adults: A Diagnostic Challenge. *J. Investig. Allergol. Clin. Immunol.* **2017**, *27*, 78–88. [CrossRef]
8. Katsarou, A.; Armenaka, M. Atopic dermatitis in older patients: Particular points. *J. Eur. Acad. Dermatol. Venereol.* **2011**, *25*, 12–18. [CrossRef]
9. Liu, P.; Zhao, Y.; Mu, Z.-L.; Lu, Q.-J.; Zhang, L.; Yao, X.; Zheng, M.; Tang, Y.-W.; Lu, X.-X.; Xia, X.-J.; et al. Clinical Features of Adult/Adolescent Atopic Dermatitis and Chinese Criteria for Atopic Dermatitis. *Chin. Med. J.* **2016**, *129*, 757–762. [CrossRef]
10. Kanwar, A.J.; Narang, T. Adult onset atopic dermatitis: Under-recognized or under-reported? *Indian Dermatol. Online J.* **2013**, *4*, 167–171. [CrossRef]
11. Agka, A.; Lambrou, G.I.; Matsopoulos, G.K.; Fytili, D.; Mirkopoulou, D.; Katsarou, A.; Chatziioannou, A.; Stratigos, A. Analysis of Prevalence and Risk Factors of Contact Sensitization with respect to the Occupational Profiles in a Greek Patient Cohort: A Retrospective Analysis of a Greek Referral Centre and Future Perspectives. *BioMed Res. Int.* **2021**, *2021*, 6672506.
12. Vakharia, P.P.; Chopra, R.; Silverberg, J.I. Systematic Review of Diagnostic Criteria Used in Atopic Dermatitis Randomized Controlled Trials. *Am. J. Clin. Dermatol.* **2018**, *19*, 15–22. [CrossRef]
13. Calzavara-Pinton, P.; Belloni Fortina, A.; Bonamonte, D.; RADAR Group. Diagnosis and management of moderate to severe atopic dermatitis in adolescents. A Consensus by the Italian Society of Dermatology and Venereology (SIDeMaST), the Italian Association of Hospital Dermatologists and Public Health (ADOI), the Italian Association of Hospital and Territorial Allergists and Immunologists (AAIITO), the Italian Society of Allergy, Asthma and Clinical Immunology (SIAAIC), the Italian Society of Pediatric Allergy and Immunology (SIAIP), the Italian Society of Allergological, Occupational and Environmental Dermatology (SIDAPA), and the Italian Society of Pediatric Dermatology (SIDerP). *Ital. J. Dermatol. Venerol.* **2021**, *156*, 184–197.
14. Eichenfield, L.F.; Tom, W.L.; Chamlin, S.L.; Feldman, S.R.; Hanifin, J.M.; Simpson, E.L.; Berger, T.G.; Bergman, J.N.; Cohen, D.E.; Cooper, K.D.; et al. Guidelines of care for the management of atopic dermatitis: Section 1. Diagnosis and assessment of atopic dermatitis. *J. Am. Acad. Dermatol.* **2014**, *70*, 338–351. [CrossRef]
15. Aral, M.; Arican, O.; Gul, M.; Sasmaz, S.; Kocturk, S.A.; Kastal, U.; Ekerbicer, H.C. The relationship between serum levels of total IgE, IL-18, IL-12, IFN-gamma and disease severity in children with atopic dermatitis. *Mediat. Inflamm.* **2006**, *2006*, 73098. [CrossRef]
16. Di Lorenzo, G.; Gangemi, S.; Merendino, R.A.; Minciullo, P.L.; Cannavò, S.P.; Martinelli, N.; Mansueto, P.; Rini, G.B.; Corrocher, R.; Pacor, M.L. Serum levels of soluble CD30 in adult patients affected by atopic dermatitis and its relation to age, duration of disease and Scoring Atopic Dermatitis index. *Mediat. Inflamm.* **2003**, *12*, 123–125. [CrossRef]
17. Chang, Y.S.; Chiang, B.L. Sleep disorders and atopic dermatitis: A 2-way street? *J. Allergy Clin. Immunol.* **2018**, *142*, 1033–1040. [CrossRef]

18. Reuveni, H.; Chapnick, G.; Tal, A.; Tarasiuk, A. Sleep fragmentation in children with atopic dermatitis. *Arch. Pediatr. Adolesc. Med.* **1999**, *153*, 249–253. [CrossRef]
19. Silverberg, J.I. Comorbidities and the impact of atopic dermatitis. *Ann. Allergy Asthma Immunol.* **2019**, *123*, 144–151. [CrossRef] [PubMed]
20. Murray, G.; O'Kane, M.; Watson, R.; Tobin, A.M. Psychosocial burden and out-of-pocket costs in patients with atopic dermatitis in Ireland. *Clin. Exp. Dermatol.* **2021**, *46*, 157–161. [CrossRef]
21. Ali, F.; Vyas, J.; Finlay, A.Y. Counting the Burden: Atopic Dermatitis and Health-related Quality of Life. *Acta Derm. Venereol.* **2020**, *100*, adv00161. [CrossRef] [PubMed]
22. Olsen, J.R.; Gallacher, J.; Finlay, A.Y.; Piguet, V.; Francis, N.A. Quality of life impact of childhood skin conditions measured using the Children's Dermatology Life Quality Index (CDLQI): A meta-analysis. *Br. J. Dermatol.* **2016**, *174*, 853–861. [CrossRef]
23. Chernyshov, P.V.; Sampogna, F.; Pustišek, N.; Marinovic, B.; Manolache, L.; Suru, A.; Salavastru, C.M.; Tiplica, G.S.; Stoleriu, G.; Kakourou, T.; et al. Validation of the dermatology-specific proxy instrument the Infants and Toddlers Dermatology Quality of Life. *J. Eur. Acad. Dermatol. Venereol.* **2019**, *33*, 1405–1411. [CrossRef] [PubMed]
24. Patrizi, A.; Costanzo, A.; Patruno, C.; Busà, V.M.; Chiricozzi, A.; Girolomoni, G. Unmet needs in atopic dermatitis management: An expert consensus. *J Dermatolog Treat.* **2022**, *33*, 2459–2465. [CrossRef]
25. Beck, L.A.; Thaçi, D.; Hamilton, J.D.; Graham, N.M.; Bieber, T.; Rocklin, R.; Ming, J.E.; Ren, H.; Kao, R.; Simpson, E.; et al. Dupilumab treatment in adults with moderate-to-severe atopic dermatitis. *N. Engl. J. Med.* **2014**, *371*, 130–139. [CrossRef]
26. Blauvelt, A.; de Bruin-Weller, M.; Gooderham, M.; Cather, J.C.; Weisman, J.; Pariser, D.; Simpson, E.L.; A Papp, K.; Hong, H.C.-H.; Rubel, D.; et al. Long-term management of moderate-to-severe atopic dermatitis with dupilumab and concomitant topical corticosteroids (LIBERTY AD CHRONOS): A 1-year, randomised, double-blinded, placebo-controlled, phase 3 trial. *Lancet* **2017**, *389*, 2287–2303. [CrossRef]
27. D'Erme, A.M.; Romanelli, M.; Chiricozzi, A. Spotlight on dupilumab in the treatment of atopic dermatitis: Design, development, and potential place in therapy. *Drug Des. Devel. Ther.* **2017**, *11*, 1473–1480. [CrossRef]
28. Bieber, T. Atopic dermatitis: An expanding therapeutic pipeline for a complex disease. *Nat. Rev. Drug Discov.* **2022**, *21*, 21–40. [CrossRef]

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Comorbidity Patterns in Patients with Atopic Dermatitis Using Network Analysis in the EpiChron Study

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Abstract: Background: Atopic dermatitis (AD) is associated with different comorbidities. Methods: Retrospective, observational study based on clinical information from the individuals of the EpiChron Cohort Study (Aragon, Spain) with a diagnosis of AD between 1 January 2010 and 31 December 2018. We calculated the tetrachoric correlations of each pair of comorbidities to analyze the weight of the association between them. We used a cut-off point for statistical significance of p-value < 0.01. Results: The prevalence of AD in the EpiChron Cohort was 3.83%. The most frequently found comorbidities were respiratory, cardio-metabolic, cardiovascular, and mental health disorders. Comorbidities were combined into 17 disease patterns (15 in men and 11 in women), with some sex and age specificities. An infectious respiratory pattern was the most consistently described pattern across all ages and sexes, followed by a cardiometabolic pattern that appeared in patients over 18 years of age. Conclusions: Our study revealed the presence of different clinically meaningful comorbidity patterns in patients with AD. Our results can help to identify which comorbidities deserve special attention in these types of patients and to better understand the physio-pathological mechanisms underlying the disease associations identified. Further studies are encouraged to validate the results obtained in different clinical settings and populations.

Keywords: atopic dermatitis; comorbidities; patterns; network analysis

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1. Introduction

Atopic dermatitis (AD) is a chronic inflammatory skin disease of multifactorial etiology characterized by dry skin, itching, erythema, inflammation and eczema formation [1]. It is estimated to affect 15–30% of children and 2–10% of adults [2]. Its prevalence has increased worldwide in recent years due to lifestyle and environmental changes, varying widely according to age and geographical area and being more prevalent in developed countries [2,3].

Advances in the understanding of the etiopathogenesis of AD suggest that its genesis is due to the interaction of several factors that act together to produce the onset and chronification of the disease. An alteration of the barrier function of the skin stands out, behind which there are underlying immune mechanisms, as well as genetic and environmental factors. Structural and functional abnormalities of the epidermis, together

with skin inflammation due to an altered immune response, are the cornerstones of the pathogenesis of AD [4,5].

AD is considered as the cutaneous manifestation of a systemic disorder that also gives rise to other pathologies, such as asthma, allergic rhinoconjunctivitis, etc. Some patients with AD have elevated blood levels of IgE and eosinophils. These levels are related to the severity of AD, and for this reason lower values are shown on debut [4].

The immunological mechanisms involved are being investigated; Those proposed to date include antigen-presenting cutaneous dendritic cells in the pathogenesis, and also the loss of immunosuppressive capacity of CD_4^+, CD_{25}^+ Treg cells [4].

As occurs in other diseases of the atopy spectrum, the predominance of Th2 cells over Th1 generates an immunological imbalance that aggravates the pathogenesis of AD, increases IgE and activates interleukins.

On the other hand, the importance of the integrity of the skin barrier should be highlighted; in recent decades, its dysfunction has been determined to be essential in the pathogenesis of AD. The structure of the skin barrier is complex. The most superficial layer of the epidermis is the stratum corneum, it is made up of proteins (filaggrin, loricin, involucrin); and by a layer of lipids composed of long-chain ceramides as the main component. The stratum corneum protects against environmental stimuli such as allergens, irritants, chemical and physical changes and infections, it also prevents trans-epidermal dehydration [5].

Family genetic studies have shown that AD is a hereditary disease. At the moment, the evidence points to chromosome 1q21 where the locus of the epidermal differentiation complex is located [4,5]. Of all the components of the skin barrier, filaggrin and its mutations are the ones that have shown the greatest association with AD. Filaggrin is a protein that interacts with intermediate filaments, producing their aggregation into macrofibrils. Filaggrin defects can lead to dysfunctions in the skin barrier, resulting in inferior protection against bacteria and allergens [5].

It is currently recognized that this disease is found, especially in children and adolescents, within the context of atopy, an entity that also includes asthma, rhino-conjunctivitis, and a significant predisposition to develop allergies [6,7]. Several studies have previously analyzed the association of AD with the presence of other diseases. A systematic review and meta-analysis by Chester et al. in 2021 [8] concluded that AD patients present an increased risk of mental and autoimmune diseases. The narrative review by Paller et al. in 2018 [9] showed that the global burden of AD is associated with mental illnesses such as depression, anxiety, and suicidal ideation, as a result of lack of sleep, itching, and stigmatization due to their skin lesions, both in children and adults. The cohort study by Mortz et al. in 2015 [10], on the other hand, showed that AD is associated with infections, neuropsychiatric disorders, metabolic syndrome, autoimmune diseases, and cancer, among others. Furthermore, the cross-sectional study by Gilaberte et al. in 2020 [11] showed that 43% of children under 18 years of age with AD in Spain have at least one additional comorbidity. The most frequent comorbidities in this study were asthma, psychosocial disorders, and visual disturbances, whereas asthma, allergic rhinitis, and irritable bowel syndrome showed the greatest strength of association with AD.

A better knowledge of the comorbidities surrounding AD could help us guide the care of these patients from a holistic perspective and better understand the etiopathogenesis of this disease. However, chronic diseases rarely appear in isolation and tend to cluster together in the form of disease patterns, which represent non-random associations among diseases. Their study could allow us to identify profiles of AD patients with specific care needs and specific preventive actions, and could also shed some light on the underlying physio-pathological mechanisms.

In this context, network science is a powerful tool that applies clustering techniques that allow us to exhaustively analyze and visualize the associations between diseases to identify disease patterns [12]. Network analysis has already been applied to study the associations among diseases in patients with specific index conditions with relevant clinical

results [13,14]. However, to our knowledge, this research approach has not been applied to the study of AD comorbidity.

This study aims to explore the existence of comorbidity patterns in patients with AD using network analysis and to clinically describe the clusters obtained.

2. Materials and Methods

We conducted a retrospective, observational study in the EpiChron Cohort, which links socio-demographic and clinical data from all the users of the public health system of the Spanish region of Aragón [15]. This cohort is based on the information registered in the electronic health records (EHRs) and clinical–administrative databases of approximately 98% of the citizens of the region (reference population: 1.3 million people). For this study, we selected all the 50,801 individuals from the cohort diagnosed with AD at some point from 1 January 2010 to 31 December 2018.

The Clinical Research Ethics Committee of Aragón (CEICA) approved this study (Research protocol PI20/633) and waived the requirement to obtain informed consent from patients given the epidemiological nature of the project and the use of anonymized data.

For all patients, we studied sex, age interval (0–2, 3–10, 11–17, 18–65, and >65 years), and all chronic diseases registered in their EHRs. Diagnoses were initially coded using the International Classification of Primary Care, First Edition (ICPC–1), or the International Classification of Diseases, Ninth Revision, Clinical Modification (ICD–9–CM). Subsequently, using the open-source algorithm Chronic Condition Indicator (CCI) [16], each ICD9 code was classified as either chronic or not. The software defines "chronic" as diseases with a duration equal to or greater than 12 months and meeting at least one of the following criteria: (a) require continuous care, that have a high risk of recurrence, and/or that continue to have implications for the management of the patient; (b) imply limitations on self-care, social interactions, and independent living. Once selected, those chronic diagnoses were grouped in 153 clinical categories through the Clinical Classifications Software (CCS) [17] based on the clinical, therapeutic and diagnostic similarities of the diseases.

First, a descriptive analysis of the demographic characteristics of the study population was performed. We summarized the results as proportions for categorical variables and as means and standard deviations for continuous variables.

Then, we performed a network analysis to study the associations between comorbidities of AD. We stratified the population by sex and age interval and built a network for each stratum, with ten networks in total. To increase the clinical interest of the study and to facilitate the interpretation of the results, only diseases with a prevalence > 1% were included in the analysis.

In a disease network, a node represents a disease, and an edge means a statistically significant correlation between a specific pair of conditions. We calculated the tetrachoric correlations of each pair of comorbidities to analyze the weight of the association between them [18]. We used a cut-off point for statistical significance of p-value < 0.01 to correct the family-wise error rate due to multiple comparisons [13,14].

Then, we used the network's modularity to search for clusters of diseases within each network based on the Louvain method [19], as previous disease pattern studies have done [13,14,20]. Modularity analyzes the number of edges in the network, comparing the density of edges inside a group to edges between groups [19]. The Louvain method optimizes the modularity in an iterated process, detecting communities or clusters of diseases. Community detection methods, such as the Louvain or Leiden algorithms, among others, allow the network's structure to determine the number and size of the cluster obtained [21,22] based on the density of edges and their weight (measured by the tetrachoric correlation) and not by the researcher.

Once we obtained the patterns of diseases for each age and sex stratum, all clinicians named the patterns by consensus. This last step was performed considering the prevalence and clinical relevance of the diseases, and the weight of the tetrachoric correlations, in line with the names already given in the literature.

We performed all the analysis in RStudio software (version 1.4.1106, Rstudio, Boston, MA, USA) and GEPHI software (version 0.9.2).

3. Results

3.1. Characteristics of the Population

We analyzed a population of 50,801 patients with AD (46.3% men). The demographic characteristics are shown in Table 1. The overall prevalence of AD in Aragon was 3.83%.

Table 1. Demographic characteristics of patients with AD.

Characteristics	Total (N = 50,801)	Men (N = 23,522)	Women (N = 27,279)
Age interval (N, %)			
0–2 years	3005 (5.9%)	1605 (6.8%)	1400 (5.1%)
3–10 years	17,797 (35.0%)	8954 (38.1%)	8843 (32.4%)
11–17 years	10,955 (21.6%)	54,318 (22.6%)	5637 (20.7%)
18–65 years	14,707 (29.0%)	5985 (25.4%)	8722 (32.0%)
>65 years	4337 (8.5%)	1660 (7.1%)	2677 (9.8%)
Area of Residence (N, %)			
Urban	31,650 (62.3%)	14,627 (62.2%)	17,023 (62.4%)
Rural	19,151 (37.7%)	8895 (37.8%)	10,256 (37.6%)

The most prevalent diseases found in patients with AD were respiratory (i.e., upper respiratory infections, asthma, and rhinitis), cardio-metabolic (i.e., hypertension, dyslipidemia and obesity), cardiovascular (i.e., cardiac dysrhythmia and coagulation disorders), and mental health diseases (i.e., anxiety and mood disorders). Diseases were combined into seventeen patterns with some sex and age specificities, which are summarized below. The complete output of the analysis is available as Supplementary Material in which we detailed the complete pattern analysis.

3.2. Comorbidity Patterns in Men

We identified fifteen patterns in men, classified as upper respiratory infections, respiratory, otorhinolaryngological (ORL), upper respiratory infections-ORL, congenital anomalies-mental, respiratory-allergic, sensitive-digestive, sensitive-metabolic, headache-mental, cardiometabolic, mental, cardiovascular, dyslipidemic, and geriatric. Their composition, disease prevalence, and correlation between conditions are described below and in Figure 1.

In children aged 0–2, we found three patterns. The *upper respiratory infections* pattern included diseases such as upper respiratory infections, which was the most prevalent condition in this network, esophageal disorders, and congenital anomalies. We found a *respiratory* pattern that had asthma and other upper respiratory diseases, among others. An *ORL* pattern was also described with otitis as its most prevalent disease.

Four patterns in boys aged 3–10 years were identified. One that combined most of the diseases from upper respiratory infections and ORL diseases in children aged 0–2. We found a *respiratory-allergic* pattern that included asthma, other upper respiratory diseases, and rhinitis. The correlation between these two last conditions was almost perfect, with a strength of the correlation of 0.99 out of 1. The *congenital anomalies-mental* pattern associated congenital anomalies, miscellaneous mental health disorders, and other skin inflammatory conditions. The last pattern found was a *sensitive-digestive* pattern which included blindness as the most prevalent condition.

Four other patterns were identified in boys aged 11–17. The *upper respiratory infections-ORL* and *respiratory-allergic* were similar to the previous in children aged 3–10. The *headache-mental* pattern included headache as the most prevalent condition; it also included anxiety and miscellaneous mental health disorders, among other diseases. The *sensitive-metabolic* pattern had diseases such as blindness and vision defects, thyroid disorders, obesity, and other nutritional/endocrine/metabolic diseases.

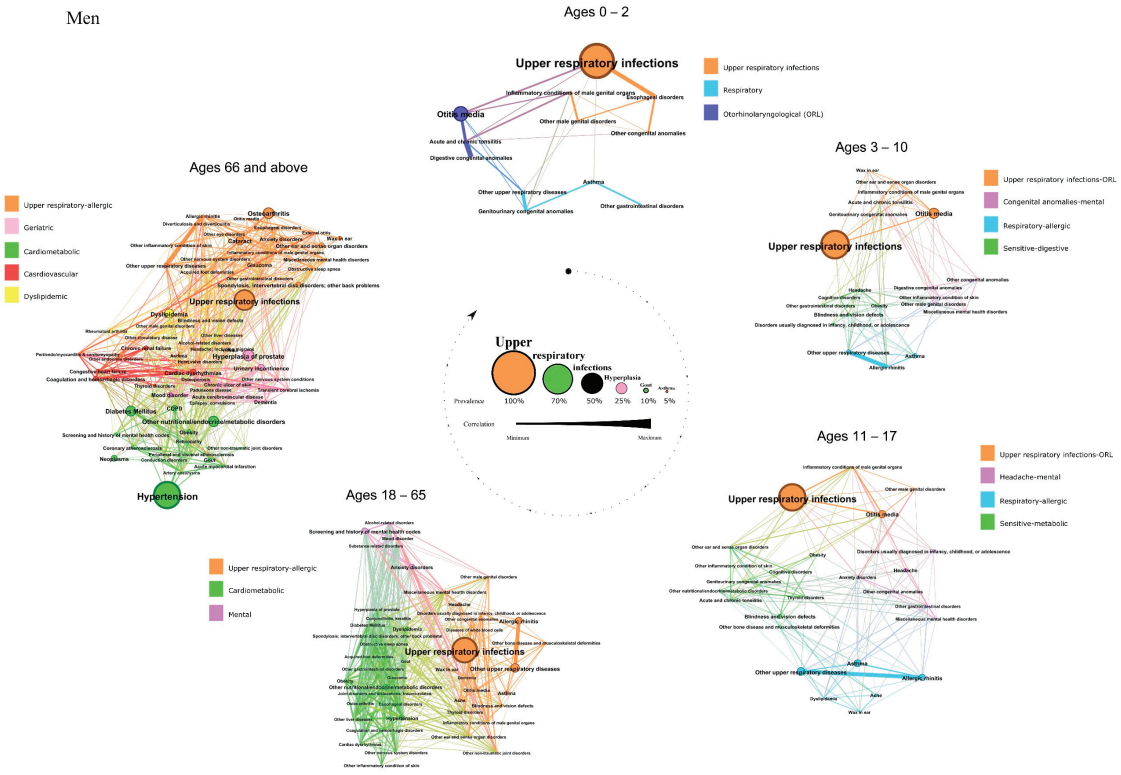

Figure 1. Comorbidity patterns in the networks of men with AD based on age. The diameter of each node and the label size are proportional to the disease prevalence. The width of each link is proportional to the correlation between disease. The colors of the nodes correspond to different patterns.

In men aged 18–65 years, three patterns were identified. One pattern combined most of the diseases from the *upper respiratory-ORL* pattern and the *respiratory-allergic* pattern from boys aged 11–17. A *cardiometabolic* pattern was the cluster with more diseases included, highlighting hypertension, obesity, other nutritional/endocrine/metabolic disorders, dyslipidemia, other inflammatory conditions of the skin, and diabetes. We also identified a *mental* pattern which included screening and history of mental health codes, anxiety, mood disorders, substance-related, and alcohol-related disorders.

In men aged 66 and older, five patterns were detected. The *upper-respiratory-allergic* pattern was similar to that found in men aged 18–65, but included other highly prevalent diseases such as osteoarthritis or cataracts. We found a *cardiometabolic* pattern with hypertension, diabetes, other nutritional/endocrine/metabolic disorders, obesity, COPD, and neoplasms as the most prevalent diseases. We identified a *cardiovascular* pattern that included cardiac dysrhythmias as its most prevalent condition. A *geriatric* pattern, with hyperplasia of the prostate, urinary incontinence, or dementia, among other diseases, was also identified. The last and less specific pattern described included dyslipidemia as its most prevalent disease.

3.3. Comorbidity Patterns in Women

We identified eleven patterns in women, which were referred as upper respiratory infections, ORL, upper respiratory infections-ORL, respiratory-allergic, sensitive-digestive, menstrual-dysphoric-metabolic, sensitive, upper respiratory-allergic, cardiometabolic, car-

diovascular, and geriatric. Their composition, disease prevalence, and correlation between conditions are described below and in Figure 2.

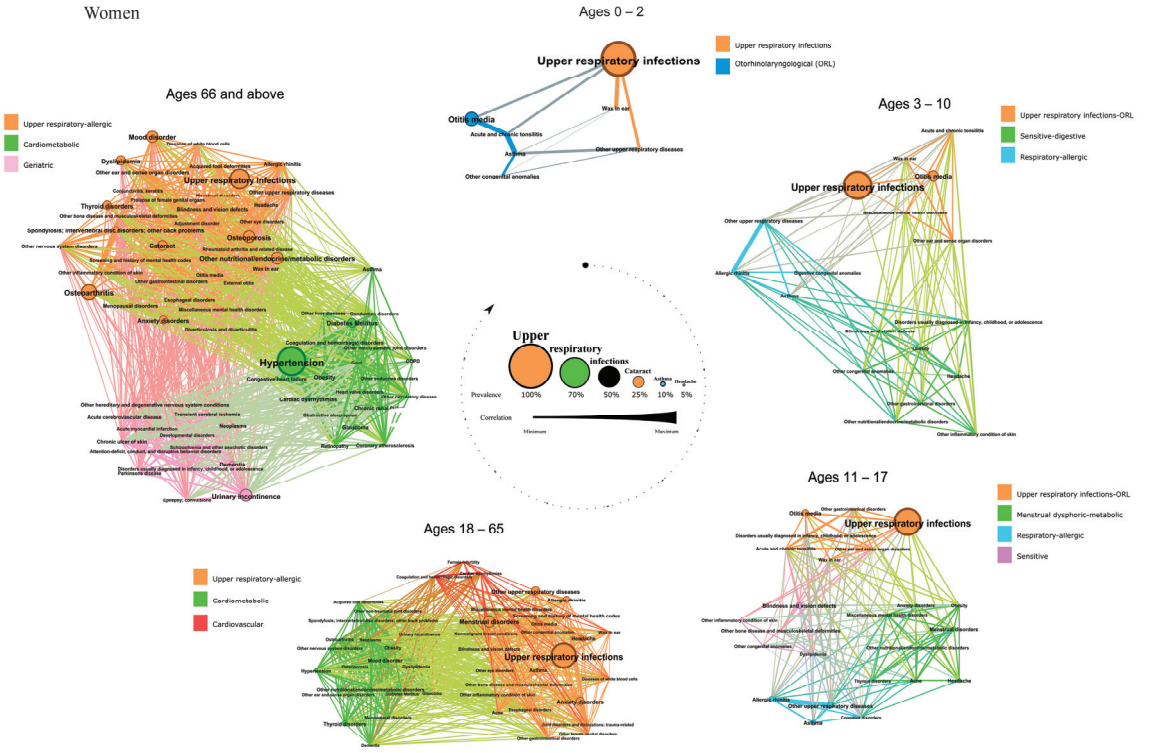

Figure 2. Comorbidity patterns in the networks of women with AD based on age. The diameter of each node and the label size are proportional to the disease prevalence. The width of each link is proportional to the correlation between disease. The colors of the nodes correspond to different patterns.

In girls aged 0–2, the two patterns found were similar to those found in boys aged 0–2 years: *upper respiratory infections* and *ORL*.

We described three patterns in girls aged 3–10, again very similar to the clusters found in boys at the same age: an *upper respiratory infections-ORL* pattern, a *respiratory-allergic* pattern, and a *sensitive-digestive* pattern.

Four other patterns were identified in girls aged 11–17: an *upper respiratory infections-ORL* pattern; a *respiratory-allergic* pattern; a *menstrual dysphoric-metabolic* pattern that included menstrual disorders as the most prevalent condition, but also anxiety, miscellaneous mental health disorders, obesity, thyroid and other metabolic disorders; and finally, the *sensitive* pattern, which included blindness as its most prevalent disease.

In women aged 18–65 years, we found three patterns. The *upper respiratory-allergic* pattern included respiratory diseases such as rhinitis, other upper respiratory diseases, and asthma, but also menstrual disorders and anxiety, among others. A *cardiometabolic* pattern was found, that was mainly characterized by thyroid diseases, obesity, and hypertension but also mood disorders. We also identified *cardiovascular* a pattern that only included three conditions: coagulation and hemorrhagic disorders, cardiac dysrhythmias, and female infertility.

In women aged 66 and older, three patterns were detected. The *upper respiratory-allergic* pattern also included osteoarthritis, osteoporosis, thyroid, and mood disorders, among

other diseases. The *cardiometabolic* pattern was found, characterized by hypertension, but also including diabetes mellitus, heart failure, COPD and obstructive sleep apnea. Finally, a *geriatric* pattern that included urinary incontinence, dementia, neoplasms, and chronic skin ulcer as the most prevalent diseases was found.

4. Discussion

This study explored the comorbidity patterns of AD through the analysis and visualization of the existing disease networks. Different clusters defined as upper respiratory infections with ORL diseases, respiratory disorders with allergic conditions, and cardiovascular diseases with metabolic disorders, among others, were identified depending on age and gender. These epidemiological findings can be helpful to guide AD patients in the primary, secondary, or even tertiary prevention of their comorbidities and understand their physio-pathological mechanisms.

The present investigation shows the infectious respiratory pattern as the most consistently described pattern across all age groups and sexes. Its main component was respiratory infectious diseases, but its weight decreased in older groups in favor of diseases with an allergic component. Asthma or allergic rhinitis are some of the disorders that have been added. In the youngest groups, this allergic component has a distinct pattern by itself. In the case of children aged 0–2 years, respiratory infections are associated with genital, esophageal, or other malformations, although this is not the case in girls.

The higher incidence of infections in patients with AD has been widely described. Dysfunction of the epithelial barrier, colonization of the skin by *Staphylococcus aureus*, and the use of immunosuppressive drugs are some of their causes. In this context, the Swiss BAMSE cohort revealed a higher incidence of pneumonia, otitis media and antibiotics use in AD patients aged 0–2 years [23]. This fact is consistent with the patterns described. Although patients with AD are colonized by *S. aureus* in up to 70% of the cases and are more likely than the general population to suffer impetigo, herpetic eczema or molluscum contagiosum [24], in our analysis skin infections did not play a relevant role or were associated with extracutaneous infections.

Cardiometabolic diseases have been associated with AD in various epidemiological studies, although this association is less clear than in other diseases such as psoriasis [8,25,26]. Multifactorial etiology has been used to explain this association: insomnia, obesity, diabetes and smoking, among other variables [27,28]. Our study found a pattern of cardiometabolic comorbidities that included hypertension, obesity, and mood and thyroid disorders, among others. This pattern was common in men and women over 18 years of age, although there were differences, including mood and endocrine disorders occurring more frequently in women. As for chronic obstructive pulmonary disease, it was more frequent in the group older than 65 years old for both sexes. The existence of this pattern confirms that patients with AD present comorbidities that are cardiovascular risk factors and that tend to be associated throughout life.

Anxiety, insomnia, and mood disorders, among other mental health problems, are comorbidities with a higher incidence in patients with AD. Recent studies have shown that the earlier the disease appears, the greater the risk of suffering from psychiatric comorbidities is [11,24,29]. We found a pattern of mental comorbidities that appeared in boys older than two years and is maintained up to 65 years of age. In the case of girls, this pattern appears intermingled with a menstrual disease first and metabolic comorbidities that we call menstrual-dysphoric-metabolic pattern. The different ways of interacting between the sexes with the environment and the interpersonal relationships they establish at an early age could explain this phenomenon [30].

In patients over 65 years of age, a geriatric pattern was found in both sexes. This pattern grouped diseases such as urinary incontinence, Parkinson's disease, dementia, skin ulcers and neoplasms, among others. The association of these diseases, typical of physiological aging, with AD is complex. An increase in neoplasms has been described in patients with AD, with lymphomas being the most strongly associated [9]. Regarding the

rest of diseases of this pattern, to our knowledge no studies support a higher incidence of these diseases in patients with AD.

Regarding the limitations of this study, the fact that the clinical information obtained in the EHRs was not originally designed for research could create over- and under-diagnosis of some chronic disorders. Another limitation is the cross-sectional retrospective nature of the study, which does not allow us to know the longitudinal characteristics of the population. Additionally, we have to consider the lack of some variables that could help us explain the results obtained, such as lifestyle information, socioeconomic factors, information on functional status, and analytical variables, among others.

One of the principal strengths of our research is that it was conducted on a population-based cohort, including 98% of the reference population. Moreover, data in the EpiChron Cohort undergo continuous quality control checkups that ensure their accuracy and reliability for research purposes. Another important strength is the innovative method applied to understand comorbidities in AD. Network analysis studies the interrelations between diseases and how patterns emerge from them. This paper shows the potentiality of applying this method to study and visualize the comorbidities of AD and achieve a more holistic understanding of these patients. In this sense, it is also important to highlight that this study exhaustively analyzed all chronic diseases obtained from the patient's EHRs created by health professionals, and not just the most relevant, prevalent or self-reported diseases.

5. Conclusions

We identified similar disease patterns in men and women with AD, with the number and complexity of such patterns increasing with age. This is the first study to analyze the comorbidity patterns of AD patients, and our results can help to guide caregivers of AD patients in the prevention of their comorbidities and to understand the physiopathological mechanisms underlying the comorbidity patterns identified. This study opens an innovative approach to analyze and help AD patients, although further studies are needed to validate the results obtained in different clinical settings and populations.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/jcm11216413/s1.

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Data Availability Statement: The data used in this study cannot be publicly shared, because of restrictions imposed by the Aragon Health Sciences Institute (IACS) and asserted by the Clinical Research Ethics Committee of Aragon (CEICA, ceica@aragon.es). The authors can establish future collaborations with other groups based on the same data. Potential collaborations should be addressed to the Principal Investigator of the EpiChron Group, Alexandra Prados-Torres, sprados.iacs@aragon.es.

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References

1. Barbarot, S.; Aubert, H.; Bernier, C.; Stalder, J.-F. Dermatitis Atópica. *EMC-Dermatol.* **2016**, *50*, 317–329. [CrossRef]
2. Bagazgoitia, L.; Gutiérrez, M.; García Blesa, C.; Martín, A.H.; Torrelo, A. Aspectos epidemiológicos, patogénicos, clínicos y diagnósticos de la dermatitis atópica. ¿Es posible la prevención? *Rev. Pediatría De Atención Primaria* **2009**, *11* (Suppl. 15), s31–s37. [CrossRef]
3. Aragonés, A.M.; Toledo, R.F.; Calatayud, A.M.; Mir, J.C.C. Epidemiologic, Clinical and Socioeconomic Factors of Atopic Dermatitis in Spain: Alergológica-2005. *J. Investig. Allergol. Clin. Immunol.* **2009**, *19*, 27–33.
4. Darlenski, R.; Kazandjieva, J.; Hristakieva, E.; Fluhr, J.W. Atopic Dermatitis as a Systemic Disease. *Clin. Dermatol.* **2014**, *32*, 409–413. [CrossRef] [PubMed]
5. Ricardo Alonso, O.E.; Rodríguez Sánchez, M.B.; Hernández Fernández, M.; Alonso González, M. Aspectos de Interés Sobre La Etiopatogenia de La Dermatitis Atópica TT–Interesting Aspects about the Etiopathogenesis of Atopic Dermatitis. *Rev. Médica. Electrónica.* **2018**, *40*, 1139–1148.
6. Silverberg, J.I.; Silverberg, N.B. Childhood Atopic Dermatitis and Warts Are Associated with Increased Risk of Infection: A US Population-Based Study. *J. Allergy Clin. Immunol.* **2014**, *133*, 1041–1047. [CrossRef]
7. Brunner, P.M.; Silverberg, J.I.; Guttman-Yassky, E.; Paller, A.S.; Kabashima, K.; Amagai, M.; Luger, T.A.; Deleuran, M.; Werfel, T.; Eyerich, K.; et al. Increasing comorbidities suggest that atopic dermatitis is a systemic disorder. *J. Investig. Dermatol.* **2017**, *137*, 18–25. [CrossRef]
8. Chester, J.; Kaleci, S.; Liberati, S.; Alicandro, T.; Rivi, M.; Bonzano, L.; Guanti, M.; Andreone, P.; Pellacani, G. Atopic Dermatitis Associated with Autoimmune, Cardiovascular and Mental Health Comorbidities: A Systematic Review and Meta-Analysis. *Eur. J. Dermatol.* **2022**, *32*, 34–48. [CrossRef]
9. Paller, A.; Jaworski, J.C.; Simpson, E.L.; Boguniewicz, M.; Russell, J.J.; Block, J.K.; Tofte, S.; Dunn, J.D.; Feldman, S.R.; Clark, A.R.; et al. Major Comorbidities of Atopic Dermatitis: Beyond Allergic Disorders. *Am. J. Clin. Dermatol.* **2018**, *19*, 821–838. [CrossRef]
10. Mortz, C.G.; Andersen, K.E.; Dellgren, C.; Barington, T.; Bindslev-Jensen, C. Atopic Dermatitis from Adolescence to Adulthood in the TOACS Cohort: Prevalence, Persistence and Comorbidities. *Allergy Eur. J. Allergy Clin. Immunol.* **2015**, *70*, 836–845. [CrossRef]
11. Gilaberte, Y.; Pérez-Gilaberte, J.B.; Poblador-Plou, B.; Bliek-Bueno, K.; Gimeno-Miguel, A.; Prados-Torres, A. Prevalence and Comorbidity of Atopic Dermatitis in Children: A Large-Scale Population Study Based on Real-World Data. *J. Clin. Med.* **2020**, *9*, 1632. [CrossRef]
12. Barabasi, A.L. Network Science. Available online: http://networksciencebook.com/ (accessed on 10 October 2022).
13. Divo, M.J.; Casanova, C.; Marin, J.M.; Pinto-Plata, V.M.; de-Torres, J.P.; Zulueta, J.J.; Cabrera, C.; Zagaceta, J.; Sanchez-Salcedo, P.; Berto, J.; et al. COPD Comorbidities Network. *Eur. Respir. J.* **2015**, *46*, 640–650. [CrossRef]
14. Carmona-Pírez, J.; Poblador-Plou, B.; Díez-Manglano, J.; Morillo-Jiménez, M.J.; Marín Trigo, J.M.; Ioakeim-Skoufa, I.; Gimeno-Miguel, A.; Prados-Torres, A. Multimorbidity Networks of Chronic Obstructive Pulmonary Disease and Heart Failure in Men and Women: Evidence from the EpiChron Cohort. *Mech. Ageing Dev.* **2021**, *193*, 111392. [CrossRef]
15. Prados-Torres, A.; Poblador-Plou, B.; Gimeno-Miguel, A.; Calderón-Larrañaga, A.; Poncel-Falcó, A.; Gimeno-Feliú, L.A.; González-Rubio, F.; Laguna-Berna, C.; Marta-Moreno, J.; Clerencia-Sierra, M.; et al. Cohort Profile: The Epidemiology of Chronic Diseases and Multimorbidity. The EpiChron Cohort Study. *Int. J. Epidemiol.* **2018**, *47*, 382e–384e. [CrossRef]
16. Chronic Condition Indicator (CCI) for ICD-9-CM. Available online: https://www.hcup-us.ahrq.gov/toolssoftware/chronic/chronic.jsp (accessed on 1 October 2021).
17. Elixhauser, A.; Steiner, C.; Palmer, L. Clinical Classifications Software (CCS). 2009. Available online: http://www.hcup-us.ahrq.gov/toolssoftware/ccs/ccs.jsp (accessed on 10 October 2022).
18. Kubinger, K. On Artificial Results Due to Using Factor Analysis for Dichotomous Variables. *Psychol. Sci.* **2003**, *45*, 106–110.
19. Blondel, V.D.; Guillaume, J.-L.; Lambiotte, R.; Lefebvre, E. Fast Unfolding of Communities in Large Networks. *J. Stat. Mech. Theory Exp.* **2008**, *2008*, 10008. [CrossRef]
20. Divo, M.J.; Celli, B.R.; Poblador-Plou, B.; Calderón-Larrañaga, A.; de-Torres, J.P.; Gimeno-Feliu, L.A.; Bertó, J.; Zulueta, J.J.; Casanova, C.; Pinto-Plata, V.M.; et al. Chronic Obstructive Pulmonary Disease (COPD) as a Disease of Early Aging: Evidence from the EpiChron Cohort. *PLoS ONE* **2018**, *13*, e0193143. [CrossRef]
21. Lancichinetti, A.; Fortunato, S. Community Detection Algorithms: A Comparative Analysis. *Phys. Rev. E* **2009**, *80*, 56117. [CrossRef]
22. Newman, M.E.J. Modularity and Community Structure in Networks. *Proc. Natl. Acad. Sci. USA* **2006**, *103*, 8577–8582. [CrossRef]
23. Böhme, M.; Lannerö, E.; Wickman, M.; Nordvall, S.L.; Wahlgren, C.F. Atopic Dermatitis and Concomitant Disease Patterns in Children up to Two Years of Age. *Acta Derm. Venereol.* **2002**, *82*, 98–103. [CrossRef]
24. Silverberg, J.I. Comorbidities and the Impact of Atopic Dermatitis. *Ann. Allergy Asthma Immunol.* **2019**, *123*, 144–151. [CrossRef] [PubMed]
25. Jung, H.J.; Lee, D.H.; Park, M.Y.; Ahn, J. Cardiovascular Comorbidities of Atopic Dermatitis: Using National Health Insurance Data in Korea. *Allergy Asthma Clin. Immunol.* **2021**, *17*, 94. [CrossRef] [PubMed]
26. Silverberg, J.I. Association between Adult Atopic Dermatitis, Cardiovascular Disease, and Increased Heart Attacks in Three Population-Based Studies. *Allergy* **2015**, *70*, 1300–1308. [CrossRef]

27. Ascott, A.; Mulick, A.; Yu, A.M.; Prieto-Merino, D.; Schmidt, M.; Abuabara, K.; Smeeth, L.; Roberts, A.; Langan, S.M. Atopic Eczema and Major Cardiovascular Outcomes: A Systematic Review and Meta-Analysis of Population-Based Studies. *J. Allergy Clin. Immunol.* **2019**, *143*, 1821–1829. [CrossRef] [PubMed]
28. Yin, J.; Jin, X.; Shan, Z.; Li, S.; Huang, H.; Li, P.; Peng, X.; Peng, Z.; Yu, K.; Bao, W.; et al. Relationship of Sleep Duration With All-Cause Mortality and Cardiovascular Events: A Systematic Review and Dose-Response Meta-Analysis of Prospective Cohort Studies. *J. Am. Heart Assoc.* **2022**, *6*, e005947. [CrossRef]
29. Cheng, B.T.; Silverberg, J.I. Depression and Psychological Distress in US Adults with Atopic Dermatitis. *Ann. Allergy Asthma Immunol.* **2019**, *123*, 179–185. [CrossRef]
30. Faravelli, C.; Alessandra Scarpato, M.; Castellini, G.; Lo Sauro, C. Gender Differences in Depression and Anxiety: The Role of Age. *Psychiatry Res.* **2013**, *210*, 1301–1303. [CrossRef]

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Case Report

Treatment of Severe Atopic Dermatitis with Dupilumab in Patients with Advanced Cancer

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Abstract: Atopic dermatitis is a chronic inflammatory intensively pruritic skin disease. Patients with moderate-to-severe atopic dermatitis or with difficult-to-treat areas are candidates for systemic therapy, especially when topical therapy is inadequate. Currently, we have available not only conventional immunosuppressive systemic therapy, but also targeted biological therapy, which has shown a remarkable reduction in clinical severity with a good safety profile. Dupilumab has been approved to treat moderate-to-severe atopic dermatitis. Even though the therapy has been available for more than 3 years, there are still limited data regarding the treatment of patients with concomitant cancer. Previous immunosuppressive treatment for atopic dermatitis, such as cyclosporine or azathioprine, poses a safety risk for patients with malignant disease. We present a case series of three patients with advanced cancer and severe atopic dermatitis treated with dupilumab for an average of 17 months with a great response toward atopic dermatitis without cancer recurrence. One patient had colorectal cancer' the second and the third both had cancer duplicity—colorectal and kidney cancer and penile squamous cell carcinoma with prostate cancer. Our cases suggest that dupilumab can safely control atopic dermatitis in patients with advanced cancer.

Keywords: atopic dermatitis; dupilumab; cancer; real-world study; biological therapy

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1. Introduction

Atopic dermatitis (AD) is one of the most common chronic inflammatory skin diseases of a noninfectious nature, which, although not life-threatening, has a significant negative impact on the patient's quality of life. The prevalence of AD has doubled to tripled in industrialized countries since the 1970s, with approximately 15% to 20% of children (of whom up to 30% have moderate-to-severe AD) and 2% to 4% of adults (of whom up to 46% have moderate-to-severe AD) affected worldwide [1,2].

Although immune dysregulation and skin barrier defects are accepted as key components in the development of the disease, the pathophysiology remains unclear despite ongoing scientific research. While previous studies have largely focused on immune dysregulation of T helper cell type 1 (Th1) and Th2 groups as the pathogenesis of the disease in genetically predisposed individuals, skin barrier disruption and systemic inflammation are the focus of current AD research [3]. In AD, disruption of the skin barrier results in increased transepidermal water loss, decreased skin hydration, and increased presentation of antigens by Langerhans cells initiating inflammation [3].

However, two hypotheses are now known regarding the origin of inflammation that leads to the triggering of atopic dermatitis [4].

The first hypothesis is primarily an immune dysfunction (increased expression of Th2 lymphocytes and, thus, production of interleukins IL-4 and IL-13), resulting in IgE sensitization, allergic inflammation, and a secondary role of impaired epithelial function [4].

The second hypothesis is that a primary role in the development of atopic dermatitis is played by impaired epithelial function of the skin, leading to immunological dysregulation

and subsequent inflammation. Genetic and environmental factors are also involved in the expression of atopic dermatitis [4].

As a result of these multiple factors, atopic dermatitis exhibits significant heterogeneity in disease phenotype, age of onset, clinical severity, persistence, comorbidity, and response to treatment. The clinical picture of atopic dermatitis, as well as the course of the disease itself, is diverse and unpredictable. The basic characteristic triad consists of persistent pruritus, dermatitis, and xerosis. Pruritus is a key and dominant feature of AD [5]. It generates comorbidities such as sleep loss and psychological distress, creating a continuous burden for patients, parents, and other family members [6]. Atopic dermatitis has a negative impact on patients' normal daily activities [7] and their work productivity [8], and it can be accompanied by many comorbidities such as psychiatric diseases, ophthalmological disability, cardiovascular disease, anxiety, depression, and different autoimmune diseases [6].

The existing evidence on cancer risk in atopic dermatitis is inconsistent, with studies limited by insufficient consideration of severity of atopic dermatitis and its treatment. A recent population-based cohort study by Wan et al. reported a reduction in the incidence of certain solid tumors (e.g., breast cancer) and an increased risk of lymphoma in patients with atopic dermatitis [9].

Furthermore, atopic dermatitis is also associated with significant direct and indirect financial costs, which correlate with disease severity [10].

The goal of therapy is to achieve long-term remission with minimal side-effects and to improve patients' quality of life. Options of topical therapy are limited to corticosteroids and topical immunomodulators [11]. Topical therapy is the mainstay of AD treatment and can be used in monotherapy for mild cases. In patients with severe atopic dermatitis, it serves as an adjunct to systemic therapy.

Systemic therapy, including biological, is indicated for patient with moderate-to-severe forms of atopic dermatitis. Among the conventional immunosuppressive drugs, cyclosporine is the drug of first choice in adult patients as it is the only on-label conventional systemic treatment in Europe [11,12]. Other immunosuppressants can also be used off-label, such as systemic corticosteroids, azathioprine, mycophenolate mofetil, methotrexate, or phototherapy [11]. Although these traditional immunosuppressive therapies can be effective in atopic dermatitis, their routine use is often limited due to adverse effects, frequent laboratory monitoring, and inappropriateness of long-term usage [12]. Furthermore, it is always necessary to consider comorbidities and contraindications, as well as possible secondary infections, and to exclude malignancy due to the immunosuppressive effect. With systemic therapy, it is important to carefully consider the risks and benefits of the selected drug. The newest treatment option for patients with atopic dermatitis is targeted therapy, the development of which has been made possible by a closer understanding of the pathogenesis of AD at the molecular level. Knowledge of the immunological background of AD has led to the synthesis and testing of monoclonal antibodies targeting these cytokines, with the first approved drug being dupilumab [12,13]. The second group of targeted therapies for AD constitutes small molecules whose mechanism of action is JAK/STAT inhibition [14].

Dupilumab is a fully human monoclonal antibody against the α chain of the interleukin (IL) 4 receptor; it blocks IL-4 and IL-13 signaling pathways and has shown a remarkable reduction in clinical severity with a good safety profile in both clinical trials and real-world studies [13,15–18]. IL-4 and IL-13 are key Th2 cytokines that play a critical role in the pathophysiology of allergic diseases and, thus, in the development of atopic eczema [4].

Dupilumab has been reimbursed in the Czech Republic since 1 June 2019 for adult patients with severe AD after failure or ineffectiveness of at least one conventional systemic immunosuppressive therapy (except systemic treatment with corticosteroids) or for patients in whom the use of conventional systemic therapy is contraindicated. Dupilumab is also reimbursed for patients aged 6–18 years after failure of maximal topical therapy and phototherapy or balneotherapy

No specific laboratory or imaging tests are required before or during therapy. Treatment of pre-existing infections is recommended [19]. The most common side-effects are application site reactions, conjunctivitis, blepharitis, and herpes simplex infection. Dupilumab can be combined with topical corticosteroids or topical immunomodulators.

Furthermore, the FDA has approved dupilumab for the treatment of bronchial asthma, chronic sinusitis with nasal polyposis, eosinophilic esophagitis, and prurigo nodularis [19].

Long-term safe therapy is the only effective way to prevent recurrence and exacerbation of AD.

Patients with a history of malignancy were generally excluded from dupilumab clinical trials [13,16]. Current conventional immunosuppressive treatment for AD such as cyclosporine or azathioprine is not suitable for patients with a history of cancer. Up to now, there are no data reporting that modulation of the IL-4 and IL-13 signaling pathways can increase the risk of malignancy [20].

Although interventional clinical trials have demonstrated the efficacy and safety of dupilumab, they may not accurately correspond to "real-life" practice, given that clinical trials often do not include patients with comorbidities (e.g., decompensated arterial hypertension, severe eye diseases, and patients with previous oncological disease), which we commonly see in clinical practice.

Only a few cases have been reported of patients with advanced cancer being treated with dupilumab [21,22]. Here, we present three patients with atopic dermatitis and previous malignancy treated with dupilumab, with good clinical response on AD and no evidence of cancer recurrence.

2. Case Series

Case 1. Our patient is a 59 year old nonsmoker female suffering from atopic dermatitis since the age of 40, which first appeared after significant psychological stress. The manifestations of atopic dermatitis were initially mild and then gradually proceeded to a generalized phenotype. She was treated with UVB phototherapy (311 nm) which triggered an exacerbation of skin lesions. The patient suffered from severe pruritus and inability to sleep. She had a personal history of arterial hypertension and hypothyroidism on pharmacological therapy. Of the atopic comorbidities, only polyvalent allergy was present. Because of severe atopic dermatitis, the patient was offered participation in a double-blind, randomized trial with nemolizumab; however, soon after the initiation, her participation had to be terminated because she was diagnosed with stage IIIb colorectal cancer. The patient underwent resection and subsequent adjuvant chemotherapy with capecitabine, which had to be terminated after the seventh cycle due to significant progression of atopic dermatitis. After consultation with the attending oncologist, the use of conventional systemic treatment (cyclosporine, methotrexate, and azathioprine) was contraindicated due to the induction of immunosuppression, which could accelerate the primary malignancy, especially due to the fact that capecitabine therapy was terminated prematurely and, thus, the risk of malignancy relapse was higher. We indicated the patient to use targeted biological treatment with dupilumab in a standard dosage regimen. At the beginning, she had an Eczema Assessment Severity Index (EASI) of 24.4, a Dermatology Life Quality Index (DLQI) of 19, and an itch Numerical Rating Scale (NRS) of 7; her serum IgE level was 1700 IU/mL. Ophthalmological atopic comorbidities were excluded after ophthalmological examination. The patient has now been on dupilumab therapy for more than 1 year. The EASI value is 0.9, which means that she is a EASI90 responder; the DLQI is 0, NRS is 2, and serum IgE level has decreased to 470 IU/mL. There have been no side-effects, and the patient is still in remission from the oncological disease.

Case 2. Our second patient is a 46 year old female with a history of atopic dermatitis, bronchial asthma, polyvalent allergy, and cancer duplicity—colorectal cancer stage IIIa and carcinoma of right kidney stage Ia, both treated with surgical removal, followed by adjuvant chemotherapy and radiotherapy, 2 years before she visited our department. Atopic dermatitis was under control with only occasional application of topical corticosteroids

until she was diagnosed with cancer. After the cancer treatment, she presented to our department with severe pruritus NRS 6/10 and a diffuse eczematous eruption—EASI score of 50 (Figure 1). Impact on quality of life was severe with a DLQI score of 15; she complained of inability to sleep due to continuous pruritus, her work productivity was seriously impaired, and she had to seek psychological help.

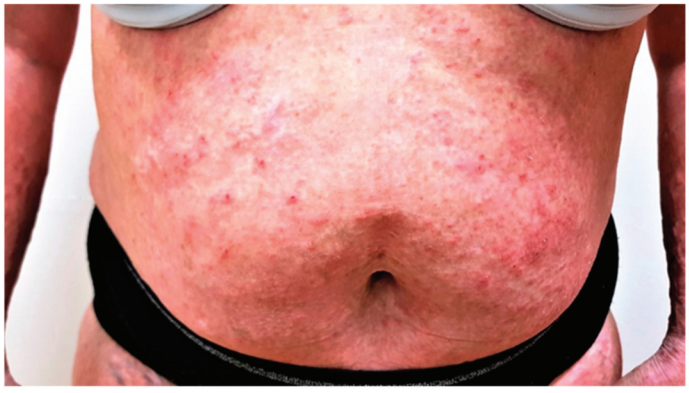

Figure 1. Patient before the initiation of dupilumab with EASI score of 50.

Considering her oncologic anamnesis, we initially treated her with topical treatment in combination with narrowband UVB phototherapy. However, the condition did not improve. Systemic immunosuppressive therapy was considered unsafe due to the previous malignancies. After discussion with her oncologist, we initiated treatment with dupilumab in a standard dosage regimen. The patient responded very well to the treatment (Figure 2); she has been currently treated with dupilumab for 2.5 years, her EASI score is 8, her subjective pruritus has decreased to 1, and her DLQI score has improved to 3. The patient is very satisfied with the results of dupilumab treatment. To date of this publication, no adverse events have occurred. The patient is following up with her oncologist, and surveillance imaging are negative to date.

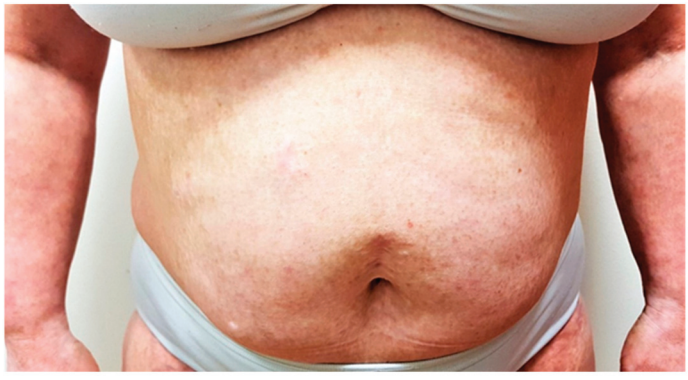

Figure 2. Patient after 2.5 years on dupilumab with EASI score of 8.

Case 3. A 56 year old man with a lifelong history of AD, bronchial asthma, and polyvalent allergy presented to our department with severe pruritus and generalized eczematous lesions, which did not respond to topical treatment or to application of systemic corticosteroids. He had a history of penile spinocellular carcinoma and prostate cancer that was diagnosed 2 years before our treatment initiation, both treated only surgically, in

remission at the time of his visit. Pruritus and inability to sleep had a high impact on his quality of life. Owing to his positive cancer history, he was unable to participate in clinical trials since he would not fulfill the inclusion criteria.

We initiated narrowband UVB phototherapy in combination with topical corticosteroid therapy. However, atopic dermatitis did not improve, and he continued to experience frequent flares. Given the severity of his AD, the impact on his quality of life, and cancer history, systemic immunosuppressive drugs were contraindicated by his oncologist; hence, we decided to start biological treatment. Dupilumab was initiated at a 600 mg loading dose, followed by 300 mg biweekly in a standard dosage regimen. At the beginning of the treatment the EASI score was 32.9 (Figure 3), reported pruritus was 8/10, and DLQI score was 16; the IgE level was 1670 IU/mL. After 4 months of treatment, the EASI score dropped to 4, pruritus score decreased to 1, and DLQI score was 2 (Figure 4). Serum levels of IgE also decreased to 937 IU/mL. The patient continues therapy with dupilumab with good response and no side-effects. After 5 months on therapy, no recurrence of cancer was observed.

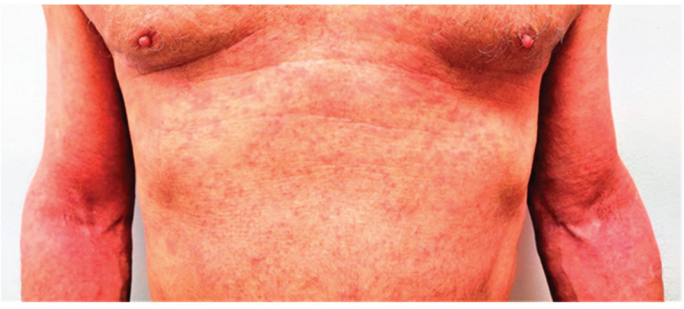

Figure 3. Patient before the initiation of dupilumab with EASI score of 32.9.

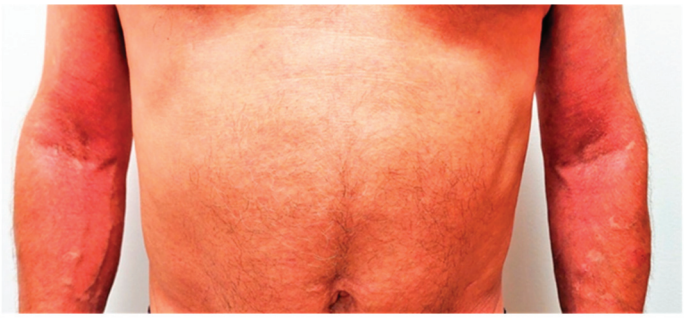

Figure 4. Patient 4 months after the initiation of dupilumab with EASI score of 4.

3. Discussion

Atopic dermatitis (AD) is one of the most common chronic inflammatory dermatoses of noninfectious nature, which, although not life-threatening, significantly affects the patient's quality of life [5]. The treatment of atopic dermatitis has been for long time very complicated due to the fact that available drugs were not suitable for long-term administration, and their use was accompanied by side-effects and is inappropriate for patients with certain comorbidities such as immunodeficiency or malignancy.

The aim of therapy is to achieve remission with minimal drug toxicity and to improve the patient's quality of life.

Significant improvements in the understanding of the etiopathogenesis of atopic dermatitis have been seen in the last few years.

A recent systematic review of preclinical and clinical studies showed that there is no increased risk of malignancy when specifically targeting IL-13 and IL-4 [20]. Data from registries have also not reported an increased risk of malignancy [23,24].

To date, few reports have described real-life experiences of the use of biological drugs in atopic dermatitis patients with a history of malignancy [21,22]. Fowler et al. described two patients with cancer (malignant melanoma and anal squamous cell carcinoma) who were treated with dupilumab safely with good clinical response on atopic dermatitis [21]. Another case report by Qiu et al. presented a patient with history of non-Hodgkin's lymphoma (NHL) with multiple relapses and a severe form of atopic dermatitis. The patient responded to treatment with dupilumab very well, with no reported side-effects [22].

None of the published case reports showed any risk of cancer recurrence for patients with an oncologic history after dupilumab treatment [21,22].

On the other hand, a case of a bladder cancer during dupilumab therapy was recently described; although the authors did not associate the tumor with dupilumab treatment, they stopped the biologic therapy [25]. Interestingly dupilumab has been used in patients with multiple myeloma (MM) for the treatment of lenalidomide rashes. Multiple myeloma remained stable during dupilumab application suggesting that it could have therapeutic benefits as an adjuvant therapy for MM [26]. There is a clinical trial currently enrolling subjects to investigate the safety, antitumor effect, and immunogenicitiy of neoadjuvant dupilumab given prior to radical prostatectomy in men with high-risk localized prostate cancer [27] and another clinical trial that is enrolling subjects to investigate whether dupilumab may be beneficial in patients with metastatic non-small-cell lung cancer following immunotherapy [28]. There is an unproven suggestion that dupilumab may be beneficial for downregulating PD-1 expression on tumor-infiltrating lymphocytes and for improving cancer immunotherapy [28].

In a murine model, IL-4 blockade led to an increase in IL-12, IFNg, and TNF in $CD8^+$ T cells and a reduction in tumor burden, and additional antitumor activity was observed in combination with PD-L1 blockade. On the basis of these preclinical data, it is hypothesized that the addition of dupilumab to anti-PD-(L)1 therapy will be well tolerated and will preserve the antitumor effect of immune checkpoint blockade [28].

We described three cases with severe atopic dermatitis unresponsive to topical therapy and to narrowband phototherapy with a history of malignancy and current dupilumab treatment (Table 1).

Table 1. Oncologic patients with atopic dermatitis treated with dupilumab.

	Cancer before Dupilumab Initiation	Type of Cancer	Previous Cancer Treatment	Year of Cancer Diagnosis	Year of Dupilumab Initiation	Cancer Recurrence	EASI at the Baseline	Current EASI Score
Case 1	Yes	Colorectal carcinoma	Surgery and chemotherapy	2020	2021	No	24.4	0.9
Case 2	Yes	Colorectal carcinoma and kidney cancer	Surgery, chemotherapy, and radiotherapy	2018	2020	No	50	8
Case 3	Yes	Penile spinocellular carcinoma and prostate cancer	Surgery	2020	2022	No	32.9	4

Our patients had a mean interval between cancer diagnosis and initiation of dupilumab treatment of 1.7 years. The tumor types differed from the previous published case series [21,22].

All our patients had a severe form of atopic dermatitis at the time of the visit to our department; they complained of severe pruritus, and their quality of life was also impaired. None of them had any previous systemic treatment for atopic dermatitis. Due to their history of malignant disease, we decided in accordance with their oncologists to not initiate

conventional systemic treatment. On contrary, dupilumab therapy is not connected to a higher risk of malignancy.

We obtained IgE levels in the patient with colorectal carcinoma, as well as in the patient with penile spinocellular carcinoma and prostate cancer, which were increased at baseline. As expected, the serum levels of IgE decreased in both patients after the initiation of dupilumab therapy. IgE plays an important role in the pathogenesis of AD and its concentration is elevated both in serum and on the skin of patients. There is a significant association between higher IgE levels and disease severity [29]. The correlation between high IgE levels and the risk of developing cancer has not been proven [30].

The patients benefited from the effect of dupilumab; they all achieved EASI75, which means an improvement in clinical status in terms of atopic dermatitis of at least 75% compared to baseline, as well as relief from intense pruritus and a significant improvement in their quality of life.

We observed an absence of recurrence or progression of malignant disease, and no patient to date developed a second malignancy. No adverse events related to the dupilumab treatment were observed. All patients handle the subcutaneous application of dupilumab on their own in the home setting, and they all come for regular visits to our department and keep in touch with their oncologist.

Considering the lack of direct association between IL-13 and IL-4 blockage and cancer development, and considering the absence of cancer recurrence, there is no evidence to exclude dupilumab for atopic dermatitis patients with a previous diagnosis of malignancy. Of note, dupilumab is not considered immunosuppressive, but rather immunomodulatory, given its targeted action on the immune system [31]. In atopic dermatitis, the efficacy of IL-13 and IL-4 inhibition has gradually accumulated, and real-world evidence of dupilumab treatment has confirmed its high effectiveness [13,16–18,23,31,32]. In 2022, we published a multicenter prospective real-life experience study in the treatment of atopic dermatitis with dupilumab, as well as its effectiveness and safety. As expected, dupilumab showed very good efficacy and was well tolerated. The effectiveness of dupilumab was expressed by a significant reduction in EASI and DLQI scores. After 4 months, EASI75 response was seen in 66.6% of the patients, which further increased after 1 year to 89.5% [23].

In our patients, continuing therapy with dupilumab did not show cancer progression or recurrence, which suggests that dupilumab can be a safe treatment for atopic dermatitis in patients with advanced cancer. It should be noted that it is always important to evaluate each patient individually since there is still a lack of clinical data regarding the use of dupilumab in patients with atopic dermatitis and malignancy.

Clinicians must be cautious and consider treatment on a case-by-case basis following discussion with an oncologist. Further studies with a higher number of patients and longer follow-up are needed.

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References

1. Barbarot, S.; Auziere, S.; Gadkari, A.; Girolomoni, G.; Puig, L.; Simpson, E.L.; Margolis, D.J.; De Bruin-Weller, M.; Eckert, L. Epidemiology of atopic dermatitis in adults: Results from an international survey. *Allergy* **2018**, *73*, 1284–1293. [CrossRef] [PubMed]
2. Bieber, T. Atopic dermatitis. *Ann. Dermatol.* **2010**, *22*, 125. [CrossRef] [PubMed]
3. Zheng, T.; Jinho, Y.; Oh, M.H.; Zhu, Z. The atopic march: Progression from atopic dermatitis to allergic rhinitis and asthma. *Allergy Asthma Immunol. Res.* **2011**, *3*, 67–73. [CrossRef] [PubMed]
4. Silverberg, N.B.; Silverberg, J.I. Inside out or outside in: Does atopic dermatitis disrupt barrier function or does disruption of barrier function trigger atopic dermatitis? *Cutis* **2015**, *96*, 359–361.
5. Martin, S.; Brown, T.; Fehnel, S.; Deal, L.S.; Katz, E.G.; Chiou, C.-F. The atopic dermatitis itch scale: Development of a new measure to assess pruritus in patients with atopic dermatitis. *J. Dermatolog. Treat.* **2020**, *31*, 484–490. [CrossRef]
6. Paller, A.; Jaworski, J.C.; Simpson, E.L.; Boguniewicz, M.; Russell, J.J.; Block, J.K.; Tofte, S.; Dunn, J.D.; Feldman, S.R.; Clark, A.R.; et al. Major comorbidities of atopic dermatitis: Beyond allergic disorders. *Am. J. Clin. Dermatol.* **2018**, *19*, 821–838. [CrossRef]
7. Li, J.C.; Fishbein, A.; Singam, V.; Patel, K.R.B.; Zee, P.C.; Attarian, H.; Cella, D.; Silverberg, J.I.M. Sleep disturbance and sleep-related impairment in adults with atopic dermatitis: A cross-sectional study. *Dermatitis* **2018**, *29*, 270–277. [CrossRef]
8. Whiteley, J.; Emir, B.; Seitzman, R.; Makinson, G. The burden of atopic dermatitis in US adults: Results from the 2013 National Health and Wellness Survey. *Curr. Med. Res. Opin.* **2016**, *32*, 1645–1651. [CrossRef]
9. Wan, J.; Shin, D.B.; Syed, M.N. 31772 malignancy risk among children and adults with atopic dermatitis in a population-based cohort. *J. Am. Acad. Dermatol.* **2022**, *87*, AB79. [CrossRef]
10. Bickers, D.R.; Lim, H.W.; Margolis, D.; Weinstock, M.A.; Goodman, C.; Faulkner, E.; Gould, C.; Gemmen, E.; Dall, T. The burden of skin diseases: 2004: A joint project of the American Academy of Dermatology Association and the Society for Investigative Dermatology. *J. Am. Acad. Dermatol.* **2006**, *55*, 490–500. [CrossRef]
11. Torres, T.; Ferreira, E.O.; Goncalo, M.; Mendes-Bastos, P.; Selores, M.; Filipe, P. Update on atopic dermatitis. *Acta Med. Port.* **2019**, *32*, 606–613. [CrossRef]
12. Wollenberg, A.; Barbarot, S.; Bieber, T.; Christen-Zaech, S.; Deleuran, M.; Fink-Wagner, A.; Gieler, U.; Girolomoni, G.; Lau, S.; Muraro, A.; et al. Consensus-based European guidelines for treatment of atopic eczema (atopic dermatitis) in adults and children: Part, I.I. *J. Eur. Acad. Dermatol. Venereol.* **2018**, *32*, 850–878. [CrossRef]
13. Simpson, E.L.; Bieber, T.; Guttman-Yassky, E.; Beck, L.A.; Blauvelt, A.; Cork, M.J.; Silverberg, J.I.; Deleuran, M.; Kataoka, Y.; Lacour, J.-P.; et al. Two phase 3 trials of dupilumab versus placebo in atopic dermatitis. *N. Engl. J. Med.* **2016**, *375*, 2335–2348. [CrossRef]
14. Chovatiya, R.; Paller, A.S. JAK inhibitors in the treatment of atopic dermatitis. *J. Allergy Clin. Immunol.* **2021**, *148*, 927–940. [CrossRef]
15. Simon, B.; Bieber, T. Systemic therapy for atopic dermatitis. *Allergy* **2014**, *69*, 46–55. [CrossRef]
16. Blauvelt, A.; de Bruin-Weller, M.; Gooderham, M.; Cather, J.C.; Weisman, J.; Pariser, D.; Simpson, E.L.; Papp, K.A.; Hong, H.C.-H.; Rubel, D.; et al. Long-term management of moderate-to-severe atopic dermatitis with dupilumab and concomitant topical corticosteroids (LIBERTY AD CHRONOS): A 1-year, randomised, double-blinded, placebo-controlled, phase 3 trial. *Lancet* **2017**, *389*, 2287–2303. [CrossRef]
17. Halling, A.-S.; Loft, N.; Silverberg, J.I.; Guttman-Yassky, E.; Thyssen, J.P. Real-world evidence of dupilumab efficacy and risk of adverse events: A systematic review and meta-analysis. *J. Am. Acad. Dermatol.* **2021**, *84*, 139–147. [CrossRef]
18. Seegräber, M.; Srour, J.; Walter, A.; Knop, M.; Wollenberg, A. Dupilumab for treatment of atopic dermatitis. *Expert Rev. Clin. Pharmacol.* **2018**, *11*, 467–474. [CrossRef]
19. FDA/CEDR Resources Page. Food and Drug Administration Web Site. Available online: https://www.accessdata.fda.gov/drugsatfda_docs/label/2022/761055s044lbl.pdf (accessed on 13 January 2023).
20. Braddock, M.; Hanania, N.A.; Sharafkhaneh, A.; Colice, G.; Carlsson, M. Potential risks related to modulating interleukin-13 and interleukin-4 signalling: A systematic review. *Drug Saf.* **2018**, *41*, 489–509. [CrossRef]
21. Fowler, E.; Rosen, J.; Lev-Tov, H.; Yosipovitch, G. Two Cancer Patients Receiving Dupilumab for Treatment of Atopic Dermatitis. *Acta Derm. Venereol.* **2019**, *99*, 899–900. [CrossRef]
22. Qiu, Y.; Ali, K.; Lou, H.; Shan, J.; Wu, L. Successful Treatment of Atopic Dermatitis with Dupilumab in a Patient with Non-Hodgkin's Lymphoma. *Acta Derm.-Venereol.* **2022**, *102*, adv00625. [CrossRef] [PubMed]
23. Kojanová, M.; Tánczosová, M.; Štrosová, D.; Cetkovska, P.; Fialova, J.; Dolezal, T.; Machovcova, A.; Gkalpakiotis, S.; on behalf of the BIOREP Study Group. Dupilumab for the treatment of atopic dermatitis: Realworld data from the Czech Republic BIOREP registry. *J. Dermatol. Treat.* **2022**, *33*, 2578–2586. [CrossRef] [PubMed]
24. Nettis, E.; Ferrucci, S.M.; Ortoncelli, M.; Pellacani, G.; Foti, C.; Di Leo, E.; Patruno, C.; Rongioletti, F.; Argenziano, G.; Macchia, L.; et al. Use of dupilumab in 543 adult patients with moderate-to-severe atopic dermatitis: A multicenter, retrospective study. *J. Investig. Allergol. Clin. Immunol.* **2022**, *32*, 124–132. [CrossRef] [PubMed]
25. Siliquini, N.; Viola, R.; Giura, M.T.; Fierro, M.T.; Ribero, S.; Ortoncelli, M. Case of bladder cancer during Dupilumab therapy: Just an incidental event? *Dermatol. Ther.* **2020**, *33*, e13854. [CrossRef] [PubMed]
26. Owji, S.; Dubin, D.P.; Yassky, D.; Han, J.; Tan, K.; Jagannath, S.; Parekh, S.; Gulati, N. Dupilumab in Multiple Myeloma: A Case Series. *Clin. Lymphoma Myeloma Leuk.* **2022**, *22*, 928–932. [CrossRef]

27. Neoadjuvant Dupilumab in Men with Localized High-Risk Prostate Cancer—Full Text View (No Date) Full Text View—ClinicalTrials.gov. Available online: https://clinicaltrials.gov/ct2/show/NCT03886493 (accessed on 13 January 2023).
28. Fitzgerald, B.G.; Marron, T.U.; Hall, N.; O'Grady, D.; LaMarche, N.; Hennequin, C.; Hegde, S.; Maier, B.; Le Berichel, J.; Chadda, U.; et al. A phase 1b/2 trial of dupilumab given in conjunction with pd-(l)1 blockade in the treatment of relapsed/refractory metastatic NSCLC. *J. Clin. Oncol.* **2022**, *40*, TPS9139. [CrossRef]
29. Wollenberg, A.; Thomsen, S.F.; Lacour, J.P.; Jaumont, X.; Lazarewicz, S. Targeting immunoglobulin E in atopic dermatitis: A review of the existing evidence. *World Allergy Organ. J.* **2021**, *14*, 100519. [CrossRef]
30. Helby, J.; Bojesen, S.; Nielsen, S.; Nordestgaard, B. IGE and risk of cancer in 37 747 individuals from the general population. *Ann. Oncol.* **2015**, *26*, 1784–1790. [CrossRef]
31. Eshtiaghi, P.; Gooderham, M.J. Dupilumab: An evidence-based review of its potential in the treatment of atopic dermatitis. *Core Evid.* **2018**, *13*, 13–20. [CrossRef]
32. Fargnoli, M.C.; Esposito, M.; Ferrucci, S.; Girolomoni, G.; Offidani, A.; Patrizi, A.; Peris, K.; Costanzo, A.; Malara, G.; Pellacani, G.; et al. A 48-week update of a multicentre real-life experience of dupilumab in adult patients with moderate-to-severe atopic dermatitis. *J. Dermatol. Treat.* **2022**, *33*, 1146–1149. [CrossRef]

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Th2 Cytokines Affect the Innate Immune Barrier without Impairing the Physical Barrier in a 3D Model of Normal Human Skin

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Abstract: (1) Background: Atopic dermatitis is one of the most common inflammatory skin diseases characterized by T helper (Th) 2 and Th22 cells producing interleukin (IL)-4/IL-13 and IL-22, respectively. The specific contribution of each cytokine to the impairment of the physical and the immune barrier via Toll-like receptors (TLRs) is poorly addressed concerning the epidermal compartment of the skin. (2) Methods: The effect of IL-4, IL-13, IL-22, and the master cytokine IL-23 is evaluated in a 3D model of normal human skin biopsies (n = 7) at the air–liquid interface for 24 and 48 h. We investigated by immunofluorescence the expressions of (i) claudin-1, zonula occludens (ZO)-1 filaggrin, involucrin for the physical barrier and (ii) TLR2, 4, 7, 9, human beta-defensin 2 (hBD-2) for the immune barrier. (3) Results: Th2 cytokines induce spongiosis and fail in impairing tight junction composition, while IL-22 reduces and IL-23 induces claudin-1 expression. IL-4 and IL-13 affect the TLR-mediated barrier largely than IL-22 and IL-23. IL-4 early inhibits hBD-2 expression, while IL-22 and IL-23 induce its distribution. (4) Conclusions: This experimental approach looks to the pathogenesis of AD through molecular epidermal proteins rather than cytokines only and paves the way for tailored patient therapy.

Keywords: human epidermis; immunofluorescence; transmission electron microscopy; keratinocytes; interleukins; toll-like receptors; human beta-defensin 2; involucrin; filaggrin; claudin-1

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1. Introduction

Atopic dermatitis (AD) is one of the most common inflammatory skin diseases associated with a wide burden and poor quality of life [1,2]. Even if AD is dominated by type T helper (Th) 2 cells and type 2 innate lymphoid cells, other immunopathogenetic pathways seem to play a role according to the many clinical phenotypes [3]. The onset of AD as well as the most acute clinical forms, are associated with high amounts of pro-inflammatory cytokines such as interleukin (IL)-4, IL-13 released by Th2, and IL-22 released by Th17 and Th22 [4]. IL-23 is a core cytokine in many chronic inflammatory disorders, and the IL-17/23 axis is crucial in the pathogenesis of psoriasis [5]. If IL-23 has a specific role in supporting autoimmunity in peripheral tissues, it is not fully validated yet, but it can stabilize the Th17 phenotype and keep their survival [6]. Dendritic cells and macrophages of lesional AD produce high amounts of IL-23, suggesting its role in the initiation and maintenance of skin inflammation [7].

Keratinocytes (KCs), the most represented cytotype in the human epidermis, actively participate in both the physical/chemical barrier and the immunological shield. KCs

proliferate in the basal layer and, in the suprabasal layers, undergo a finely tuned and dynamic morpho-functional rearrangement of the cytoskeleton and intercellular junctions, defined as terminal differentiation. A switch occurs from keratin (K) K5/K14 in the basal compartment to K1/K10 in the suprabasal differentiating layers [8]. Inducible keratins K16 and K17 are associated, respectively, with keratinocyte activation and wound healing, and keratinocyte proliferation [9,10]. In parallel with the cytoskeletal rearrangement, the key structural protein filaggrin, present in the keratohyalin granules in the granular layer, binds to keratin intermediate filaments promoting the formation of the most differentiated KCs, i.e., corneocytes [11–13]. Finally, yet importantly, the late differentiation stage involves the expression of involucrin, providing mechanical strength to corneocytes themselves [14].

Among the different intercellular junctions, tight junctions (TJs), together with desmosomes, represent the first element for the inside-out barrier in the epidermis. Their structure consists of transmembrane integral membrane proteins belonging to the claudin family and scaffold proteins, i.e., zonula occludens family (ZOs), found in the plaque, permitting the connection between TJ transmembrane proteins and cytoskeletal actin filaments [15].

As mentioned above, KCs actively participate as initiators in innate immunity via Toll-like receptors (TLRs) expression and signaling pathways [16]. TLRs belong to the family of pattern recognition receptors (PRRs), which are expressed on both immune cells and non-immune cells [17]. At least ten TLR subtypes have been identified in humans with specific cellular localization and ligands. TLR1, TLR2, TLR4, TLR5, and TLR6 are located on the cell surface. TLR2 is involved in the binding of residues from Gram-positive bacteria, fungi, parasites, and viruses, and TLR4 responds to LPS, a lipopolysaccharide component of the outer membrane of Gram-negative bacteria. TLR7, TLR8, and TLR9 are nucleic acid-sensing TLRs in the endoplasmic reticulum. TLR7 and TLR8 recognize viral single-strand RNA (ssRNA), whereas TLR9 binds unmethylated 2′-deoxyribocytidine-phosphate-guanosine (CpG) DNA motifs frequently present in bacteria and viruses but rare in mammalians [18].

In the normal human epidermis, TLR4 expression is restricted to basal keratinocytes [19,20]. On the other hand, TLR2 and TLR7 are spread throughout the entire epidermal compartment [21,22], while TLR9 can be expressed only occasionally in the granular layer but is often absent [23].

Activation of different TLRs positively regulates the expression of antimicrobial peptides that include, but are not limited to, defensins. The expression of some of these peptides, such as human beta-defensin 1 (hBD-1), is constitutive. In contrast, the expression of others, including human beta-defensin 2 (hBD-2), is triggered by injury or inflammation of the skin [24].

Skin barrier defects have been considered an initial step in developing AD [25], and all components of the barrier can participate in this process. IL-4 and IL-13 are known to be major players [26], but their precise involvement and role are still debated. The inflammatory environment affects the cytoskeletal arrangement, inducing an increase of K16 expression in the suprabasal epidermis [27] and a downregulation of K10 expression [28]. Filaggrin [29,30], loricrin, and involucrin [31] expressions may also be reduced in AD patients. Inappropriate TLR response and AMP expression are associated with autoimmune skin diseases, such as psoriasis and atopic dermatitis (AD) [24,32], which share some immune-mediated steps, but their aetiopathogenesis is different and involves specific pro-inflammatory cytokines [33,34]. The approval of dupilumab, the fully human monoclonal anti-Th2 cytokine, shed light on AD pathogenesis, demonstrating an effect beyond Th2 inhibition [35]. A possible role is thus emerging with regard to IL-22 and IL-23, classically considered “psoriatic” cytokines. Moreover, the functions of IL-4 and IL-13 overlap but are not identical, and the need to elucidate the specific contribution of each cytokine in different processes is not fading in view of identifying more and more precise pharmacological targets.

A 3D model of normal human skin biopsies maintained at the air–liquid interface and standardized in our laboratory [36–40] represents a clear and simple approach to

investigating the early keratinocyte response to a specific inflammatory stimulus. The presence of the physiological epithelial–mesenchymal cross-talk between the epidermis and the underlying dermis mimics as closely as possible the physiological condition. As blood and lymphatic vessels are virtually absent, this setting allows the study of the response induced by each cytokine within the epidermal compartment, paying specific attention to keratinocytes.

In the present study, the impact of the proinflammatory cytokines Th2, i.e., IL-4 and IL-13, IL-22, and IL-23, on (i) the immune epidermal barrier, i.e., TLR2, 4, 7, 9, and hBD-2 expression, and (ii) TJ molecular composition—claudin-1 and ZO-1 were evaluated by immunofluorescence using the 3D model of normal human skin biopsies (n = 7). As for TLRs, TLR2 and TLR4 were chosen for the quantitative analysis as they comprise the recognition of both Gram-positive and Gram-negative bacteria, respectively. To better characterize the specific influence exerted by IL-4 and IL-13, we investigated the expression of biomarkers of cell differentiation, i.e., K14, K10, K16, K17, filaggrin, and involucrin in the same experimental setting. IL-22 effects on these biomarkers have been reported previously [34]. Finally, ultrastructural analysis by transmission electron microscopy (TEM) allowed the measurements of the intercellular distance as an index of spongiosis. All the experiments were performed with biopsies obtained from all subjects.

2. Materials and Methods

2.1. 3D Organotypic Human Skin Culture

Bioptic fragments of normal human skin were obtained after abdominal aesthetic surgery from healthy, non-smoking, 20- to 40-year-old caucasian women (n = 7) after written informed consent, in accordance with the ethical standards of the Institutional Committee on human experimentation and the Helsinki Declaration. Biopsies were reduced with sterile scalpel to fragments 1 × 1 cm and overnight cultured at air–liquid interface in a Transwell system with the dermis immersed in the culture medium and the epidermis facing the air [36,37]. The samples were then exposed to IL-4 (50 ng/mL), IL-13 (50 ng/mL), IL-22 (100 ng/mL), or IL-23 (50 ng/mL) (PeproTech, London, UK) for 24 and 48 h, culturing parallel control groups. All the experiments were performed with biopsies obtained from all subjects. Skin fragments (5 × 5 mm) were immersion-fixed in 4% formalin in PBS 0.1 M, paraffin-embedded, and cut by a rotatory microtome (Bio-Optica, Milan, Italy), obtaining at least 40 serial sections (5 μm thickness) for each sample.

2.2. Immunofluorescence Qualitative Analysis

Specimens were routinely processed for fixation, paraffin embedding, and microtome cut. At least two immunofluorescence experiments were carried out for the qualitative analysis of each marker in each sample in the experimental conditions reported in Table 1. Unspecific binding site saturation was always carried out with 10% goat serum in 0.1 M PBS pH 7.4 (30 min at RT). Negative technical control was always considered on each slide, thus omitting the primary antibody. In samples incubated with Th2 cytokines, K10/K16 double immunostaining was performed. For secondary antibodies, either Alexa Fluor 488 goat anti-mouse or Alexa Fluor 488 goat anti-rabbit (ThermoFisher Scientific, Rockford, IL, USA; dilution 1:200, 1 h at RT) were used as secondary antibodies. Nuclei were counterstained with 4′,6′-diamidino-2-phenylindole dihydrochloride (DAPI; Sigma-Aldrich, St. Louis, MI, USA; dilution 1:50,000, 5 min at RT), and slides were finally mounted with Mowiol 4-88 (Sigma-Aldrich, St. Louis, MI, USA).

Immunofluorescence analysis was performed with a laser scanning confocal microscope Nikon A1R, using constant acquisition parameters for all the experimental groups (Nikon, Tokyo, Japan). hBD-2 experiments were evaluated by a Nikon Eclipse 80i microscope (Nikon, Tokyo, Japan).

Table 1. Antibodies and protocols for indirect immunofluorescence analysis.

Antibody	Antigen Retrieval	Incubation (Antibody Diluted in PBS/BSA 2%)
Polyclonal rabbit anti-human CLDN-1 (ThermoFisher Scientific, Rockford, IL, USA)	0.01 M citrate buffer pH 6 in MW	dilution 1:100 1 h at 37 °C
Polyclonal rabbit anti-human ZO-1 (ThermoFisher Scientific)	Pronase E 10 min at 37°C	dilution 1:100 1 h at 37 °C
Monoclonal mouse anti-human TLR2 (Novus Bio, Littleton, CO, USA)	0.01 M Na citrate buffer pH 6 in MW	1:100 overnight at 4 °C
Monoclonal mouse anti-human TLR4 (Novus Bio)		1:300 1 h at 37 °C
Monoclonal mouse anti-human TLR9 (Novus Bio)		dilution 1:10 overnight at 4 °C
Polyclonal rabbit anti-human TLR7 (Novus Bio)	0.05 M Tris HCl pH 8.5 in MW	1:300 overnight at 4 °C
Polyclonal rabbit-anti-human hBD2 (Santa Cruz Biotechnology, Dallas, TX, USA)	0.01M Na citrate buffer in autoclave 120 °C 6 min	dilution 1:50 overnight 4 °C
Monoclonal mouse anti-human filaggrin (Santa Cruz Biotechnology)	0.01 M Na citrate buffer pH 6 in MW	dilution 1:250, overnight at 4 °C
Monoclonal mouse anti-human involucrin (ThermoFisher Scientific)		dilution 1:1000, overnight at 4 °C
Monoclonal mouse anti-human K10 (Santa Cruz Biotechnology)		dilution 1:50 overnight at 4 °C
Monoclonal rabbit anti-human K16 (Bio SB, Santa Barbara, CA, USA)		dilution 1:100, 1 h at 37 °C
Rabbit anti-human K17 (Abcam, Cambridge, UK)		dilution 1:200, overnight at 4 °C
Monoclonal mouse anti-human K14 (Santa Cruz Biotechnology)	pepsin 0.05% 15′ RT and 0.01M Na citrate buffer pH 6 in MW	1:200 overnight at 4 °C

CLDN-1: claudin-1; ZO-1: zonula occludens 1; TLR: Toll-like receptor; hBD-2: human beta-defensin 2; K: keratin; MW: microwave; RT: Room Temperature; PBS: Phosphate Buffer Saline; BSA: Bovine Serum Albumin.

2.3. Immunofluorescence Quantitative Analysis

For K10, K14, K16, K17, TLR2, and TLR4, at least three experiments were carried out (two slides/sample; two sections/slide), and two blind investigators measured the positive area in μm^2 by ImageJ 1.53 on the whole section and normalized on living epidermis area, excluding the stratum corneum on serial photomicrographs acquired with constant parameters. Results are expressed as mean of the ratio positive area/living epidermal area + 1 SD.

2.4. Transmission Electron Microscopy and Morphometric Analysis

Specimens were routinely processed for TEM analysis and examined by Talos 120 electron microscope (ThermoFisher Scientific, Rockford, IL, USA).

The quantitative analysis of intercellular spaces in both the basal and the suprabasal compartments was performed on ultrathin sections by ImageJ 1.53 on at least 10 random fields per sample, and results were expressed as the mean of intercellular distance (μm) + 1 SD.

2.5. Statistical Analysis

Statistically significant differences were always obtained via Kruskal–Wallis analysis of variance followed by Dunn's post-hoc test using Prism 9.0.0 (GraphPad Software, Boston, MA, USA). Differences were considered statistically significant when $p < 0.05$.

3. Results

3.1. Th2 Cytokines Affect the Epidermal Innate Immune Barrier without Impairing the TJ Composition

In control samples, the membrane-associated expression of claudin-1 (Figure 1A,B) increased from the spinous layer upwards. After Th2 cytokine exposure, claudin-1 immunostaining was always confined in the uppermost epidermis (Figure 1, panels C–F), similar to controls. IL-22 strongly reduced claudin-1 immunopositivity (Figure 1, panels G and H), while IL-23 induced the expression of this TJ protein, particularly in the granular layer (Figure 1, panels I and J).

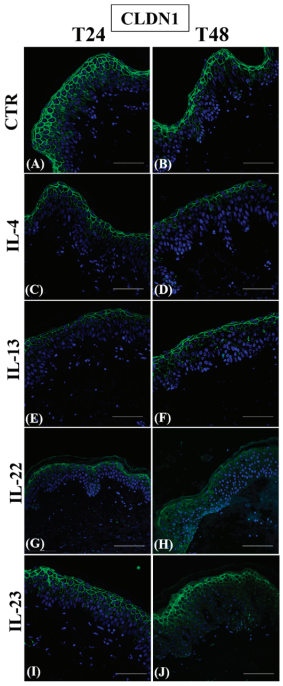

Figure 1. Immunofluorescence of claudin-1 expression on paraffin human skin sections. Representative claudin-1 immunostaining in normal human skin paraffin sections. (**A**,**C**,**E**,**G**,**I**): samples harvested at 24 h; (**B**,**D**,**F**,**H**,**J**): samples harvested at 48 h. (**A**,**B**): CTR samples; (**C**,**D**): IL-4-treated samples; (**E**,**F**): IL-13-treated samples; (**G**,**H**): IL-22-treated samples; (**I**,**J**): IL-23-treated samples. Nuclei are counterstained with DAPI. CTR: control; IL-4: interleukin 4; IL-13: interleukin 13; IL-22: interleukin 22; IL-23: interleukin 23; DAPI: 4′, 6-diamidino-2-phenylindoledihydrochloride—scale bars: 50 μm.

ZO-1 cytoplasmic expression was detected in the most differentiated epidermal layers in control samples (Figure 2, panels A and B).

After Th2 incubation, ZO-1 expression was weak in all the suprabasal layers at 24 h (Figure 2, panels C and E) and even more after 48 h (Figure 2, panels D and F). Conversely, IL-22 and IL-23 induced an evident upregulation of the ZO-1 cytoplasmic expression at T48 in the lower epidermal layers (Figure 2, panels H and J). However, no effect was detected at T24 (Figure 2, panels G and I).

As expected, control groups always showed a homogeneous cytoplasmic TLR2 distribution in the entire epidermal compartment (Figure 3, panels A and B), while TLR4 expression was restricted to basal keratinocytes (Figure 3C) and, at 48 h, only a slight immunopositivity extended upwards (Figure 3D). Similarly to TLR2, TLR7 immunostaining

was always present throughout the epidermis, with both a cytoplasmic and perinuclear localization (Figure 3, panels E and F), while TLR9 expression was never detected (Figure 3, panels G and H).

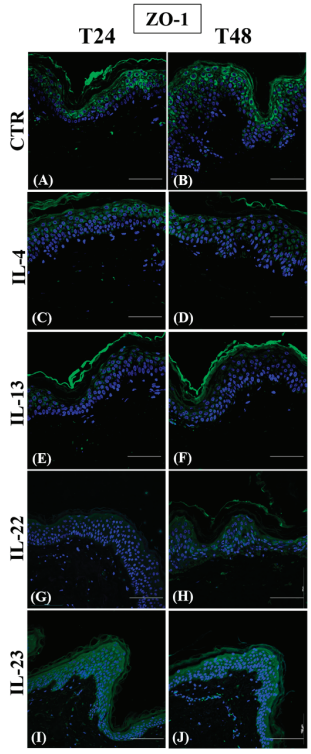

Figure 2. Immunofluorescence of ZO-1 expression on paraffin human skin sections. Representative ZO-1 immunostaining in normal human skin paraffin sections. (**A**,**C**,**E**,**G**,**I**): samples harvested at 24 h; (**B**,**D**,**F**,**H**,**J**): samples harvested at 48 h. (**A**,**B**): CTR samples; (C, D): IL-4-treated samples; (**E**,**F**): IL-13-treated samples; (**G**,**H**): IL-22-treated samples; (**I**,**J**): IL-23-treated samples. Nuclei are counterstained with DAPI. ZO-1: zonula occludens 1. CTR: control; IL-4: interleukin 4; IL-13: interleukin 13; IL-22: interleukin 22; IL-23: interleukin 23; DAPI: 4′, 6-diamidino-2-phenylindoledihydrochloride—scale bars: 50 μm.

TLR2 immunostaining was reduced transiently only in IL-13-exposed samples after 24 h (Figure 4C), while it was comparable in all other cytokine-exposed samples at both time points (Figure 4, panels A, B, and D; Figure 5, panels A–D), similarly to controls (see Figure 3, panels A and B).

Quantitative immunofluorescence analysis of the TLR2-positive area indicated that only IL-13 induced a statistically significant decrease (Figure 6A).

TLR4 appeared discontinuously expressed in the basal layer after cytokine incubation at 24 h (Figure 4, panels E and G; Figure 5, panels E and G) and was reduced to a variable extent by the different cytokines (Figure 6B), with a statistically significant difference only for IL-4 and IL-22. However, at 48 h, IL-4 inhibited TLR4 expression even more evidently than at 24 h (Figures 4F and 6B), while TLR4 immunopositivity was partially restored in IL-22 samples (Figure 5F) or even higher than in controls in IL-13 and IL-23 groups (Figures 4H and 5H) spreading towards the lower spinous layer (Figure 5H, white arrows).

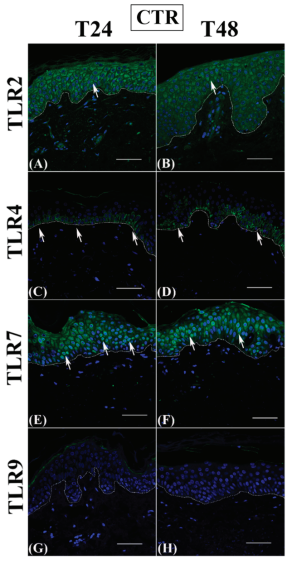

Figure 3. TLR2, TLR4, TLR7, and TLR9 immunofluorescence analysis on paraffin human skin sections. Representative TLR2 (**A**,**B**), TLR4 (**C**,**D**), TLR7 (**E**,**F**), and TLR9 (**G**,**H**) immunostainings in normal human skin paraffin sections. (**A**,**C**,**E**,**G**): samples harvested at 24 h; (**B**,**D**,**F**,**H**): samples harvested at 48 h. (**A**–**H**): control samples. Nuclei are counterstained with DAPI. TLR2: Toll-like receptor 2; TLR4: Toll-like receptor 4; TLR7: Toll-like receptor 7; TLR9: Toll-like receptor 9; DAPI: 4′, 6-diamidino-2-phenylindoledihydrochloride. White dotted line indicates the basal membrane. White arrows indicate positive immunostaining—scale bars: 50 μm.

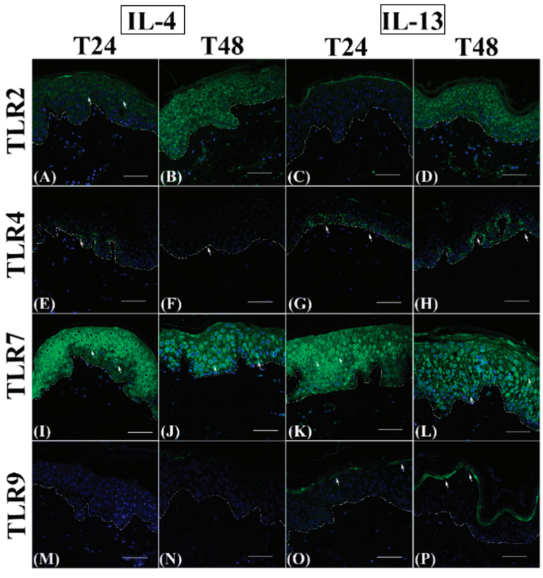

Figure 4. TLR2, TLR4, TLR7, and TLR9 immunofluorescence analysis on IL-4 and IL-13 incubated paraffin human skin sections. Representative TLR2 (**A**–**D**), TLR4 (**E**–**H**), TLR7 (**I**–**L**), and TLR9 (**M**–**P**) immunostainings in normal human skin paraffin sections. (**A**,**E**,**I**,**M**,**C**,**G**,**K**,**O**): samples harvested at

24 h; (**B**,**F**,**J**,**N**,**D**,**H**,**L**,**P**): samples harvested at 48 h. Nuclei are counterstained with DAPI. TLR2: Toll-like receptor 2; TLR4: Toll-like receptor 4; TLR7: Toll-like receptor 7; TLR9: Toll-like receptor 9; IL-4: interleukin 4; IL-13: interleukin 13; DAPI: 4′, 6-diamidino-2-phenylindoledihydrochloride. White dotted line indicates the basal membrane. White arrows indicate positive immunostaining—scale bars: 50 μm.

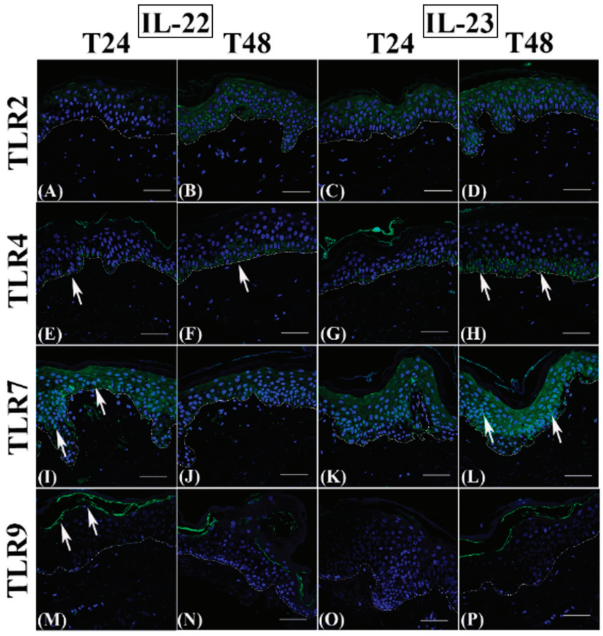

Figure 5. TLR2, TLR4, TLR7, and TLR9 immunofluorescence analysis on IL-22 and IL-23 incubated paraffin human skin sections. Representative TLR2 (**A**–**D**), TLR4 (**E**–**H**), TLR7 (**I**–**L**), and TLR9 (**M**–**P**) immunostainings in normal human skin paraffin sections. (**A**,**E**,**I**,**M**,**C**,**G**,**K**,**O**): samples harvested at 24 h; (**B**,**F**,**J**,**N**,**D**,**H**,**L**,**P**): samples harvested at 48 h. Nuclei are counterstained with DAPI. TLR2: Toll-like receptor 2; TLR4: Toll-like receptor 4; TLR7: Toll-like receptor 7; TLR9: Toll-like receptor 9; IL-22: interleukin 22; IL-23: interleukin 23; DAPI: 4′,6-diamidino-2-phenylindoledihydrochloride. White dotted line indicates the basal membrane. White arrows indicate positive immunostaining—scale bars: 50 μm.

TLR7 immunolabelling intensity was always induced after Th2 cytokine incubation (Figure 4, panels I-L; compared to Figure 3, panels E and F), with a clear perinuclear localization. At 24 h, TLR7 expression was reduced by IL-23 (Figure 5K) but not by IL-22 (Figure 5I), while at T48, TLR4 immunopositivity was inhibited by IL-22 (Figure 5J) but not by IL-23 (Figure 5 L). In all samples, a cytoplasmic localization was evident (Figure 5, panels I–K), with the exception of the IL-23 group at 48 h (Figure 5L). TLR9 induction was never detected in IL-4 (Figure 4, panels M and N) and IL-23 samples (Figure 5, panels O and P), while IL-22 induced a faint and temporary TLR9 induction in samples incubated for 24 h (Figure 5M). Only IL-13 triggered an evident upregulation of TLR9 expression in the granular layer starting from 24 h (Figure 4, panels O and P).

In controls, hBD-2 expression was always localized in the keratinocyte cytoplasm of the medium spinous layer (Figure 7, panels A and B). Only IL-4 completely inhibited hBD-2 expression throughout the entire epidermal compartment (Figure 7, panels C and D), while IL-13 had no effect (Figure 7, panels E and F). On the other hand, IL-22 and IL-23 induced hBD-2 expression throughout the suprabasal compartment at 24 h, an event which became more and more evident at 48 h (Figure 7, panels G–J).

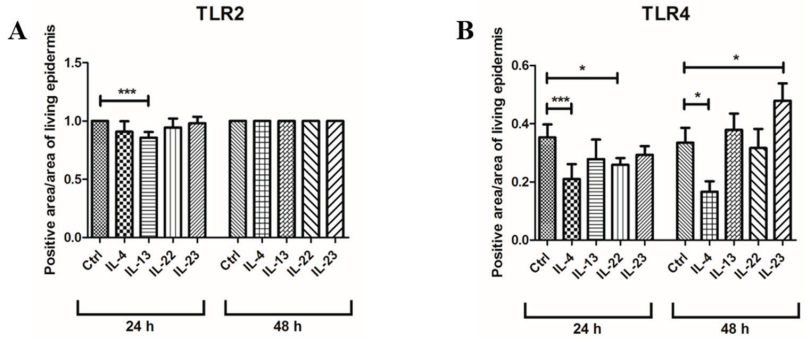

Figure 6. Quantitative analysis of TLR2 and TLR 4 epidermal distribution after immunofluorescence analysis after 24 and 48 h of cytokine incubation. (**A**): TLR2; (**B**): TLR4. TLR2: Toll-like receptor 2; TLR4: Toll-like receptor 4; IL-4: interleukin 4; IL-13: interleukin 13; IL-22: interleukin 22; IL-23: interleukin 23. Statistical analysis was performed by Prism 9.0.0 via Kruskal–Wallis non-parametric analysis of variance, followed by Dunn's post-hoc multiple comparison test. Differences were considered statistically significant when $p < 0.05$. * $p < 0.05$; *** $p < 0.01$.

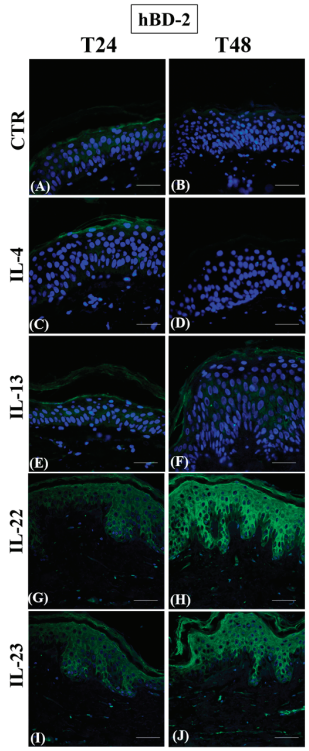

Figure 7. Immunofluorescence of hBD-2 expression on paraffin human skin sections. Representative hBD-2 immunostaining in normal human skin paraffin sections. (**A**,**C**,**E**,**G**,**I**): samples harvested at 24 h; (**B**,**D**,**F**,**H**,**J**): samples harvested at 48 h. (**A**,**B**): CTR samples; (**C**,**D**): IL-4-treated samples; (**E**,**F**): IL-13-treated samples; (**G**,**H**): IL-22-treated samples; (**I**,**J**): IL-23-treated samples. Nuclei are counterstained with DAPI. hBD-2: human beta-defensin 2. CTR: control; IL-4: interleukin 4; IL-13: interleukin 13; IL-22: interleukin 22; IL-23: interleukin 23; DAPI: 4′, 6-diamidino-2-phenylindoledihydrochloride—scale bars: 50 µm.

3.2. Epidermal Homeostasis Is Affected Differently by IL-4 and IL-13

In control groups, immunostainings for filaggrin (Figure 8, panels A and B) and involucrin (Figure 8, panels C and D) were homogeneously distributed in the cytoplasm of granular keratinocytes with a continuous pattern in between adjacent cells. In all cytokine-treated samples, filaggrin immunolabelling was limited to the uppermost region of keratinocyte cytoplasm (Figure 8, panels E, F, I, and J, inserts) and was interrupted after the incubation only with IL-13 for 48 h (Figure 8, panel J, arrowheads). Involucrin distribution in the epidermal compartment was not affected by any cytokine treatment at T24 (Figure 8, panels G and K), while the immunopositivity faded after the exposure to cytokines for 48 h (Figure 8, panels H and L), in particular in IL-4 group.

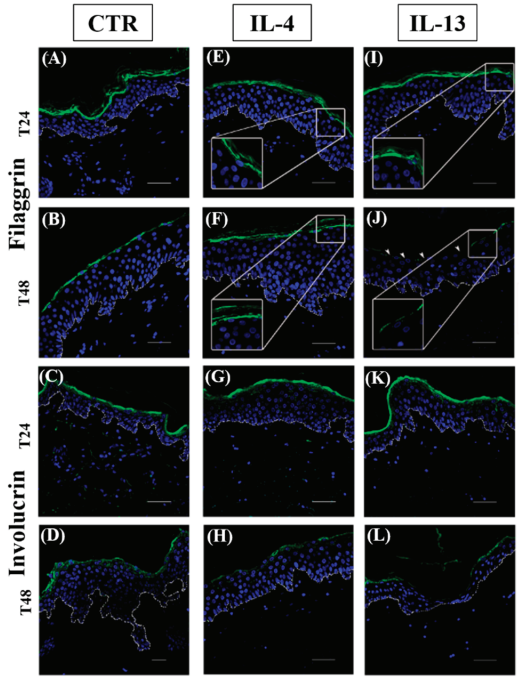

Figure 8. Immunofluorescence analysis of filaggrin and involucrin. Representative filaggrin (**A**,**B**,**E**,**F**,**I**,**J**) and involucrin (**C**,**D**,**G**,**H**,**K**,**L**) immunostainings in normal human skin paraffin sections. (**A**,**C**,**E**,**G**,**I**,**K**): samples harvested at 24 h; (**B**,**D**,**F**,**H**,**J**,**L**): samples harvested at 48 h. (**A**–**D**): CTR samples; (**E**–**H**): IL-4-treated samples; (**I**–**L**): IL-13-treated samples. Nuclei are counterstained with DAPI. CTR: control; IL-4: interleukin 4; IL-13: interleukin 13; DAPI: 4′, 6-diamidino-2-phenylindoledihydrochloride. White dotted line indicates the basal membrane—scale bars: 50 μm.

In all the samples, when present, keratin immunostaining was localized in the cytoplasm of epidermal keratinocytes (Figure 9).

Similarly to controls (Figure 9, panels A and B), K14 immunolabelling was limited to the basal layer. Its intensity decreased in all cytokine-exposed samples (Figure 9, panels E, F, I, and J), except for the group incubated with IL-4 for 48 h, where the immunopositivity spread toward the suprabasal compartment (Figure 9F, arrows). Its intracellular localization was always evident in correspondence with the basal lamina.

K10 and K16 were expressed homogeneously in the suprabasal and the basal layers of all control samples, respectively (Figure 9, panels C and D). The double immunostaining in cytokine-incubated skin revealed a constant decrease of K10-positive area in the lower spinous layer and a time- and cytokine-dependent induction of K16 expression (Figure 9,

panels G, H, K, and L), peaking in IL-13 group after 48 h when it extended upwards the upper spinous layer (Figure 9L).

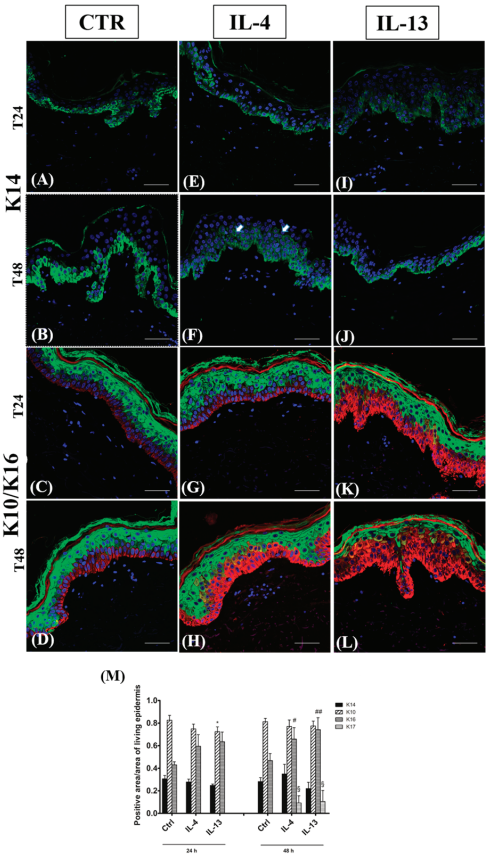

Figure 9. K14 and K10/K16 qualitative and quantitative immunofluorescence analysis. Representative K14 (**A**,**B**,**E**,**F**,**I**,**J**) and K10/K16 (**C**,**D**,**G**,**H**,**K**,**L**) qualitative immunostainings, and (**M**) quantitative analysis in normal human skin paraffin sections. Green staining for K10 and red staining for K16. (**A**,**C**,**E**,**G**,**I**,**K**): samples harvested at 24 h; (**B**,**D**,**F**,**H**,**J**,**L**): samples harvested at 48 h. (**A**–**D**): CTR samples; (**E**–**H**): IL-4-treated samples; (**I**–**L**): IL-13-treated samples. (**M**) Quantitative analysis of K14-, K10-, K16-, and K17-positive areas in normal human skin paraffin sections. Results are expressed as the ratio positive area/area of living epidermis + 1 SD; bars indicate standard error. * $p < 0.05$ vs. all CTR samples; # $p < 0.01$ vs. all CTR samples; ## $p < 0.005$ vs. all CTR samples, § $p < 0.001$ vs. all CTR samples (Kruskal–Wallis analysis of variance followed by Dunn's post-hoc test). Nuclei are counterstained with DAPI. K14: keratin 14; K10: keratin 10; K16: keratin 16; K17: keratin 17; CTR: control; IL-4: interleukin 4; IL-13: interleukin 13; DAPI: 4′, 6-diamidino-2-phenylindoledihydrochloride. Arrowheads indicate the lower spinous layer. White dotted line indicates the basal membrane—scale bars: 50 μm.

K17 expression was absent in controls and all samples harvested at 24 h (Figure S1, panels A–C and E). Only scattered K17-positive keratinocytes were observed after 48 h of cytokine exposure (Figure S1, panels D and F).

The quantitative analysis of the different keratin-positive areas is reported in Figure 9M.

3.3. Spongiosis Is an Early AD Event Triggered by Both Th2 Cytokines

By TEM, in all samples, no detachment between the dermis and the epidermis occurred, the basal membrane was uninterrupted, and the fine structure of desmosomes was always preserved (Figure 10, arrows).

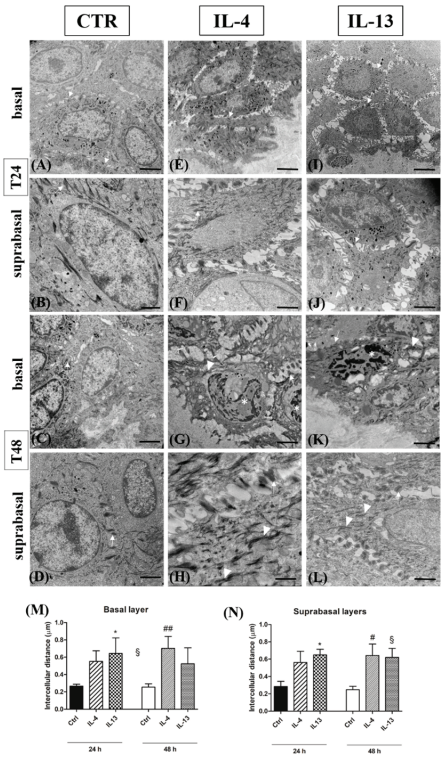

Figure 10. Transmission electron microscopy analysis. Representative photomicrographs of normal human skin araldite ultrathin sections (**A**–**L**) and quantitative analysis of intercellular spaces in the basal (**M**) and the suprabasal layers (**N**). (**A**,**B**,**E**,**F**,**I**,**J**): Samples harvested at 24 h; (**C**,**D**,**G**,**H**,**K**,**L**): samples harvested at 48 h. (**A**,**C**,**E**,**G**,**I**,**K**): Basal layer; (**B**,**D**,**F**,**H**,**J**,**L**): suprabasal layers. (**A**–**D**): CTR samples; (E-H): IL-4-treated samples; (I-L): IL-13-treated samples. Results are expressed as the mean of intercellular distance (μm) + 1 SD; bars indicate standard error. * $p < 0,05$ vs. all CTR samples; # $p < 0.01$ vs. all CTR samples; ## $p < 0.005$ vs. all CTR samples, § $p < 0.001$ vs. all CTR samples (Kruskal–Wallis analysis of variance followed by Dunn's post-hoc test). CTR: control; IL-4: interleukin 4; IL-13: interleukin 13. Arrows indicate desmosomes; arrowheads indicate keratin filament aggregation; asterisks indicate chromatin condensation—scale bars (**A**,**C**,**E**,**G**,**I**,**K**): 2 μm; scale bars (**B**,**D**,**F**,**H**,**J**,**L**): 1 μm.

In all samples harvested at 24 h, keratin bundles were regularly organized, and chromatin appeared finely dispersed in the nuclei of keratinocytes in both the basal (Figure 10, panels A, E, and I) and the suprabasal compartments (Figure 8, panels B, F and J). In cytokine-treated samples harvested at 48 h, chromatin condensation was observed in the nuclei of basal keratinocytes (Figure 10, panels G and K, asterisks). Aggregated keratin filaments were evident in the basal (Figure 10, panels G and K, arrowheads) and the spinous layer (Figure 10, panels H and L, arrowheads). Compared to controls, the cell-to-cell distance was increased in the cytokine-exposed groups (Figure 10, panels E, F, I, and J), with a statistically significant difference for IL-13 in the basal layer at both time points and for both Th2 cytokines in the suprabasal layers at 48 h (Figure 10, panels M and N).

4. Discussion

The identification of the main cellular events underlying AD is more and more challenging because of the multifaceted nature of this disease and the tendency to have heterogeneous clinical features according to the age and medical history of each patient. Furthermore, the initial trigger for AD, the progression mechanisms, and the potential pharmacological intervention require proper experimental skin models to obtain novel insights into new pharmacological targets and tools. In particular, as recently highlighted, "there is an unmet need to better understand epidermal barrier regulation, not only as it applies to general skincare, but also to treatment of common skin conditions, from atopic dermatitis to xerosis" [41]. The 3D normal human skin culture standardized in our laboratory represents a good experimental approach and offers the possibility to study the early impairment of the epidermal barrier in the presence of specific inflammatory stimuli [20,36,39,42].

Our results demonstrate that if, on the one hand, Th2 cytokines fail to impair the physical epidermal barrier, they strongly affect the TLR-mediated innate immune barrier to a greater extent than IL-22. Interestingly, the master cytokine IL-23 does not have any relevant effect on TLR expression, with the exception of TLR4 at 48 h. We can thus hypothesize that, in the early phases of AD, the epidermal barrier is simultaneously impaired on the innate side by Th2 cytokines and on the physical side by IL-22, as indicated in Figure 11.

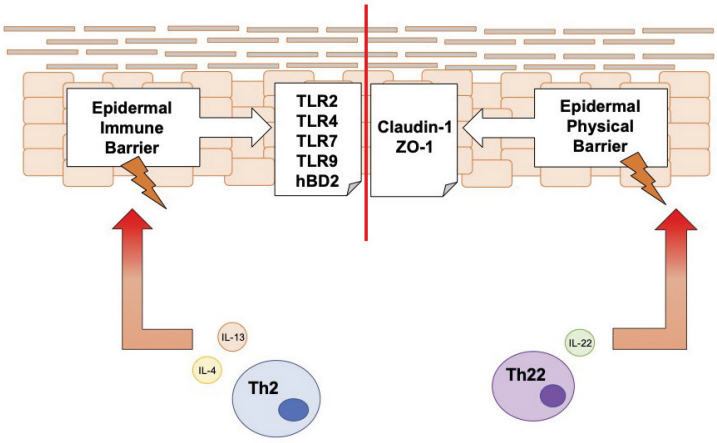

Figure 11. Schematic representation of the early impairment of the epidermal barrier in a proinflammatory microenvironment mimicking atopic dermatitis. TLR: Toll-like receptor; hBD-2: human beta-defensin 2; IL-4: interleukin 4; IL-13: interleukin 13; Th2: T helper (Th) 2 cells; ZO-1: zonula occludens 1; IL-22: interleukin 22; Th22: T helper (Th) 22 cells.

The clinical features observed in AD lesions can be considered the combined result of these two different cytokine-mediated effects. We also demonstrated that although IL-4 and IL-13 are known to share many regulatory mechanisms, their effects are not identical and interfere specifically with some epidermal phenomena. The histo-morphological approach herein presented is pivotal to understanding if the impairment of protein expression is accompanied by a change of the protein epidermal distribution, an issue otherwise neglected with the molecular analysis and/or in in vitro systems.

We report that claudin-1 expression was not affected when either IL-4 or IL-13 alone was added, confirming the experimental evidence obtained in primary human keratinocytes by Yuki et al. [43] and recently in reconstructed human epidermis by Cadau and Coll. [44]. Our study stands in continuation with in vitro studies [45] and Honzke's study performed in skin equivalents incubated with Th2 cytokines [46], with the evident advantage of displaying a fully developed epidermal barrier, thus allowing a more realistic and direct

extrapolation to the clinics. Conversely, claudin-1 expression was inhibited in the presence of IL-22, suggesting that the impaired TJ structure reported in clinics is due not to Th2 cytokines but to IL-22 itself. The reduced expression of ZO-1 in Th2-incubated samples is in agreement with the findings that (i) only a small amount of ZO proteins is required for nucleating claudin strand assembly, (ii) a minimal scaffold at the junction may be sufficient to set up the initial claudin fibrils, and (iii) claudins have the capacity to self-organize [47]. While the role of IL-23 has been widely discussed in the pathogenesis of psoriasis, due to the specific role of dendritic cells [48], less has been reported concerning AD. To the best of our knowledge, the impact of this master cytokine on TJ molecular composition in the epidermis is not elucidated yet. For this reason, the increased expression of both claudin-1 and ZO-1 induced by IL-23 needs further investigation to understand if this effect can relate to mechanisms other than intercellular junctions, in particular for ZO-1.

Filaggrin expression was reduced slightly in the presence of Th2 cytokines, suggesting that this effect is not one of the earliest and constant features occurring during the initial phases of AD lesion formation, but it can represent a later pathogenic event in AD as a clear impairment of its expression is clinically reported in bioptic samples [49]. An early filaggrin expression downregulation was observed in normal human keratinocytes incubated with IL-4 or IL-13 [49] and in engineered skin equivalents [50], but the different settings can account for this discrepancy. On the whole, our evidence is in accordance with the observations that i) FLG gene mutations are not found in all AD patients with a penetration of just 40% [51], and ii) FLG mutation carriers do not always develop AD [52,53]. A similar conclusion can be drawn for involucrin, as its expression was not affected in our experiments, but the reduction of its presence in AD lesions has been reported both at the gene and the protein level [31].

For the immune barrier, Th2 cytokines involved in AD pathogenesis exert a specific and significant tuning in accordance with the literature for TLR2 and TLR4 in bioptic AD skin [21], demonstrating that this shift in TLR expression may be related to a need for enhanced immune surveillance against microbe invasion. Similarly, TLR7 upregulation is reported in peripheral blood monocytes of AD patients [54]. Regarding TLR9 upregulation induced by IL-13, our findings agree with the boost of IL-1α secretion, an interleukin known to be induced in AD patients [55] and observed in HaCaT cells after incubation with strains of *S. aureus* [56]. The relationship between TLR9 and AD was also confirmed by the blockade of the increase of IL-1α levels by the pretreatment with iODNs, a TLR9 antagonist [55].

Considering the "psoriatic" pro-inflammatory cytokines, i.e., IL-22 and IL-23, contrasting results describing the surface TLR pattern distribution in psoriatic plaques are reported [21,57–59]. TLR2/TLR4 upregulation in a psoriatic milieu was reported after stimulation of normal human keratinocytes with IFN-γ and TNF-α [60], but i) the different experimental setting, i.e., 2D vs. 3D, and ii) the different inflammatory stimulus can explain this discrepancy. The absence of a persistent modulation of TLR2/TLR4 expression after exposure either to IL-22 or to IL-23 in our experimental conditions can thus be meaningful. In parallel, the limited effect exerted by IL-22 and IL-23 on TLR7/TLR9 expression should be discussed based on recent observations reporting their upregulation in plaque psoriasis biopsies [61] and after the incubation of 3D organotypic cultures of normal human skin with TNF-alpha, IL-17, IL-22, IL-23, which can reproduce the psoriatic plaque milieu [42]. Both evidences suggest that a complete psoriatic microenvironment is required to modulate TLR7 and TLR9 expression and that a single cytokine is not able to induce any significant tuning in the considered experimental conditions.

Discussing hBD-2 immunoreactivity, considered a useful marker to identify some clinical forms of psoriasis requiring differential diagnosis from DA, the interesting result was that IL-4, but not IL-13, early inhibited its epidermal expression, demonstrating the specificity of the downstream effects induced by each Th2 cytokine. In accordance with previous studies [62], both IL-22 and IL-23 strongly induced hBD-2 expression, thus giving

a clear explanation for the higher levels of this antimicrobial protein found in psoriasis than in AD [63].

In our experimental conditions, both Th2 cytokines similarly reduced K10 expression, i.e., an epidermal differentiation marker of terminal differentiation in the suprabasal layers, but only IL-4 impairs the homeostasis in the basal layer, as shown by the rise of K14 expression. Interestingly, the keratinocyte proliferation rate is never influenced by any cytokine (unpublished personal observations). These data stand in continuation with the existing evidence obtained in in vitro keratinocytes [64,65] and strongly suggest that the alteration of the epidermal differentiation early occurs also in the presence of Th2 cytokines. Furthermore, the enlargement of intercellular spaces was evident as early as 24 h after Th2 cytokine incubation, in particular in the IL-13 group, suggesting that spongiosis is an initial AD pathogenetic event specifically triggered by Th2 cytokines, as recently reported [44–46].

We previously demonstrated that K17 expression, an important and specific psoriatic marker, can be early induced in the same experimental model after the incubation with a mixture of TNF-alpha, IL-17, IL-22, and IL-23 with a pattern distribution in the epidermal compartment very similar to psoriatic skin described in the literature [42]. As expected, in the present study, Th2 cytokines samples did not induce K17 expression, in accordance with the existing literature [10]. In the presence of an AD milieu, the K16-positive epidermal area progressively increased with time, reaching a statistically significant difference compared to the control group at 48 h. This observation stands in continuation with the existing literature [27] and indicates that, once again, the normal epidermis is ready for early and specifically responding to a proinflammatory microenvironment with the expression of inducible keratins. Similarly to the psoriatic microenvironment, in this case, it seems relevant that the stimulus should be prolonged, thus reproducing as strictly as possible the pathological condition in which pro-inflammatory cytokines are involved. Future studies considering the co-presence of IL-4, IL-13, and IL-22 are needed to investigate a potential additive/synergic effect among these cytokines.

The four compartments of the epidermal barrier, i.e., the physical, chemical, immunological, and microbial barrier [66], can be affected by genetic and environmental factors [67]. AD and psoriasis are the main inflammatory skin diseases associated with impaired skin barrier function. The mechanisms related to epidermal barrier dysfunction may be primary and/or secondary.

Indeed, mutations in the filaggrin gene were identified in a subset of patients with AD. On the other hand, considering that not all patients with AD display filaggrin mutations, a combination of primary and secondary barrier defects underlies the disease process. Thus, the inflammation in AD patients, predominantly characterized by a strong and inappropriate Th2 cell activation [68], affects the integrity of the epidermal barrier on multiple levels. Secondary factors adversely affecting the epidermal barrier integrity are predominant in the pathogenesis of psoriasis. Moreover, lipid abnormalities in the stratum corneum were found in both AD and psoriasis [69]. The epithelial barrier function is also crucial for intestinal homeostasis. Indeed, in some subsets of inflammatory bowel disease (IBD), as emerged from the animal model data, barrier dysfunction may be a primary contributor to the disease (a primary defect) and not a consequence of mucosal inflammation [70].

5. Conclusions

TJ dysfunction in the presence of a physiological epidermal stratification/barrier is secondary to Th2 inflammatory processes. Future studies evaluating the functional properties of the epidermal barrier and permeability tests are needed to complete this complex tableau. Nevertheless, this experimental approach looks to the pathogenesis of AD through molecular epidermal proteins rather than cytokines only and paves the way for tailored patient therapy.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/jcm12051941/s1, Figure S1. K17 immunofluorescence analysis.

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References

1. Fölster-Holst, R. Management of Atopic Dermatitis: Are There Differences between Children and Adults? *J. Eur. Acad. Dermatol. Venereol.* **2014**, *28*, 5–8. [CrossRef]
2. Mortz, C.G.; Andersen, K.E.; Dellgren, C.; Barington, T.; Bindslev-Jensen, C. Atopic Dermatitis from Adolescence to Adulthood in the TOACS Cohort: Prevalence, Persistence and Comorbidities. *Allergy Eur. J. Allergy Clin. Immunol.* **2015**, *70*, 836–845. [CrossRef] [PubMed]
3. Kobayashi, T.; Ricardo-Gonzalez, R.R.; Moro, K. Skin-Resident Innate Lymphoid Cells—Cutaneous Innate Guardians and Regulators. *Trends Immunol.* **2020**, *41*, 100–112. [CrossRef] [PubMed]
4. Gittler, J.K.; Shemer, A.; Suárez-Fariñas, M.; Fuentes-Duculan, J.; Gulewicz, K.J.; Wang, C.Q.F.; Mitsui, H.; Cardinale, I.; de Guzman Strong, C.; Krueger, J.G.; et al. Progressive Activation of TH2/TH22 Cytokines and Selective Epidermal Proteins Characterizes Acute and Chronic Atopic Dermatitis. *J. Allergy Clin. Immunol.* **2012**, *130*, 1344–1354. [CrossRef]
5. Teng, M.W.L.; Bowman, E.P.; McElwee, J.J.; Smyth, M.J.; Casanova, J.-L.; Cooper, A.M.; Cua, D.J. IL-12 and IL-23 Cytokines: From Discovery to Targeted Therapies for Immune-Mediated Inflammatory Diseases. *Nat. Med.* **2015**, *21*, 719–729. [CrossRef] [PubMed]
6. Li, Y.; Wang, H.; Lu, H.; Hua, S. Regulation of Memory T Cells by Interleukin-23. *Int. Arch. Allergy Immunol.* **2016**, *169*, 157–162. [CrossRef]
7. Werfel, T.; Allam, J.-P.; Biedermann, T.; Eyerich, K.; Gilles, S.; Guttman-Yassky, E.; Hoetzenecker, W.; Knol, E.; Simon, H.-U.; Wollenberg, A.; et al. Cellular and Molecular Immunologic Mechanisms in Patients with Atopic Dermatitis. *J. Allergy Clin. Immunol.* **2016**, *138*, 336–349. [CrossRef] [PubMed]
8. Coulombe, P.A.; Wong, P. Cytoplasmic Intermediate Filaments Revealed as Dynamic and Multipurpose Scaffolds. *Nat. Cell Biol.* **2004**, *6*, 699–706. [CrossRef]
9. Zhang, X.; Yin, M.; Zhang, L.J. Keratin 6, 16 and 17-Critical Barrier Alarmin Molecules in Skin Wounds and Psoriasis. *Cells* **2019**, *8*, 807. [CrossRef] [PubMed]
10. Komine, M.; Freedberg, I.M.; Blumenberg, M. Regulation of Epidermal Expression of Keratin K17 in Inflammatory Skin Diseases. *J. Investig. Dermatol.* **1996**, *107*, 569–575. [CrossRef] [PubMed]
11. Presland, R.B.; Kimball, J.R.; Kautsky, M.B.; Patrick Lewis, S.; Lo, C.Y.; Dale, B.A. Evidence for Specific Proteolytic Cleavage of the N-Terminal Domain of Human Profilaggrin During Epidermal Differentiation. *J. Investig. Dermatol.* **1997**, *108*, 170–178. [CrossRef] [PubMed]
12. Furue, M. Regulation of Filaggrin, Loricrin, and Involucrin by IL-4, IL-13, IL-17A, IL-22, AHR, and NRF2: Pathogenic Implications in Atopic Dermatitis. *Int. J. Melecular Sci.* **2020**, *21*, 5382. [CrossRef] [PubMed]
13. Proksch, E.; Brandner, J.M.; Jensen, J.-M. The Skin: An Indispensable Barrier. *Exp. Dermatol.* **2008**, *17*, 1063–1072. [CrossRef]
14. Candi, E.; Schmidt, R.; Melino, G. The Cornified Envelope: A Model of Cell Death in the Skin. *Nat. Rev. Mol. Cell Biol.* **2005**, *6*, 328–340. [CrossRef]
15. Crawford, M.; Dagnino, L. Tissue Barriers Scaffolding Proteins in the Development and Maintenance of the Epidermal Permeability Barrier. *Tissue Barriers* **2017**, *5*, e1341969. [CrossRef] [PubMed]
16. Miller, L.S. Toll-Like Receptors in Skin. *Adv. Dermatol.* **2008**, *24*, 71–87. [CrossRef]

17. Santegoets, K.C.M.; van Bon, L.; van den Berg, W.B.; Wenink, M.H.; Radstake, T.R.D.J. Toll-like Receptors in Rheumatic Diseases: Are We Paying a High Price for Our Defense against Bugs? *FEBS Lett.* **2011**, *585*, 3660–3666. [CrossRef]
18. Kawai, T.; Akira, S. The Role of Pattern-Recognition Receptors in Innate Immunity: Update on Toll-like Receptors. *Nat. Immunol.* **2010**, *11*, 373–384. [CrossRef]
19. Liang, N.; Yang, Y.-P.; Li, W.; Wu, Y.-Y.; Zhang, Z.-W.; Luo, Y.; Fan, Y.-M. Overexpression of NLRP3, NLRC4 and AIM2 Inflammasomes and Their Priming-Associated Molecules (TLR2, TLR4, Dectin-1, Dectin-2 and NFκB) in *Malassezia* Folliculitis. *Mycoses* **2018**, *61*, 111–118. [CrossRef]
20. Donetti, E.; Lombardo, G.; Indino, S.; Cornaghi, L.; Arnaboldi, F.; Pescitelli, L.; Baruffaldi Preis, F.; Prignano, F. The Psoriatic Shift Induced by Interleukin 17 Is Promptly Reverted by a Specific Anti-IL-17A Agent in a Three-Dimensional Organotypic Model of Normal Human Skin Culture. *Eur. J. Histochem.* **2020**, *64*, 3115. [CrossRef]
21. Panzer, R.; Blobel, C.; Fölster-Holst, R.; Proksch, E. TLR2 and TLR4 Expression in Atopic Dermatitis, Contact Dermatitis and Psoriasis. *Exp. Dermatol.* **2014**, *23*, 364–366. [CrossRef]
22. Donetti, E.; Cornaghi, L.; Arnaboldi, F.; Ricceri, F.; Pescitelli, L.; Maiocchi, M.; Carriero, F.; Baruffaldi Preis, F.; Prignano, F. Epidermal Barrier Reaction to an in Vitro Psoriatic Microenvironment. *Exp. Cell Res.* **2017**, *360*, 180–188. [CrossRef] [PubMed]
23. Miller, L.S.; Sørensen, O.E.; Liu, P.T.; Jalian, H.R.; Eshtiaghpour, D.; Behmanesh, B.E.; Chung, W.; Starner, T.D.; Kim, J.; Sieling, P.A.; et al. TGF-α Regulates TLR Expression and Function on Epidermal Keratinocytes. *J. Immunol.* **2005**, *174*, 6137–6143. [CrossRef] [PubMed]
24. Ong, P.Y.; Ohtake, T.; Brandt, C.; Strickland, I.; Boguniewicz, M.; Ganz, T.; Gallo, R.L.; Leung, D.Y.M. Endogenous Antimicrobial Peptides and Skin Infections in Atopic Dermatitis. *N. Engl. J. Med.* **2002**, *347*, 1151–1160. [CrossRef] [PubMed]
25. Kim, B.E.; Leung, D.Y.M. Significance of Skin Barrier Dysfunction in Atopic Dermatitis. *Allergy Asthma Immunol. Res.* **2018**, *10*, 207. [CrossRef] [PubMed]
26. Bäsler, K.; Bergmann, S.; Heisig, M.; Naegel, A.; Zorn-Kruppa, M.; Brandner, J.M. The Role of Tight Junctions in Skin Barrier Function and Dermal Absorption. *J. Control. Release* **2016**, *242*, 105–118. [CrossRef]
27. Hamilton, J.D.; Suárez-Fariñas, M.; Dhingra, N.; Cardinale, I.; Li, X.; Kostic, A.; Ming, J.E.; Radin, A.R.; Krueger, J.G.; Graham, N.; et al. Dupilumab Improves the Molecular Signature in Skin of Patients with Moderate-to-Severe Atopic Dermatitis. *J. Allergy Clin. Immunol.* **2014**, *134*, 1293–1300. [CrossRef]
28. Dai, X.; Shiraishi, K.; Muto, J.; Utsunomiya, R.; Mori, H.; Murakami, M.; Sayama, K. Nuclear IL-33 Plays an Important Role in IL-31-Mediated Downregulation of FLG, Keratin 1, and Keratin 10 by Regulating Signal Transducer and Activator of Transcription 3 Activation in Human Keratinocytes. *J. Investig. Dermatol.* **2022**, *142*, 136–144.e3. [CrossRef]
29. Brown, S.J.; Mclean, W.H.I. One Remarkable Molecule: Filaggrin. *J. Investig. Dermatol.* **2012**, *132*, 751–762. [CrossRef]
30. Elias, P.M. Primary Role of Barrier Dysfunction in the Pathogenesis of Atopic Dermatitis. *Exp. Dermatol.* **2018**, *27*, 847–851. [CrossRef]
31. Kim, B.E.; Leung, D.Y.M.; Boguniewicz, M.; Howell, M.D. Loricrin and Involucrin Expression Is Down-Regulated by Th2 Cytokines through STAT-6. *Clin. Immunol.* **2008**, *126*, 332–337. [CrossRef]
32. Ermertcan, A.T.; Öztürk, F.; Gündüz, K. Toll-like Receptors and Skin. *J. Eur. Acad. Dermatol. Venereol.* **2011**, *25*, 997–1006. [CrossRef]
33. Schäbitz, A.; Eyerich, K.; Garzorz-Stark, N. So Close, and yet so Far Away: The Dichotomy of the Specific Immune Response and Inflammation in Psoriasis and Atopic Dermatitis. *J. Intern. Med.* **2021**, *290*, 27–39. [CrossRef] [PubMed]
34. Guttman-Yassky, E.; Krueger, J.G. Atopic Dermatitis and Psoriasis: Two Different Immune Diseases or One Spectrum? *Curr. Opin. Immunol.* **2017**, *48*, 68–73. [CrossRef]
35. Renert-Yuval, Y.; Guttman-Yassky, E. New Treatments for Atopic Dermatitis Targeting beyond IL-4/IL-13 Cytokines. *Ann. Allergy Asthma Immunol.* **2020**, *124*, 28–35. [CrossRef] [PubMed]
36. Bedoni, M.; Sforza, C.; Dolci, C.; Donetti, E. Proliferation and Differentiation Biomarkers in Normal Human Breast Skin Organotypic Cultures. *J. Dermatol. Sci.* **2007**, *46*, 139–142. [CrossRef] [PubMed]
37. Donetti, E.; Cornaghi, L.; Gualerzi, A.; Baruffaldi Preis, F.W.; Prignano, F. An Innovative Three-Dimensional Model of Normal Human Skin to Study the Proinflammatory Psoriatic Effects of Tumor Necrosis Factor-Alpha and Interleukin-17. *Cytokine* **2014**, *68*, 1–8. [CrossRef]
38. Prignano, F.; Arnaboldi, F.; Cornaghi, L.; Landoni, F.; Tripo, L.; Preis, F.W.B.; Donetti, E. Tumor Necrosis Factor-Alpha and Interleukin-17 Differently Affects Langerhans Cell Distribution and Activation in an Innovative Three-Dimensional Model of Normal Human Skin. *Eur. J. Cell Biol.* **2015**, *94*, 71–77. [CrossRef]
39. Chiricozzi, A.; Romanelli, M.; Panduri, S.; Donetti, E.; Prignano, F. Relevance of in Vitro 3-D Skin Models in Dissecting Cytokine Contribution to Psoriasis Pathogenesis. *Histol. Histopathol.* **2017**, *32*, 893–898. [CrossRef] [PubMed]
40. Cornaghi, L.; Gagliano, N.; Preis, F.W.B.; Prignano, F.; Donetti, E. Inside-out and Outside-in Organotypic Normal Human Skin Culture: JAK-STAT Pathway Is Activated after pro-Inflammatory Psoriatic Cytokine Exposure. *Tissue Cell* **2022**, *74*, 101675. [CrossRef]
41. de Benedetto, A. Tight Junctions in the Skin: Still a Lot to Learn. *Br. J. Dermatol.* **2021**, *184*, 388–389. [CrossRef] [PubMed]
42. Donetti, E.; Lombardo, G.; Baruffaldi Preis, F.W.; Cornaghi, L.; Pescitelli, L.; Prignano, F. 3D Skin Model to Investigate the Early Epidermal Morphological Psoriatic Features. *J. Transl. Sci.* **2020**, *6*, 1–4. [CrossRef]

43. Yuki, T.; Tobiishi, M.; Kusaka-Kikushima, A.; Ota, Y.; Tokura, Y. Impaired Tight Junctions in Atopic Dermatitis Skin and in a Skin-Equivalent Model Treated with Interleukin-17. *PLoS ONE* **2016**, *11*, e0161759. [CrossRef] [PubMed]
44. Cadau, S.; Gault, M.; Berthelemy, N.; Hsu, C.-Y.; Danoux, L.; Pelletier, N.; Goudounèche, D.; Pons, C.; Leprince, C.; André-Frei, V.; et al. An Inflamed and Infected Reconstructed Human Epidermis to Study Atopic Dermatitis and Skin Care Ingredients. *Int. J. Mol. Sci.* **2022**, *23*, 12880. [CrossRef] [PubMed]
45. de Benedetto, A.; Rafaels, N.M.; McGirt, L.Y.; Ivanov, A.I.; Georas, S.N.; Cheadle, C.; Berger, A.E.; Zhang, K.; Vidyasagar, S.; Yoshida, T.; et al. Tight Junction Defects in Patients with Atopic Dermatitis. *J. Allergy Clin. Immunol.* **2011**, *127*, 773–786.e7. [CrossRef] [PubMed]
46. Hönzke, S.; Wallmeyer, L.; Ostrowski, A.; Radbruch, M.; Mundhenk, L.; Schäfer-Korting, M.; Hedtrich, S. Influence of Th2 Cytokines on the Cornified Envelope, Tight Junction Proteins, and β-Defensins in Filaggrin-Deficient Skin Equivalents. *J. Investig. Dermatol.* **2016**, *136*, 631–639. [CrossRef]
47. van Itallie, C.M.; Anderson, J.M. Architecture of Tight Junctions and Principles of Molecular Composition. *Semin. Cell Dev. Biol.* **2014**, *36*, 157–165. [CrossRef] [PubMed]
48. Nakamizo, S.; Dutertre, C.-A.; Khalilnezhad, A.; Zhang, X.M.; Lim, S.; Lum, J.; Koh, G.; Foong, C.; Yong, P.J.A.; Tan, K.J.; et al. Single-Cell Analysis of Human Skin Identifies CD14+ Type 3 Dendritic Cells Co-Producing IL1B and IL23A in Psoriasis. *J. Exp. Med.* **2021**, *218*, e20202345. [CrossRef]
49. Howell, M.D.; Kim, B.E.; Gao, P.; Grant, A.V.; Boguniewicz, M.; DeBenedetto, A.; Schneider, L.; Beck, L.A.; Barnes, K.C.; Leung, D.Y.M. Cytokine Modulation of Atopic Dermatitis Filaggrin Skin Expression. *J. Allergy Clin. Immunol.* **2007**, *120*, 150–155. [CrossRef] [PubMed]
50. Danso, M.O.; van Drongelen, V.; Mulder, A.; van Esch, J.; Scott, H.; van Smeden, J.; el Ghalbzouri, A.; Bouwstra, J.A. TNF-α and Th2 Cytokines Induce Atopic Dermatitis-like Features on Epidermal Differentiation Proteins and Stratum Corneum Lipids in Human Skin Equivalents. *J. Investig. Dermatol.* **2014**, *134*, 1941–1950. [CrossRef]
51. Strid, J.; McLean, W.H.I.; Irvine, A.D. Too Much, Too Little or Just Enough: A Goldilocks Effect for IL-13 and Skin Barrier Regulation? *J. Investig. Dermatol.* **2016**, *136*, 561–564. [CrossRef] [PubMed]
52. Kezic, S.; O'Regan, G.M.; Yau, N.; Sandilands, A.; Chen, H.; Campbell, L.E.; Kroboth, K.; Watson, R.; Rowland, M.; Irwin McLean, W.H.; et al. Levels of Filaggrin Degradation Products Are Influenced by Both Filaggrin Genotype and Atopic Dermatitis Severity. *Allergy* **2011**, *66*, 934–940. [CrossRef] [PubMed]
53. Weidinger, S.; O'Sullivan, M.; Illig, T.; Baurecht, H.; Depner, M.; Rodriguez, E.; Ruether, A.; Klopp, N.; Vogelberg, C.; Weiland, S.K.; et al. Filaggrin Mutations, Atopic Eczema, Hay Fever, and Asthma in Children. *J. Allergy Clin. Immunol.* **2008**, *121*, 1203–1209.e1. [CrossRef] [PubMed]
54. dos Santos, V.G.; Orfali, R.L.; de Oliveira Titz, T.; da Silva Duarte, A.J.; Sato, M.N.; Aoki, V. Evidence of Regulatory Myeloid Dendritic Cells and Circulating Inflammatory Epidermal Dendritic Cells-like Modulated by Toll-like Receptors 2 and 7/8 in Adults with Atopic Dermatitis. *Int. J. Dermatol.* **2017**, *56*, 630–635. [CrossRef]
55. Kezic, S.; O'Regan, G.M.; Lutter, R.; Jakasa, I.; Koster, E.S.; Saunders, S.; Caspers, P.; Kemperman, P.M.J.H.; Puppels, G.J.; Sandilands, A.; et al. Filaggrin Loss-of-Function Mutations Are Associated with Enhanced Expression of IL-1 Cytokines in the Stratum Corneum of Patients with Atopic Dermatitis and in a Murine Model of Filaggrin Deficiency. *J. Allergy Clin. Immunol.* **2012**, *129*, 1031–1039.e1. [CrossRef]
56. Moriwaki, M.; Iwamoto, K.; Niitsu, Y.; Matsushima, A.; Yanase, Y.; Hisatsune, J.; Sugai, M.; Hide, M. Staphylococcus Aureus from Atopic Dermatitis Skin Accumulates in the Lysosomes of Keratinocytes with Induction of IL-1α Secretion via TLR9. *Allergy* **2019**, *74*, 560–571. [CrossRef]
57. Seung, N.R.; Park, E.J.; Kim, C.W.; Kim, K.H.; Kim, K.J.; Cho, H.J.; Park, H.R. Comparison of Expression of Heat-Shock Protein 60, Toll-like Receptors 2 and 4, and T-Cell Receptor Γδ in Plaque and Guttate Psoriasis. *J. Cutan. Pathol.* **2007**, *34*, 903–911. [CrossRef]
58. Baker, B.S.; Ovigne, J.-M.; Powles, A.V.; Corcoran, S.; Fry, L. Normal Keratinocytes Express Toll-like Receptors (TLRs) 1, 2 and 5: Modulation of TLR Expression in Chronic Plaque Psoriasis. *Br. J. Dermatol.* **2003**, *148*, 670–679. [CrossRef]
59. Curry, J.L.; Qin, J.-Z.; Bonish, B.; Carrick, R.; Bacon, P.; Panella, J.; Robinson, J.; Nickoloff, B.J. Innate Immune-Related Receptors in Normal and Psoriatic Skin. *Arch. Pathol. Lab. Med.* **2003**, *127*, 178–186. [CrossRef]
60. Begon, É.; Michel, L.; Flageul, B.; Beaudoin, I.; Jean-Louis, F.; Bachelez, H.; Dubertret, L.; Musette, P. Expression, Subcellular Localization and Cytokinic Modulation of Toll-like Receptors (TLRs) in Normal Human Keratinocytes: TLR2 up-Regulation in Psoriatic Skin. *Eur. J. Dermatol.* **2007**, *17*, 497–506. [CrossRef]
61. Prignano, F.; Lombardo, G.; Indino, S.; Ricceri, F.; Cornaghi, L.; Donetti, E.B.; Donetti, E. Evaluation of Expression of Toll-Like Receptors 7 and 9, Proliferation, and Cytoskeletal Biomarkers in Plaque and Guttate Psoriasis: A Pilot Morphological Study. *Eur. J. Histochem.* **2021**, *65*, 3218. [CrossRef] [PubMed]
62. Liang, S.C.; Tan, X.-Y.; Luxenberg, D.P.; Karim, R.; Dunussi-Joannopoulos, K.; Collins, M.; Fouser, L.A. Interleukin (IL)-22 and IL-17 Are Coexpressed by Th17 Cells and Cooperatively Enhance Expression of Antimicrobial Peptides. *J. Exp. Med.* **2006**, *203*, 2271–2279. [CrossRef]
63. de Jongh, G.J.; Zeeuwen, P.L.J.M.; Kucharekova, M.; Pfundt, R.; van der Valk, P.G.; Blokx, W.; Dogan, A.; Hiemstra, P.S.; van de Kerkhof, P.C.; Schalkwijk, J. High Expression Levels of Keratinocyte Antimicrobial Proteins in Psoriasis Compared with Atopic Dermatitis. *J. Investig. Dermatol.* **2005**, *125*, 1163–1173. [CrossRef] [PubMed]

64. Omori-Miyake, M.; Yamashita, M.; Tsunemi, Y.; Kawashima, M.; Yagi, J. In Vitro Assessment of IL-4- or IL-13-Mediated Changes in the Structural Components of Keratinocytes in Mice and Humans. *J. Investig. Dermatol.* **2014**, *134*, 1342–1350. [CrossRef]
65. Totsuka, A.; Omori-Miyake, M.; Kawashima, M.; Yagi, J.; Tsunemi, Y. Expression of Keratin 1, Keratin 10, Desmoglein 1 and Desmocollin 1 in the Epidermis: Possible Downregulation by Interleukin-4 and Interleukin-13 in Atopic Dermatitis. *Eur. J. Dermatol.* **2017**, *27*, 247–253. [CrossRef]
66. Niehues, H.; Bouwstra, J.A.; El Ghalbzouri, A.; Brandner, J.M.; Zeeuwen, P.L.J.M.; van den Bogaard, E.H. 3D skin models for 3R research: The potential of 3D reconstructed skin models to study skin barrier function. *Exp. Dermatol.* **2018**, *27*, 501–511. [CrossRef]
67. Lee, A.Y. Molecular Mechanism of Epidermal Barrier Dysfunction as Primary Abnormalities. *Int. J. Mol. Sci.* **2020**, *21*, 1194. [CrossRef] [PubMed]
68. Pescitelli, L.; Rosi, E.; Ricceri, F.; Pimpinelli, N.; Prignano, F. Novel Therapeutic Approaches and Targets for the Treatment of Atopic Dermatitis. *Curr. Pharm. Biotechnol.* **2021**, *22*, 73–84. [CrossRef]
69. Schmuth, M.; Blunder, S.; Dubrac, S.; Gruber, R.; Moosbrugger-Martinz, V. Epidermal barrier in hereditary ichthyoses, atopic dermatitis, and psoriasis. *J. Der Dtsch. Dermatol. Ges.* **2015**, *13*, 1119–1123. [CrossRef]
70. McGuckin, M.A.; Eri, R.; Simms, L.A.; Florin, T.H.; Radford-Smith, G. Intestinal barrier dysfunction in inflammatory bowel diseases. *Inflamm. Bowel Dis.* **2009**, *15*, 100–113. [CrossRef]

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Review

The Imprint of Exposome on the Development of Atopic Dermatitis across the Lifespan: A Narrative Review

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Abstract: Atopic dermatitis (AD) is a chronic inflammatory skin condition that affects more than 200 million people worldwide, including up to 20% of children and 10% of the adult population. Although AD appears frequently in childhood and often continues into adulthood, about 1 in 4 adults develop the adult-onset disease. The prenatal period, early childhood, and adolescence are considered critical timepoints for the development of AD when the exposome results in long-lasting effects on the immune system. The exposome can be defined as the measure of all the exposures of an individual during their lifetime and how these exposures relate to well-being. While genetic factors could partially explain AD onset, multiple external environmental exposures (external exposome) in early life are implicated and are equally important for understanding AD manifestation. In this review, we describe the conceptual framework of the exposome and its relevance to AD from conception and across the lifespan. Through a spatiotemporal lens that focuses on the multi-level phenotyping of the environment, we highlight a framework that embraces the dynamic complex nature of exposome and recognizes the influence of additive and interactive environmental exposures. Moreover, we highlight the need to understand the developmental origins of AD from an age-related perspective when studying the effects of the exposome on AD, shifting the research paradigm away from the per se categorized exposome factors and beyond clinical contexts to explore the trajectory of age-related exposome risks and hence future preventive interventions.

Keywords: exposome; atopic dermatitis; age; skin of color; COVID-19; war

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1. Introduction

Atopic dermatitis (AD) or atopic eczema is a common inflammatory skin disease with early onset which affects 200 million people worldwide, including up to 20% of children and 10% of adults, but the prevalence of the disease varies greatly throughout the world. In general, AD prevalence is driven by a complex relationship between environmental, genetic predispositions, and immunologic factors. An awareness of environmental diversity is crucial in determining disease phenotypes. More specifically, AD is recognized as a heterogeneous disease with multifactorial etiology and complex pathophysiology involving the immune system and epidermal barrier dysfunction, which are influenced by genetic, epigenetic and environmental factors [1,2]. Genes encoding skin barrier proteins, such as filaggrin (FLG), have been shown to play a role in the inheritance of AD. FLG is synthesized from profilaggrin, which is then transformed into FLG monomers which in turn interact with intermediate filaments in the stratum corneum (SC), causing them to aggregate into dense parallel arrays of macrofilaments. This promotes cellular compaction and keratin crosslinking in the SC, which forms a highly insoluble matrix that acts as a protective barrier. Filaggrin deregulation and loss-of-function (LOF) variants leading to

abnormal FLG production result in skin barrier disruption in individuals carrying FLG LOF variants and are characterized by dry, cracked and infection-prone skin [3]. Dysfunction of the skin barrier is a cardinal clinical sign of AD as this facilitates allergen penetration and immune dysfunction and has been associated with the etiopathogenesis of the AD itch-scratch cycle. This complex pathophysiology translates into heterogeneous clinical phenotypes with differences depending on the age of onset, severity, sensitization profiles, disease persistence, presence of comorbid atopic and nonatopic conditions and longitudinal trajectories of disease progression [4].

The exposome can be defined as the measure of all the exposures of an individual during their lifetime and how these exposures relate to well-being. The external environmental exposures (external exposome) to which an individual is exposed before and after conception and their consequences at the organ and cellular level (internal exposome) are examined to explain the onset, development, and exacerbations of allergic diseases such as AD. Humidity, pollution, ultraviolet ray exposure, average time spent indoors, lifestyle and allergen exposure vary widely throughout different regions and may aggravate AD. Climate change, urbanization and loss of biodiversity are affecting the sources, emissions and concentrations of major airborne allergens and air pollutants and are among the most critical health and quality of life challenges for the increasing number of allergic patients both today and in future decades [5]. As such, the exposome has emerged as a key factor in the development of AD during the lifespan and it is now studied in correlation to the known genetic or biochemical variations that contribute to AD onset.

Globally, the prevalence of AD increases with socioeconomic status and is usually higher in high-income countries, with variations between and within countries. AD may be underdiagnosed in patients with skin of color due to lack of erythema, and therefore, black children are six times more at risk for increased disease severity [6]. African American children and infants of color have a higher prevalence than white American children and infants (37.0% for Black, 25.8% for Asian, 24.1% for Hispanic, 23.0% for multiracial and 17.9% for White). Studies have demonstrated a nearly twofold higher prevalence of AD in black-Caribbean children compared to whites [7]. In addition, the increased prevalence of AD in black patients may extend into old age; furthermore, the socioeconomic environment appears to play a role, as differences in economic burden may reflect in the disease burden [8,9].

Notably, AD prevalence varies between urban and rural populations. The presence of patients in rural areas with limited healthcare access likely results in an underrepresentation of AD prevalence estimations in these areas. Changes in the composition of the gut and skin microbiome due to environmental or lifestyle factors are key mediators of allergic diseases (Figure 1). A better understanding of the impact of external exposome on the development of AD is crucial to encourage patients, health professionals and policymakers to take action to mitigate the impact of and adapt to environmental changes [10].

Previous reviews have attempted to summarize the multitude of factors contributing to AD prevalence and progression, which illustrates the necessity of understanding the exposome to manage AD. However, the exposure to these factors varies not only from person to person but also over the course of a patient's lifetime. In this review, we aim to provide an overview of the AD exposome as an ever-changing variable over the human lifespan. By highlighting patient uniqueness and the temporal variability of their environment in the context of the natural history of AD, insights to guide better population-scale as well as personalized interventions could emerge as an important tool for effective and personalized treatment.

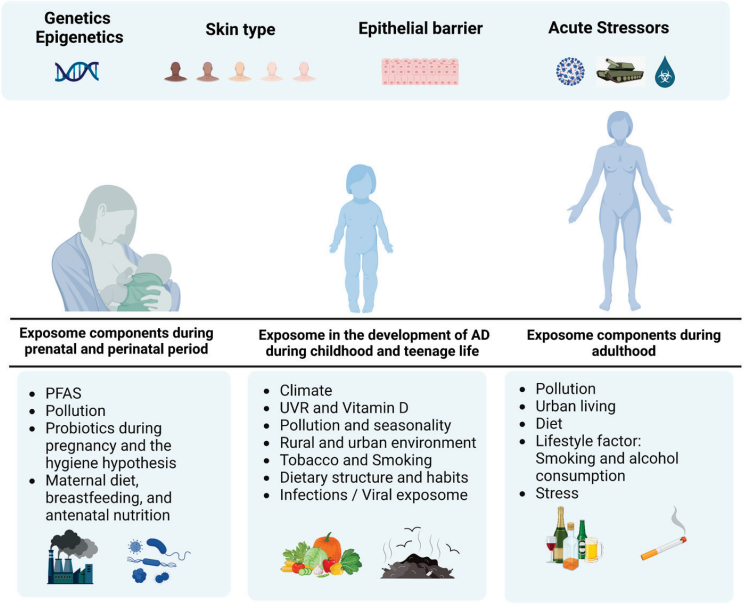

Figure 1. Exposome on the development of atopic dermatitis across the lifespan. Genetic, epigenetic, skin type and epithelial dysfunction only partially explain AD onset. The exposome including pollution, climate, rural and urban environment, lifestyle factors and acute stress differentially affects the trajectory of AD starting from conception, during pregnancy, childhood, adulthood and across the lifespan.

2. The Role of Exposome Components during Prenatal and Perinatal Period

2.1. Perfluoroalkyl and Polyfluoroalkyl Substances (PFASs)

PFASs, widely known as "forever chemicals", are a large group of compounds used for non-stick or stain-resistant surfaces of many products, including several household items. They are widely used in industrial products, i.e., lubricants, surfactants, fire-fighting foams and in everyday products, such as non-stick cookware, greaseproof paper, food packaging, carpets, furniture, waxes, paints, clothing, and personal care products such as shampoo, eye-makeup, nail varnish, and dental floss (Figure 1). Prenatal exposure to PFAS, such as perfluorooctane sulfonate (PFOS), perfluorooctanoic acid (PFOA), perfluorohexane sulfonate (PFHxS), perfluorononanoic acid (PFNA), and perfluorodecanoic acid (PFDA), has been detected in 90% or more of pregnant women in the US, Europe, and Asia and are associated with childhood AD in girls during the first 2 years [11]. In addition, prenatal exposure to polychlorinated biphenyls (PCBs) increased the risks for asthma and eczema in offspring [12]. Prenatal exposure to PFAS occurs through placental transfer in utero and postnatal exposure through breastfeeding and has been associated with a higher risk of early AD in children under 5 years of age [13,14]. Studies have correlated prenatal PFOA and PFOS exposures with elevated cord blood IgE levels, especially in boys, and it has been hypothesized that PFAS might augment hypersensitivity to allergens [15]. Moreover, in utero exposure to PFOA has been associated with a higher risk of AD development as early as the age of 2 years old, with children carrying GSTT1-null or GSTM1-null genotypes that affect the glutathione S-transferase (GST) activity which is essential in chemical detoxification. GSTM1-null and GSTP1 Ile/Ile genotypes are also associated with increased risk of AD in children with prenatal smoke exposure [16]. Finally, in the home environment, exposure to PFAS places infants and young children at increased risk of exposure due to their exploring and hand-to-mouth behavior.

2.2. Pollution

On the other hand, air pollution which is the contamination of the indoor or outdoor environment by any chemical, physical or biological agent that modifies the natural characteristics of the atmosphere can trigger AD onset in childhood. Domestic combustion appliances, motor vehicles, industrial installations and forest fires are common sources of pollutants. Pollutants of major public health concern are grouped into air pollutants (e.g., sulfur dioxide, nitrogen oxide, carbon monoxide, ozone and volatile organic compounds), persistent organic pollutants (e.g., dioxins), heavy metals (e.g., cadmium, lead, mercury) and particulate matter (PM). Exposure to lead in late pregnancy increases the risk of AD in boys at 6 months of age. Prenatal exposure to inorganic arsenic and co-exposure to inorganic arsenic and cadmium were associated with a higher risk of AD in young children [17]. Mono-benzyl phthalate (MBzP) increased the risk of developing eczema in early childhood [18]. Additionally, the combination of prenatal exposure to bisphenol A (BPA) and phthalates could be associated with AD in 6-month-old infants [19].

According to the US Environmental and Protection Agency (EPA), PM is classified according to particle size; hence, PM0.1 (ultrafine particles, $\leq$0.1 μm), PM2.5 (fine particles, $\leq$2.5 μm), PM10 (coarse particles, $\leq$10 μm) [20]. PM causes skin barrier dysfunction and the formation of reactive oxygen species, leading to induced oxidative stress, possibly epigenetic changes, and skin inflammation via direct and indirect mechanisms [20]. Of interest, is the association between maternal exposure to traffic-related pollution and the prevalence of AD in offspring.

Air pollution and the incidence of AD in East Asia were investigated in birth cohort studies, in which exposure to typical traffic-related air pollution from exhaust fumes during pregnancy resulted in increased rates of AD in children, whereas higher concentrations of particulate (PM10, PM2.5) and gaseous (NO_2, VOC, O_3, SO_2) air pollutants increased the intensity of symptoms of existing AD. The effects of prenatal exposure to PM over three trimesters on skin barrier dysfunction were studied. Prenatal exposure to PM in the first trimester and skin barrier dysfunction were positively associated with AD in offspring, early-onset, and greater severity at three years of age. Notably, higher PM2.5 in the first trimester of pregnancy, higher maternal prenatal stress and male sex were associated with AD at age 1. In addition, a significant association between PM2.5 exposure among younger AD individuals aged between 2–30 years old, but not for PM10 exposure has been described. With their smaller size and higher number of metal components, PM2.5 can easily penetrate deep into skin cells and therefore may cause a higher risk of AD than PM10. Interestingly, high levels of PM2.5 during the first trimester and low cord blood vitamin D influenced the early onset of persistent AD, with the most sensitive period being 6 to 7 weeks of gestation mediated by placental DNA methylation [21]. Furthermore, increased maternal exposure to fine PM synergistically interacts with postnatal environmental tobacco smoke inhalation on the development of infantile eczema. Gestational and prenatal exposure to cigarette smoke were both correlated with the development of adult-onset AD. Smoking may affect the immune processes of babies born to smoking mothers, which may have a reduced innate immune response.

2.3. Probiotics during Pregnancy and the Hygiene Hypothesis

Supplementation with probiotics by pregnant women during prenatal and postnatal lactation and postnatal fetuses can efficiently reduce the risk of AD in children, while the effect of multi-strain probiotic mixtures is superior to single-strain formulations [22,23]. In an 11-year, double-blind, placebo-controlled study, pregnant women in the experimental group continued to receive probiotics from 35 weeks gestation to 6 months postpartum, and infants received oral probiotics from birth till the age of 2 years. The rate of AD in the probiotic group was significantly lower than that in the placebo group. In an observational clinical trial, a multi-strain probiotic was administered orally to pregnant women from 4 weeks before delivery to 4 weeks after delivery. The secretion of sIgA in the

infant's feces suggested that oral probiotics during pregnancy and delivery contribute to the improvement of intestinal barrier function during the neonatal period [24].

Disorders of the intestinal microbiota are closely related to the onset and development of AD. Maternal diet, specifically a diet rich in fish, has protective effects on the fetus, due to the immunological effects of long-chain polyunsaturated fatty acids and their contribution to the homeostasis of cell membranes. In addition, alcohol consumption during pregnancy was associated with a significant and dose-dependent increased risk of AD in early infancy [25].

The hygiene hypothesis is further refined to the "old friends' hypothesis", implying that increases in allergies reflect a lack of exposure to beneficial microbiota, which have co-evolved with humans. These include the largely non-harmful commensal microbes acquired from the skin and gut of other humans, as well as micro-organisms such as helminths, Helicobacter pylori, and hepatitis A virus that can persist for life and need to be tolerated [26].

A meta-analysis of the role of probiotics in the prevention and treatment of AD in children shows that oral probiotics given to pregnant women and newborns can effectively reduce the prevalence of AD in children. Children with AD over 1 year of age responded better to oral probiotics, with better efficacy in children with moderate to severe AD than mild AD. In addition, both single and multi-strain preparations, especially the probiotic mixture containing lactobacillus and bifidobacterial, have a therapeutic effect with a superiority of the multi-strain preparation. Lactobacillus sp., such as *Lactobacillus acidophilus* alone or *Lactobacillus acidophilus* mixed preparations with other strains of probiotics can improve clinical symptoms in children with AD. Moreover, combinatorial use of *Lactobacillus casei* and *Lactobacillus salivarius* can reduce IgE levels. Oral and topical probiotics can regulate the distribution of local microbiota and improve the body's immune response, thus proving to be a promising method of prevention and adjuvant treatment of AD in children [27].

2.4. Maternal Diet and Antenatal Nutrition

Maternal diet and antenatal nutrition could affect fetal development by altering fetal programming, which may in turn alter immune response and atopy. Nutrition during infancy and childhood is very important for the development of AD, and breastfeeding for the first 6 months is considered to be effective in preventing the development of atopic diseases (Figure 1). Moreover, breastfeeding for the first 4 months reduces the risk of eczema in the first 4 years. Additionally, feeding infants with extensively hydrolyzed formula (eHF) in the first 4–6 months, avoiding cow's milk and dairy products, and starting solid foods after 4 months have been shown to prevent the development of AD [28].

Inadequate vitamin E intake during pregnancy was associated with a higher risk of AD in two-year-old children [29]. However, reduced risk in offspring was associated with maternal intake of beta-carotene, vitamin E, zinc, calcium, magnesium, and copper during pregnancy. While intake of allergenic foods and foods rich in n-6 polyunsaturated fatty acids during pregnancy may increase the risk of AD in offspring, foods rich in n-3 polyunsaturated fatty acids, fruits, vegetables, and prebiotics may decrease it [30]. Interestingly, an unhealthier diet—which is evaluated as a low score between Prudent Dietary Pattern (PDP) score and Western Dietary Pattern (WDP) score ((PDP-WDP) score)—during pregnancy was associated with a lower risk of atopic dermatitis and a higher risk of infections [31]. Moreover, maternal infection during gestation and exposure to antibiotics in early life was identified as a risk factor for the development of AD [32,33].

3. Exposome in the Development of AD during Childhood and Teenage Life

3.1. Climate, UVR and Vitamin D

AD exacerbates seasonally and in response to climate changes, including UV exposure, humidity, temperature, precipitation, and indoor heating. Combined high UV exposure and temperature appear to have protective effects specific to AD, in contrast to the combination of high humidity and precipitation (Figure 1). In early childhood, direct exposure to

UV radiation is beneficial for reducing the risk of eczema, whereas high latitude and thus reduced exposure to UV radiation is associated with an increased risk [34,35]. In the meantime UV radiation stimulates vitamin D production, while its deficiency has been associated with increased incidence and severity of AD symptoms. Vitamin D3 supplementation significantly improves clinical symptoms in patients with AD, especially in those with winter-related AD [36]. Finally, there is an inverse association between AD severity and serum vitamin D3 levels, whereas low maternal fish consumption and reduced VD3 intake during pregnancy increase the incidence of AD in offspring [37].

Of note, Hispanic and black children are more prone to persistent AD than white children, with black children being more at-risk for incident AD in the first place. In a large study on children residing in urban US areas, black children have the highest prevalence and severity of AD among the examined ethnic groups, as well as the smallest chance for full disease control, while AD onset occurs later in life for Hispanic children [38]. Black children are also exposed to more known risk factors, including social risk factors, such as living in older or rental houses or between two homes, tobacco exposure, lower income, and lower parental educational level [39]. The latter two also affect Hispanic children. It is known that people with darker skin types have lower 25(OH)-vitamin D levels and low maternal 25(OH)D during pregnancy increases the offspring's AD risk. Compared with non-Hispanic whites, AD incidence and persistence are higher among certain non-white racial subgroups. In a longitudinal cohort including black children with AD aged 0 to 2 years, all analyses were stratified by race. It appeared that despite lower vitamin D levels in black participants, allergic sensitization load was associated with FLG expression in the skin without lesions in non-black children, but not in black children with low vitamin D levels [40,41]. Further research is warranted to identify environmental, socioeconomic, and genetic factors that may be responsible for the observed differences.

3.2. Pollution and Seasonality

As children grow older and are exposed to air pollution, there is a higher prevalence or recurrence of AD in areas with high concentrations of PM, benzene, carbon monoxide, sulfur dioxide and nitrogen oxides, with a slight preference for girls over boys. A study examining a large prospective birth cohort showed a significant positive correlation between NO_2 exposure and AD at 6 years. Moreover, a study of pediatric AD patients living in Korean urban industrial areas for two 6-month periods, showed a positive correlation between exposure to PM and exacerbation of AD, with a longitudinal association between elevated PM levels and an increase in reported AD symptoms [42,43].

Apart from climatic factors per se, other factors such as seasonal climate variations, fluctuations in tropospheric ozone levels and air pollutants that have seasonal trends and vary with climate, contribute to the onset of childhood AD. Interestingly, among children, there is a significant seasonal variation, with children showing symptoms mainly in late winter ("winter type") and children showing exacerbation of symptoms in the summer, especially when exposed to grass pollen ("summer type") [44].

Higher odds of eczema were found in areas with a warm, humid, and rainy climate and high levels of SO_2, SO_3, OC and PM2.5, while lower prevalence was found in areas with a warm and sunny climate along with high PM10 and high ozone levels, as well as in areas with high levels of NO_2, NO_3 and PM2.5. In contrast, CO was inversely associated with eczema severity. Higher levels of lead, zinc, nickel, vanadium and arsenic in PM2.5 were associated with increased eczema prevalence, in contrast to copper, potassium and cadmium which were inversely associated with eczema [45].

A study in children aged 0–17 years showed that in the USA, states with higher average annual NO_2, SO_2 and SO_3 were associated with a higher prevalence of eczema, especially in the cold and warm months. In addition, higher levels of tropospheric ozone in the warm months, CO and PM2.5 in the cool months, and NO_3, OC and PM10 in the cool and warm months, were associated with lower odds of eczema. Among countries, the amount of UVR exposure and AD prevalence varies between countries and depends

on both latitude and stratospheric ozone concentration. As for other climatic factors, humidity, high temperatures and low precipitation are inversely correlated with AD. A possible underlying mechanism for UV radiation involvement in AD could be through the reduction in inflammation by cis-uronic acid and modulation of the skin and gut microbiome [20,45,46].

From a different scope, cold outdoor temperatures and exposure to shampoo were associated with eczema aggravation. Currently, it remains unclear whether parabens, which are included in many daily consumer products such as cosmetics, shampoos and personal care products as preservative antimicrobial agents, induce or aggravate it [47,48]. However, a population study suggested an increased association between children aged 0–3 years exposed to parabens [49]. In addition, water hardness is associated with eczema in genetically predisposed infants, especially in carriers of the FLG mutation. Swimming in chlorinated pools was associated with disease worsening and comprises one of the 'irritant factors' currently mentioned in 'eczema school' programs [50].

3.3. Rural and Urban Environment

Living on a farm or in a rural environment in childhood had a protective effect on sensitization even in middle age, but these factors did not protect from new-onset sensitization in adults [51]. It also increases exposure to a wide variety of environmental and pet allergens at an early age, which according to the hygiene hypothesis, reduces the risk of sensitization and development of atopy later in life. In contrast, it increases the risk of allergic disease later in childhood, despite the protective advantage from early life [52,53]. Exposure to a high allergen load may trigger atopy in people who are genetically susceptible. In a cohort of rural Senegalese children and teenagers under the age of 15, contact with cows significantly increased the odds of AD, while by contrast, the presence of a cat in the house showed a protective effect [54]. However, contact with pets, but not cats, has a protective effect mainly in younger ages with a favorable effect of exposure to dogs. Concerning cats, birth cohort studies showed a significant interaction between cat ownership at birth and mutations in FLG (R501X, 2282del4) on the development of early-onset AD [55]. However, a recent meta-analysis could not confirm this hypothesis, implying the complexity of gene-environment interactions in AD pathogenesis [56,57].

Interestingly, both physician-diagnosed and self-reported atopic eczema were rare in Russian Karelia (rural), as compared to Finnish Karelia (urban). An indication of higher microbial exposure in Russian Karelia is that skin and nasal microbiota were found to be significantly more diverse than in Finnish Karelia. In the Russian population of Karelia, contact with nature and a higher prevalence of Acinetobacter colonization was associated with protection from atopic dermatitis, alongside other allergic diseases [58,59]. On the other hand, living in a home with dampness and mold increases the risk for AD. It is important to highlight the significance of urban planning towards a green environment to improve maternal and child health following the associations of residential greenery with eczema in infants and pregnancy which appears to be the critical exposure window [60].

A much higher (25.6% vs. 2.0%) prevalence of eczema was observed in children aged 1 to 3 years in urban areas of South Africa compared to rural areas. Lower exposure to house dust endotoxin in urban and rural households was associated with a higher prevalence of AD, and significant differences in the composition of the house dust microbiota were observed between children with AD and healthy controls [61]. The detection of specific cat antigens in rural households was significantly lower in the AD cohort [41]. A reduced risk of AD was associated with urban children who consumed unpasteurized or fermented milk as well with frequent treatment with anthelmintic drugs [62]. The use of paraffin or kerosene as a means of heating reduced the odds of AD for all households. Overall significant differences in nutrition and cooking habits, contact with nature and animals, increased IL-6 and TNF-a, especially in patients from rural areas [63].

3.4. Tobacco and Smoking

Cigarette smoke is now an important independent risk factor in children aged 6–13 years, especially when there is a history of maternal smoking during pregnancy and infancy (Figure 1) [43]. When the disease occurs in childhood, it raises the hypothesis that a history of passive smoking becomes relevant only later in life [64]. It appears that allergic sensitization occurs over time, while tobacco smoke contributes to the destruction of the skin barrier and increased transdermal TEWL water loss, allowing contact with allergens and pathogens [65]. The worldwide prevalence of AD has been associated with active smoking [66].

3.5. Dietary Structure and Habits

Studies have tried to identify associations between dietary structure, nutrients, and AD. More specifically, protein consumption and especially protein derived from cereal, nuts, fish and seafood, as well as vegetables and vitamins A and E are linked with decreased AD in adolescents aged 13–14 years (Figure 1) [67]. Studies conducted on children and adults have found an inverse relationship between serum vitamin C and E levels with AD, and supplementation of vitamin E reduced AD symptoms [68,69]. The opposite, however, occurs with carbohydrates and fat, especially saturated fat, although olive oil demonstrated a negative association with eczema. In the meantime, adherence to a Mediterranean diet and consumption of fermented milk products might be protective factors for AD. In addition, specific food consumption, besides food allergies, may aggravate AD [70]. Although frequent consumption of fast foods, energy drinks, and convenience food has been related to the recently diagnosed AD in adolescents, extensive studies are needed to determine causality. Global epidemiological studies of children have shown the association of higher parental socioeconomic status with childhood allergic sensitization, likely due to a reduction in allergen exposure during immune system development by living in a healthier environment [71].

3.6. Infections/Viral Exposome

With increased hygiene and lack of exposure to bacteria, viruses and parasites, the immune system is not stimulated to develop immune responses. However, this hypothesis fails to explain the increase in allergy even in areas lacking basic hygiene services and the lack of reduction in atopic disease in those exposed to childhood viral diseases [53].

The composition of bacterial communities at disease preference sites was dramatically different in AD patients compared with controls. Microbial diversity during AD flares depended on the presence or absence of recent AD treatments, with even intermittent treatment being associated with greater bacterial diversity than the absence of recent treatment. In AD, the proportion of *S. aureus* was greater during disease flares than at baseline or after treatment and was associated with worsening disease severity. The S. epidermidis skin commensal also increased significantly during flare-ups. Increases in Streptococcus, Propionibacterium and Corynebacterium species were observed after treatment. Furthermore, the flexures and neck show site-specific microbial colonization. the flexures and neck. In lesions, the flexures have lower alpha diversity and a high abundance of *S. aureus*, while the neck has a high abundance of Malassezia species. [72–74].

The composition of the gut microbiome, although associated with allergic sensitization, does not affect the risk of atopic dermatitis in children up to 6 years of age [75]. The gut microbiome composition in young people shows a reduced presence of Bifidobacterium in AD; helminths play no role in AD in children, as they do not suffer the additional risk of eczema from anti-helminthic treatment, unlike patients with childhood AD [76].

4. The Exposome Effect on AD of Adulthood

4.1. Pollution

At least 1 in 4 cases of AD are adult-onset, with significant heterogeneity in the age of onset (Figure 1). Some adults may have the disease from infancy or school age,

namely childhood-onset AD that persists into adulthood. While others may report onset in adolescence or adulthood, known as adult-onset or recurrent AD [77].

Throughout life, PM exposure exacerbates AD. Air pollutants cause dermal oxidative stress and have been shown to harm the integrity of the skin barrier by altering TEWL, inflammatory cascade, stratum corneum pH and the microbiome. Various oxidative stress markers in the stratum corneum of AD biopsies have shown a correlation with AD severity. Therefore, supporting the hypothesis that environmentally generated ROS may induce oxidative protein damage in the stratum corneum, leading to the disruption of barrier function and exacerbation of AD [78]. There is a perception that air pollution disproportionately affects adult AD compared to pediatric AD.

4.2. Urban Living

Urban living often increases exposure to environmental irritants, such as NO2 which contributes to the formation of trophospheric ozone and greenhouse gases. Short-term exposure to NO2 or VOC caused significantly increased TEWL in both healthy individuals and those with AD [79–81]. Interestingly, exposure to diesel exhaust and proximity to heavy traffic leads to increased IgE production, and has been shown to be an environmental risk factor associated with AD [48,82,83].

Polycyclic aromatic hydrocarbons (PAHs) fluorene and phenanthrene are potentially associated with the pathogenesis of AD in adults [84]. It is hypothesized that PAHs may exert biological effects through the binding of aryl hydrocarbon receptor (AhR) which is suggested to be overexpressed in AD patients [85]. Various organic components of pollutants interact with the AhR in keratinocytes to induce epidermal hyperstimulation through the induction of the neurotrophic factor artemin that causes nerve growth hypersensitivity pruritus and AD pathophysiology [86]. Dry environmental conditions can markedly enhance epidermal structure and function. Low humidity and low temperatures decrease skin barrier function and increase susceptibility towards mechanical stress, while the skin also becomes more reactive towards skin irritants and allergens as pro-inflammatory cytokines and cortisol are released by keratinocytes, and the number of dermal mast cells increases. Cold and dry weather appears to increase the prevalence and risk of flares in patients with atopic dermatitis [87]. However, cold alone for short periods of time did not affect TEWL or skin irritation. TEWL is greater in black skin compared with white skin because the former is characterized by a lower ceramide/cholesterol ratio loss, both contributing to skin dryness [88]. In US and Italy, increased hospital admissions occur during winter due to low temperature, humidity and UVR exposure, as well as increased indoor heating. Whereas admissions increase in the south during summer due to the high temperature and humidity, which induces increased body temperature, sweating and itching [89]. A Nigerian prospective study reported exacerbation of symptoms with hot and humid weather with tropical-black AD patients visiting the clinic in the dry season, (higher temperature and UV index and lower precipitation. Exposure to tropical meteorological variables may affect the occurrence of AD [90].

4.3. Diet

As with childhood AD, data from the 2010 NHIS suggest that increased household income, higher household education level, and households with more individuals are significantly associated with an increased prevalence of AD in adults [91].

Although some studies investigating the effects of dietary fatty acids on AD in adults have indicated that low n-3 intake is inversely correlated with AD in women, other studies have found no association between n-3 intake and AD, and other clinical studies reported that n-3 supplementation in adults did not show any benefit over placebo in AD. Oxidative stress has been shown to induce AD by increasing the pro-inflammatory response. An anti-inflammatory diet includes oily fish (high in n-3), fruit, vegetables, seeds and probiotics and limit meat, whole grains, sugar, and flour. Meantime, 'free radical' foods, with antioxidant qualities, include berries, cherries, citrus fruit, prunes, olives, and green tea [92–94].

Certain foods and dietary patterns can trigger acute changes that lead to visible skin effects. AD has increased globally across all age groups, and this increase has also been associated with the westernization of dietary patterns and increased consumption of processed food. Processed foods and some food additives such as monosodium glutamate (a popular flavor enhancer) could act as pseudo-allergens and increase the occurrence and severity of AD [95]. On the other hand, fermented food intake is associated with a reduced likelihood of atopic dermatitis in a Korean adult population [96].

4.4. Lifestyle Factors—Smoking and Alcohol Consumption

Although, there is no consistent evidence that drinking can cause eczema or a flare-up, adults with AD had higher rates of cigarette smoking and greater odds of ever drinking 12 or more alcoholic beverages in low or high quantities. Current heavier drinking was associated with eczema in all racial groups compared with a lifetime abstaining from or current light drinking. Interestingly, eczema was associated with higher odds of ever drinking 12 or more alcoholic beverages in whites (aOR, 1.15; 95% CI, 1.14–1.15), blacks (aOR, 1.46; 95% CI, 1.46–1.47), and American Indians (aOR, 5.92; 95% CI, 5.83–6.01) but not in Asian-Americans (aOR, 1.00; 95% CI, 0.99–1.00) [97].

The relationship between active and passive smoking and adult AD remains controversial. Early and current cigarette smoking, as well as exposure to environmental cigarette smoking during childhood, have both been associated with a higher incidence of adult AD and current smoking. Both current and ever smoking were significant risk factors for adult AD, compared with non-smoking. Moreover, packs per year were significantly associated with adult AD, suggesting a lifelong cumulative risk in current smokers. In addition, non-smokers with adult AD reported significantly greater environmental tobacco exposure. Therefore, adults should be discouraged from smoking to prevent adult AD in themselves and their family members [98].

Data from German registries show that smoking AD patients have a higher disease burden with a different pattern of lesion distribution in adult AD, with a 2.5 times greater likelihood of foot involvement. Although the scoring of atopic dermatitis showed no difference between smokers and non-smokers, lesional severity of oozing, crusts, and excoriations, along with patient global assessment scores (PGA) of AD severity being higher in smokers compared to non-smokers. In addition, smokers reported increased pruritus intensity and a lower number of weeks with well-controlled AD than non-smokers. Finally, smokers had increased total IgE levels and an early age initial diagnosis of asthma. Interestingly, smokers were predominantly males (58.7%) in their forties [99]. On the other hand a study on US women, did not find a significant association between current smoking and incident AD [100].

In addition, a study of adult AD from Germany suggests increased positive screening for problematic drinking, drug use disorders, Internet addiction and gambling problems compared to the general population [101].

4.5. Stress

Psychological stress has long been observed to affect the course of AD. While chronic stress generally drives pathogenic immune responses, acute stress, in its effort to restore homeostasis, activates the sympathetic axis of the autonomous nervous system (SA), the endocrine hypothalamus–pituitary–adrenal axis (HPA), and the neuronal neuropeptidergic axis (NNA). This in turn causes vasoconstriction, neurogenic inflammation, and the release of pro-inflammatory neuromediators, followed by the anti-inflammatory cholinergic axis of the autonomic nervous system (CA) [102].

Adults with atopic dermatitis exhibit SA hyperresponsiveness, resulting in the transient release of cortisol from the adrenal glands into the bloodstream, while corticotrophin-releasing hormone (CRH), adrenocorticotropic hormone (ACTH) and cortisol from skin cells are released, which favor the Th2 response and inhibit the Th1 response. Endoge-

nous glucocorticoids (GCs) compromise stratum corneum cohesion, epidermal barrier homeostasis and innate immunity, in normal skin [103].

Acute stress has been shown to aggravate the clinical manifestations of AD in both adults and children, and a correlation has been observed between psychosocial stress and the onset or worsening of AD. It has been shown that patients have a significantly higher sympathetic tone compared to healthy controls at rest and, in general, are less able to handle acute stress [104]. We previously mentioned the effect of the maternal psychological state during pregnancy on the development of atopic dermatitis in the offspring, but a psychological aspect of this disease exists until adulthood. Stress and sleep disturbances seem to have a two-way relationship with atopic dermatitis as both potential causes and sequelae of the disease [105,106]. The mechanisms underlying the psychological aspects of eczema across all ages involve stress responses and glucocorticoid secretion in immune dysregulation and the development of scratching behavior in response to pruritus [107,108].

5. Acute Stress Challenges: COVID-19 Pandemic and Russo–Ukrainian War

Despite the fact that children faced a plethora of pandemic-related issues, a reduction in air pollution and lack of contact with outdoor allergens resulted in the improvement of allergies. Preliminary data from the CORAL birth cohort revealed higher rates of egg sensitization and eczema in children born during the first pandemic lockdown [109]. Intensive hand hygiene with warm water and soap, and alcohol-based hand sanitizers (ABHS), were reported to be associated with the rapid development of hand eczema among a high proportion of young children and adults. Avoiding ABHS at school and washing their hands with a non-alcohol and additives soap and water solved their problem and brought their AD back to good control. "School triggers" seem to be important to recognize, avoid and prevent exposure [110].

During the strict government measures to contain the spread of SARS-CoV-2, intensive hand hygiene increased the risk of hand eczema, especially in AD patients (Figure 1). The use of face masks, gloves and repeated hand sanitization has been associated with high rates of adverse skin reactions among healthcare professionals with reports of acute and chronic dermatitis and secondary infection. In healthcare workers wearing protective equipment, TEWL, temperature and erythema were all significantly increased after 2 h of glove and mask use, indicating impaired epidermal barrier function. Adult AD is associated with a significant healthcare burden and loss of work productivity [111,112]. From another perspective, the COVID-19 pandemic impacted the exposome due to excessive use of disposable protective equipment, plastic packaging, and increased use of antibacterials.

The exposome is constantly being exposed to unprecedented factors, such as natural disasters or hostile activities such as the Russo–Ukrainian war in 2022 (Figure 1). There are serious long-term environmental consequences that threaten both the environment and human health. After each explosion, particles of toxic substances and heavy metals such as lead, mercury and depleted uranium are released into the air, water and soil [113]. In addition, service members encounter environmental extremes, physical stress, military gear, and hygiene difficulties, conditions which may flare an AD patient [104,114,115]. Friction, sweating and irritation can lead to increased itch and scratching, refueling the itch-scratch cycle. The real effects of natural disasters, war and pandemics will only become visible in the coming decades.

6. Conclusions

While genetic, epigenetic, racial diversity and epithelial barrier function could partially explain AD onset, multiple exposomal factors and acute stressors are involved across the lifespan and are equally important to understanding the development of AD. While the exposome of AD remains to be elucidated, some factors demonstrate significant age-specificity. Exposomal factors play a role starting from conception, during pregnancy, childhood, adulthood and across the lifespan with an impact on the trajectory of atopic dermatitis. Infants and young children are most affected due to PFAS, pollution and

maternal nutrition. As they grow older, during adolescence, climate, infections, and rural and urban environments take their toll. Later on, lifestyle factors and stress predominate. In light of understanding the spatiotemporal effect of the exposome, investing in prevention strategies in public health and social protection is a mark of responsible action.

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References

1. Langan, S.M.; Irvine, A.D.; Weidinger, S. Atopic Dermatitis. *Lancet* **2020**, *396*, 345–360. [CrossRef] [PubMed]
2. Moltrasio, C.; Romagnuolo, M.; Marzano, A.V. Epigenetic Mechanisms of Epidermal Differentiation. *Int. J. Mol. Sci.* **2022**, *23*, 4874. [CrossRef]
3. Smieszek, S.P.; Welsh, S.; Xiao, C.; Wang, J.; Polymeropoulos, C.; Birznieks, G.; Polymeropoulos, M.H. Correlation of Age-of-Onset of Atopic Dermatitis with Filaggrin Loss-of-Function Variant Status. *Sci. Rep.* **2020**, *10*, 2721. [CrossRef] [PubMed]
4. Irvine, A.D.; Mina-Osorio, P. Disease Trajectories in Childhood Atopic Dermatitis: An Update and Practitioner's Guide. *Br. J. Dermatol.* **2019**, *181*, 895–906. [CrossRef]
5. Stefanovic, N.; Irvine, A.D.; Flohr, C. The Role of the Environment and Exposome in Atopic Dermatitis. *Curr. Treat. Options Allergy* **2021**, *8*, 222–241. [CrossRef] [PubMed]
6. Ben-Gashir, M.A.; Seed, P.T.; Hay, R.J. Reliance on Erythema Scores May Mask Severe Atopic Dermatitis in Black Children Compared with Their White Counterparts. *Br. J. Dermatol.* **2002**, *147*, 920–925. [CrossRef] [PubMed]
7. Mei-Yen Yong, A.; Tay, Y.-K. Atopic Dermatitis. *Dermatol. Clin.* **2017**, *35*, 395–402. [CrossRef] [PubMed]
8. Chovatiya, R.; Begolka, W.S.; Thibau, I.J.; Silverberg, J.I. Financial Burden and Impact of Atopic Dermatitis Out-of-Pocket Healthcare Expenses among Black Individuals in the United States. *Arch. Dermatol. Res.* **2021**, *314*, 739–747. [CrossRef] [PubMed]
9. Lusignan, S.; Alexander, H.; Broderick, C.; Dennis, J.; McGovern, A.; Feeney, C.; Flohr, C. The Epidemiology of Eczema in Children and Adults in England: A Population-based Study Using Primary Care Data. *Clin. Exp. Allergy* **2021**, *51*, 471–482. [CrossRef]
10. West, C.E.; Jenmalm, M.C.; Prescott, S.L. The Gut Microbiota and Its Role in the Development of Allergic Disease: A Wider Perspective. *Clin. Exp. Allergy* **2015**, *45*, 43–53. [CrossRef]
11. Chan, C.; Law, B.; Liu, Y.-H.; Ambrocio, A.; Au, N.; Jiang, M.; Chow, K. The Association between Maternal Stress and Childhood Eczema: A Systematic Review. *Int. J. Environ. Res. Public Health* **2018**, *15*, 395. [CrossRef] [PubMed]
12. Parker-Lalomio, M.; McCann, K.; Piorkowski, J.; Freels, S.; Persky, V.W. Prenatal Exposure to Polychlorinated Biphenyls and Asthma, Eczema/Hay Fever, and Frequent Ear Infections. *J. Asthma* **2018**, *55*, 1105–1115. [CrossRef] [PubMed]
13. Mogensen, U.B.; Grandjean, P.; Nielsen, F.; Weihe, P.; Budtz-Jørgensen, E. Breastfeeding as an Exposure Pathway for Perfluorinated Alkylates. *Environ. Sci. Technol.* **2015**, *49*, 10466–10473. [CrossRef] [PubMed]
14. Xue, J.; Zartarian, V.; Moya, J.; Freeman, N.; Beamer, P.; Black, K.; Tulve, N.; Shalat, S. A Meta-Analysis of Children's Hand-to-Mouth Frequency Data for Estimating Nondietary Ingestion Exposure. *Risk Anal.* **2007**, *27*, 411–420. [CrossRef] [PubMed]
15. von Holst, H.; Nayak, P.; Dembek, Z.; Buehler, S.; Echeverria, D.; Fallacara, D.; John, L. Perfluoroalkyl Substances Exposure and Immunity, Allergic Response, Infection, and Asthma in Children: Review of Epidemiologic Studies. *Heliyon* **2021**, *7*, e08160. [CrossRef] [PubMed]
16. Chen, Q.; Huang, R.; Hua, L.; Guo, Y.; Huang, L.; Zhao, Y.; Wang, X.; Zhang, J. Prenatal Exposure to Perfluoroalkyl and Polyfluoroalkyl Substances and Childhood Atopic Dermatitis: A Prospective Birth Cohort Study. *Environ. Health* **2018**, *17*, 8. [CrossRef]
17. Tsai, T.-L.; Wang, S.-L.; Hsieh, C.-J.; Wen, H.-J.; Kuo, C.-C.; Liu, H.-J.; Sun, C.-W.; Chen, M.-L.; Wu, M.-T.; TMICS Study Group. Association between Prenatal Exposure to Metals and Atopic Dermatitis Among Children Aged 4 Years in Taiwan. *JAMA Netw. Open* **2021**, *4*, e2131327. [CrossRef] [PubMed]
18. Just, A.C.; Whyatt, R.M.; Perzanowski, M.S.; Calafat, A.M.; Perera, F.P.; Goldstein, I.F.; Chen, Q.; Rundle, A.G.; Miller, R.L. Prenatal Exposure to Butylbenzyl Phthalate and Early Eczema in an Urban Cohort. *Environ. Health Perspect.* **2012**, *120*, 1475–1480. [CrossRef] [PubMed]
19. Lee, S.; Park, S.K.; Park, H.; Lee, W.; Lee, J.H.; Hong, Y.-C.; Ha, M.; Kim, Y.; Lee, B.-E.; Ha, E. Joint Association of Prenatal Bisphenol-A and Phthalates Exposure with Risk of Atopic Dermatitis in 6-Month-Old Infants. *Sci. Total Environ.* **2021**, *789*, 147953. [CrossRef] [PubMed]
20. Shaw, T.E.; Currie, G.P.; Koudelka, C.W.; Simpson, E.L. Eczema Prevalence in the United States: Data from the 2003 National Survey of Children's Health. *J. Investig. Dermatol.* **2011**, *131*, 67–73. [CrossRef] [PubMed]

21. Yang, S.-I.; Lee, S.-H.; Lee, S.-Y.; Kim, H.-C.; Kim, H.-B.; Kim, J.-H.; Lim, H.; Park, M.J.; Cho, H.-J.; Yoon, J.; et al. Prenatal PM2.5 Exposure and Vitamin D–Associated Early Persistent Atopic Dermatitis via Placental Methylation. *Ann. Allergy Asthma Immunol.* **2020**, *125*, 665–673.e1. [CrossRef]
22. Foolad, N.; Armstrong, A.W. Prebiotics and Probiotics: The Prevention and Reduction in Severity of Atopic Dermatitis in Children. *Benef. Microbes* **2014**, *5*, 151–160. [CrossRef]
23. Makrgeorgou, A.; Leonardi-Bee, J.; Bath-Hextall, F.J.; Murrell, D.F.; Tang, M.L.; Roberts, A.; Boyle, R.J. Probiotics for Treating Eczema. *Cochrane Database Syst. Rev.* **2018**, *2018*, CD006135. [CrossRef]
24. Liu, Y.; Du, X.; Zhai, S.; Tang, X.; Liu, C.; Li, W. Gut Microbiota and Atopic Dermatitis in Children: A Scoping Review. *BMC Pediatr.* **2022**, *22*, 323. [CrossRef]
25. Prescott, S.L.; Calder, P.C. N-3 Polyunsaturated Fatty Acids and Allergic Disease. *Curr. Opin. Clin. Nutr. Metab. Care* **2004**, *7*, 123–129. [CrossRef]
26. Bloomfield, S.F.; Rook, G.A.; Scott, E.A.; Shanahan, F.; Stanwell-Smith, R.; Turner, P. Time to Abandon the Hygiene Hypothesis: New Perspectives on Allergic Disease, the Human Microbiome, Infectious Disease Prevention and the Role of Targeted Hygiene. *Perspect. Public Health* **2016**, *136*, 213–224. [CrossRef]
27. Amalia, N.; Orchard, D.; Francis, K.L.; King, E. Systematic Review and Meta-analysis on the Use of Probiotic Supplementation in Pregnant Mother, Breastfeeding Mother and Infant for the Prevention of Atopic Dermatitis in Children. *Australas. J. Derm.* **2020**, *61*, 628–643. [CrossRef] [PubMed]
28. Nicolaou, N.; Pancheva, R.; Karaglani, E.; Sekkidou, M.; Marinova-Achkar, M.; Popova, S.; Tzaki, M.; Kapetanaki, A.; Iacovidou, N.; Boutsikou, T.; et al. The Risk Reduction Effect of a Nutritional Intervention With a Partially Hydrolyzed Whey-Based Formula on Cow's Milk Protein Allergy and Atopic Dermatitis in High-Risk Infants Within the First 6 Months of Life: The Allergy Reduction Trial (A.R.T.), a Multicenter Double-Blinded Randomized Controlled Study. *Front. Nutr.* **2022**, *9*, 863599. [CrossRef]
29. LISA Study Group; Sausenthaler, S.; Koletzko, S.; Schaaf, B.; Lehmann, I.; Borte, M.; Herbarth, O.; von Berg, A.; Wichmann, H.-E.; Heinrich, J. Maternal Diet during Pregnancy in Relation to Eczema and Allergic Sensitization in the Offspring at 2 y of Age. *Am. J. Clin. Nutr.* **2007**, *85*, 530–537. [CrossRef] [PubMed]
30. Gardner, K.G.; Gebretsadik, T.; Hartman, T.J.; Rosa, M.J.; Tylavsky, F.A.; Adgent, M.A.; Moore, P.E.; Kocak, M.; Bush, N.R.; Davis, R.L.; et al. Prenatal Omega-3 and Omega-6 Polyunsaturated Fatty Acids and Childhood Atopic Dermatitis. *J. Allergy Clin. Immunol. Pract.* **2020**, *8*, 937–944. [CrossRef] [PubMed]
31. Brzozowska, A.; Podlecka, D.; Jankowska, A.; Król, A.; Kaleta, D.; Trafalska, E.; Nowakowska-Świrta, E.; Kałużny, P.; Hanke, W.; Bal-Gierańczyk, K.; et al. Maternal Diet during Pregnancy and Risk of Allergic Diseases in Children up to 7–9 Years Old from Polish Mother and Child Cohort Study. *Environ. Res.* **2022**, *208*, 112682. [CrossRef]
32. Mubanga, M.; Lundholm, C.; D'Onofrio, B.M.; Stratmann, M.; Hedman, A.; Almqvist, C. Association of Early Life Exposure to Antibiotics With Risk of Atopic Dermatitis in Sweden. *JAMA Netw. Open* **2021**, *4*, e215245. [CrossRef]
33. Cui, H.; Mu, Z. Prenatal Maternal Risk Factors Contributing to Atopic Dermatitis: A Systematic Review and Meta-Analysis of Cohort Studies. *Ann. Derm.* **2023**, *35*, 11. [CrossRef]
34. Silverberg, J.I.; Hanifin, J.; Simpson, E.L. Climatic Factors Are Associated with Childhood Eczema Prevalence in the United States. *J. Investig. Dermatol.* **2013**, *133*, 1752–1759. [CrossRef]
35. Rueter, K.; Jones, A.P.; Siafarikas, A.; Chivers, P.; Prescott, S.L.; Palmer, D.J. The Influence of Sunlight Exposure and Sun Protecting Behaviours on Allergic Outcomes in Early Childhood. *Int. J. Environ. Res. Public Health* **2021**, *18*, 5429. [CrossRef] [PubMed]
36. Camargo, C.A.; Ganmaa, D.; Sidbury, R.; Erdenedelger, K.; Radnaakhand, N.; Khandsuren, B. Randomized Trial of Vitamin D Supplementation for Winter-Related Atopic Dermatitis in Children. *J. Allergy Clin. Immunol.* **2014**, *134*, 831–835.e1. [CrossRef] [PubMed]
37. Thyssen, J.P.; Zirwas, M.J.; Elias, P.M. Potential Role of Reduced Environmental UV Exposure as a Driver of the Current Epidemic of Atopic Dermatitis. *J. Allergy Clin. Immunol.* **2015**, *136*, 1163–1169. [CrossRef] [PubMed]
38. McKenzie, C.; Silverberg, J.I. The Prevalence and Persistence of Atopic Dermatitis in Urban United States Children. *Ann. Allergy Asthma Immunol.* **2019**, *123*, 173–178.e1. [CrossRef]
39. Wan, J.; Oganisian, A.; Spieker, A.J.; Hoffstad, O.J.; Mitra, N.; Margolis, D.J.; Takeshita, J. Racial/Ethnic Variation in Use of Ambulatory and Emergency Care for Atopic Dermatitis among US Children. *J. Investig. Dermatol.* **2019**, *139*, 1906–1913.e1. [CrossRef] [PubMed]
40. Tackett, K.J.; Jenkins, F.; Morrell, D.S.; McShane, D.B.; Burkhart, C.N. Structural Racism and Its Influence on the Severity of Atopic Dermatitis in African American Children. *Pediatr. Derm.* **2020**, *37*, 142–146. [CrossRef]
41. Botha, M.; Basera, W.; Facey-Thomas, H.E.; Gaunt, B.; Genuneit, J.; Gray, C.L.; Kiragu, W.; Ramjith, J.; Watkins, A.; Levin, M.E. Nutrition and Allergic Diseases in Urban and Rural Communities from the South African Food Allergy Cohort. *Pediatr. Allergy Immunol.* **2019**, *30*, 511–521. [CrossRef]
42. Lee, J.-T.; Cho, Y.-S.; Son, J.-Y. Relationship between Ambient Ozone Concentrations and Daily Hospital Admissions for Childhood Asthma/Atopic Dermatitis in Two Cities of Korea during 2004–2005. *Int. J. Environ. Health Res.* **2010**, *20*, 1–11. [CrossRef] [PubMed]
43. Yi, O.; Kwon, H.-J.; Kim, H.; Ha, M.; Hong, S.-J.; Hong, Y.-C.; Leem, J.-H.; Sakong, J.; Lee, C.G.; Kim, S.-Y.; et al. Effect of Environmental Tobacco Smoke on Atopic Dermatitis among Children in Korea. *Environ. Res.* **2012**, *113*, 40–45. [CrossRef]

44. Krämer, U.; Weidinger, S.; Darsow, U.; Möhrenschlager, M.; Ring, J.; Behrendt, H. Seasonality in Symptom Severity Influenced by Temperature or Grass Pollen: Results of a Panel Study in Children with Eczema. *J. Investig. Dermatol.* **2005**, *124*, 514–523. [CrossRef] [PubMed]
45. Kathuria, P.; Silverberg, J.I. Association of Pollution and Climate with Atopic Eczema in US Children. *Pediatr. Allergy Immunol.* **2016**, *27*, 478–485. [CrossRef]
46. Patra, V.; Byrne, S.N.; Wolf, P. The Skin Microbiome: Is It Affected by UV-Induced Immune Suppression? *Front. Microbiol.* **2016**, *7*, 1235. [CrossRef] [PubMed]
47. Herrero-Fernandez, M.; Montero-Vilchez, T.; Diaz-Calvillo, P.; Romera-Vilchez, M.; Buendia-Eisman, A.; Arias-Santiago, S. Impact of Water Exposure and Temperature Changes on Skin Barrier Function. *J. Clin. Med.* **2022**, *11*, 298. [CrossRef]
48. Tamagawa-Mineoka, R.; Katoh, N. Atopic Dermatitis: Identification and Management of Complicating Factors. *Int. J. Mol. Sci.* **2020**, *21*, 2671. [CrossRef] [PubMed]
49. Arafune, J.; Tsujiguchi, H.; Hara, A.; Shimizu, Y.; Hori, D.; Nguyen, T.T.T.; Suzuki, F.; Hamagishi, T.; Yamada, Y.; Nakamura, H.; et al. Increased Prevalence of Atopic Dermatitis in Children Aged 0–3 Years Highly Exposed to Parabens. *Int. J. Environ. Res. Public Health* **2021**, *18*, 11657. [CrossRef] [PubMed]
50. Langan, S.M.; Silcocks, P.; Williams, H.C. What Causes Flares of Eczema in Children? *Br. J. Dermatol.* **2009**, *161*, 640–646. [CrossRef]
51. Haarala, A.K.; Sinikumpu, S.-P.; Vaaramo, E.; Jokelainen, J.; Timonen, M.; Auvinen, J.; Pekkanen, J.; Huilaja, L. A Childhood Farm Environment Protects from Allergic Sensitization until Middle Age but Not from New-Onset Sensitization in Adulthood: A 15 Year Longitudinal Study. *Int. J. Environ. Res. Public Health* **2021**, *18*, 7078. [CrossRef]
52. Penders, J.; Gerhold, K.; Thijs, C.; Zimmermann, K.; Wahn, U.; Lau, S.; Hamelmann, E. New Insights into the Hygiene Hypothesis in Allergic Diseases: Mediation of Sibling and Birth Mode Effects by the Gut Microbiota. *Gut Microbes* **2014**, *5*, 239–244. [CrossRef] [PubMed]
53. Akdis, C.A.; Akdis, M.; Boyd, S.D.; Sampath, V.; Galli, S.J.; Nadeau, K.C. Allergy: Mechanistic Insights into New Methods of Prevention and Therapy. *Sci. Transl. Med.* **2023**, *15*, eadd2563. [CrossRef]
54. Herrant, M.; Loucoubar, C.; Boufkhed, S.; Bassène, H.; Sarr, F.D.; Baril, L.; Mercereau-Puijalon, O.; Mécheri, S.; Sakuntabhai, A.; Paul, R. Risk Factors Associated with Asthma, Atopic Dermatitis and Rhinoconjunctivitis in a Rural Senegalese Cohort. *Allergy Asthma Clin. Immunol.* **2015**, *11*, 24. [CrossRef] [PubMed]
55. Thyssen, J.P.; Ahluwalia, T.S.; Paternoster, L.; Ballardini, N.; Bergström, A.; Melén, E.; Chawes, B.L.; Stokholm, J.; Hourihane, J.O.; O'Sullivan, D.M.; et al. Interaction between Filaggrin Mutations and Neonatal Cat Exposure in Atopic Dermatitis. *Allergy* **2020**, *75*, 1481–1485. [CrossRef]
56. Langan, S.M.; Flohr, C.; Williams, H.C. The Role of Furry Pets in Eczema: A Systematic Review. *Arch. Derm.* **2007**, *143*, 1570–1577. [CrossRef] [PubMed]
57. Pelucchi, C.; Galeone, C.; Bach, J.-F.; La Vecchia, C.; Chatenoud, L. Pet Exposure and Risk of Atopic Dermatitis at the Pediatric Age: A Meta-Analysis of Birth Cohort Studies. *J. Allergy Clin. Immunol.* **2013**, *132*, 616–622.e7. [CrossRef] [PubMed]
58. Ruokolainen, L.; Paalanen, L.; Karkman, A.; Laatikainen, T.; von Hertzen, L.; Vlasoff, T.; Markelova, O.; Masyuk, V.; Auvinen, P.; Paulin, L.; et al. Significant Disparities in Allergy Prevalence and Microbiota between the Young People in Finnish and Russian Karelia. *Clin. Exp. Allergy* **2017**, *47*, 665–674. [CrossRef]
59. Haahtela, T.; Laatikainen, T.; Alenius, H.; Auvinen, P.; Fyhrquist, N.; Hanski, I.; von Hertzen, L.; Jousilahti, P.; Kosunen, T.U.; Markelova, O.; et al. Hunt for the Origin of Allergy—Comparing the Finnish and Russian Karelia. *Clin. Exp. Allergy* **2015**, *45*, 891–901. [CrossRef]
60. Wang, J.; Janson, C.; Malinovschi, A.; Holm, M.; Franklin, K.A.; Modig, L.; Johannessen, A.; Schlünssen, V.; Gislason, T.; Jogi, N.O.; et al. Asthma, Allergic Rhinitis and Atopic Dermatitis in Association with Home Environment—The RHINE Study. *Sci. Total Environ.* **2022**, *853*, 158609. [CrossRef]
61. Mahdavinia, M.; Greenfield, L.R.; Moore, D.; Botha, M.; Engen, P.; Gray, C.; Lunjani, N.; Hlela, C.; Basera, W.; Hobane, L.; et al. House Dust Microbiota and Atopic Dermatitis; Effect of Urbanization. *Pediatr. Allergy Immunol.* **2021**, *32*, 1006–1012. [CrossRef] [PubMed]
62. Levin, M.E.; Botha, M.; Basera, W.; Facey-Thomas, H.E.; Gaunt, B.; Gray, C.L.; Kiragu, W.; Ramjith, J.; Watkins, A.; Genuneit, J. Environmental Factors Associated with Allergy in Urban and Rural Children from the South African Food Allergy (SAFFA) Cohort. *J. Allergy Clin. Immunol.* **2020**, *145*, 415–426. [CrossRef]
63. Lunjani, N.; Tan, G.; Dreher, A.; Sokolowska, M.; Groeger, D.; Warwyzniak, M.; Altunbulakli, C.; Westermann, P.; Basera, W.; Hobane, L.; et al. Environment-dependent Alterations of Immune Mediators in Urban and Rural South African Children with Atopic Dermatitis. *Allergy* **2022**, *77*, 569–581. [CrossRef]
64. Thacher, J.D.; Gruzieva, O.; Pershagen, G.; Neuman, Å.; Wickman, M.; Kull, I.; Melén, E.; Bergström, A. Pre- and Postnatal Exposure to Parental Smoking and Allergic Disease Through Adolescence. *Pediatrics* **2014**, *134*, 428–434. [CrossRef]
65. Biagini Myers, J.M.; Khurana Hershey, G.K. Eczema in Early Life: Genetics, the Skin Barrier, and Lessons Learned from Birth Cohort Studies. *J. Pediatr.* **2010**, *157*, 704–714. [CrossRef] [PubMed]
66. Kantor, R.; Kim, A.; Thyssen, J.P.; Silverberg, J.I. Association of Atopic Dermatitis with Smoking: A Systematic Review and Meta-Analysis. *J. Am. Acad. Dermatol.* **2016**, *75*, 1119–1125.e1. [CrossRef]

67. Morales, E.; Strachan, D.; Asher, I.; Ellwood, P.; Pearce, N.; Garcia-Marcos, L. Combined Impact of Healthy Lifestyle Factors on Risk of Asthma, Rhinoconjunctivitis and Eczema in School Children: ISAAC Phase III. *Thorax* **2019**, *74*, 531–538. [CrossRef]
68. Jaffary, F.; Faghihi, G.; Mokhtarian, A.; Hosseini, S. Effects of Oral Vitamin E on Treatment of Atopic Dermatitis: A Randomized Controlled Trial. *J. Res. Med. Sci.* **2015**, *20*, 1053. [CrossRef]
69. Shin, J.; Kim, Y.J.; Kwon, O.; Kim, N.-I.; Cho, Y. Associations among Plasma Vitamin C, Epidermal Ceramide and Clinical Severity of Atopic Dermatitis. *Nutr. Res. Pract.* **2016**, *10*, 398. [CrossRef] [PubMed]
70. Oh, S.-Y.; Chung, J.; Kim, M.-K.; Kwon, S.O.; Cho, B.-H. Antioxidant Nutrient Intakes and Corresponding Biomarkers Associated with the Risk of Atopic Dermatitis in Young Children. *Eur. J. Clin. Nutr.* **2010**, *64*, 245–252. [CrossRef]
71. Cho, S.I.; Lee, H.; Lee, D.H.; Kim, K.-H. Association of Frequent Intake of Fast Foods, Energy Drinks, or Convenience Food with Atopic Dermatitis in Adolescents. *Eur. J. Nutr.* **2020**, *59*, 3171–3182. [CrossRef]
72. Kong, H.H.; Oh, J.; Deming, C.; Conlan, S.; Grice, E.A.; Beatson, M.A.; Nomicos, E.; Polley, E.C.; Komarow, H.D.; NISC Comparative Sequence Program; et al. Temporal Shifts in the Skin Microbiome Associated with Disease Flares and Treatment in Children with Atopic Dermatitis. *Genome Res.* **2012**, *22*, 850–859. [CrossRef] [PubMed]
73. Grice, E.A.; Kong, H.H.; Conlan, S.; Deming, C.B.; Davis, J.; Young, A.C.; NISC Comparative Sequencing Program; Bouffard, G.G.; Blakesley, R.W.; Murray, P.R.; et al. Topographical and Temporal Diversity of the Human Skin Microbiome. *Science* **2009**, *324*, 1190–1192. [CrossRef] [PubMed]
74. Bjerre, R.D.; Holm, J.B.; Palleja, A.; Sølberg, J.; Skov, L.; Johansen, J.D. Skin Dysbiosis in the Microbiome in Atopic Dermatitis Is Site-Specific and Involves Bacteria, Fungus and Virus. *BMC Microbiol.* **2021**, *21*, 256. [CrossRef] [PubMed]
75. Bisgaard, H.; Li, N.; Bonnelykke, K.; Chawes, B.L.K.; Skov, T.; Paludan-Müller, G.; Stokholm, J.; Smith, B.; Krogfelt, K.A. Reduced Diversity of the Intestinal Microbiota during Infancy Is Associated with Increased Risk of Allergic Disease at School Age. *J. Allergy Clin. Immunol.* **2011**, *128*, 646–652.e5. [CrossRef] [PubMed]
76. Namara, B.; Nash, S.; Lule, S.A.; Akurut, H.; Mpairwe, H.; Akello, F.; Tumusiime, J.; Kizza, M.; Kabagenyi, J.; Nkurunungi, G.; et al. Effects of Treating Helminths during Pregnancy and Early Childhood on Risk of Allergy-Related Outcomes: Follow-up of a Randomized Controlled Trial. *Pediatr. Allergy Immunol.* **2017**, *28*, 784–792. [CrossRef] [PubMed]
77. Lee, H.H.; Patel, K.R.; Singam, V.; Rastogi, S.; Silverberg, J.I. A Systematic Review and Meta-Analysis of the Prevalence and Phenotype of Adult-Onset Atopic Dermatitis. *J. Am. Acad. Dermatol.* **2019**, *80*, 1526–1532.e7. [CrossRef]
78. Niwa, Y.; Sumi, H.; Kawahira, K.; Terashima, T.; Nakamura, T.; Akamatsu, H. Protein Oxidative Damage in the Stratum Corneum: Evidence for a Link between Environmental Oxidants and the Changing Prevalence and Nature of Atopic Dermatitis in Japan. *Br. J. Derm.* **2003**, *149*, 248–254. [CrossRef]
79. Folster-Holst, R.; Pape, M.; Buss, Y.L.; Christophers, E.; Weichenthal, M. Low Prevalence of the Intrinsic Form of Atopic Dermatitis among Adult Patients. *Allergy* **2006**, *61*, 629–632. [CrossRef] [PubMed]
80. Tang, K.-T.; Ku, K.-C.; Chen, D.-Y.; Lin, C.-H.; Tsuang, B.-J.; Chen, Y.-H. Adult Atopic Dermatitis and Exposure to Air Pollutants—A Nationwide Population-Based Study. *Ann. Allergy Asthma Immunol.* **2017**, *118*, 351–355. [CrossRef] [PubMed]
81. Kantor, R.; Silverberg, J.I. Environmental Risk Factors and Their Role in the Management of Atopic Dermatitis. *Expert Rev. Clin. Immunol.* **2017**, *13*, 15–26. [CrossRef] [PubMed]
82. Rothenberg, M.E. The Climate Change Hypothesis for the Allergy Epidemic. *J. Allergy Clin. Immunol.* **2022**, *149*, 1522–1524. [CrossRef] [PubMed]
83. Kim, K. Influences of Environmental Chemicals on Atopic Dermatitis. *Toxicol. Res.* **2015**, *31*, 89–96. [CrossRef]
84. Tang, K.-T.; Chen, P.-A.; Lee, M.-R.; Lee, M.-F.; Chen, Y.-H. The Relationship between Exposure to Polycyclic Aromatic Hydrocarbons and Adult Atopic Dermatitis. *Asian Pac. J. Allergy Immunol.* **2020**. [CrossRef]
85. Hidaka, T.; Ogawa, E.; Kobayashi, E.H.; Suzuki, T.; Funayama, R.; Nagashima, T.; Fujimura, T.; Aiba, S.; Nakayama, K.; Okuyama, R.; et al. The Aryl Hydrocarbon Receptor AhR Links Atopic Dermatitis and Air Pollution via Induction of the Neurotrophic Factor Artemin. *Nat. Immunol.* **2017**, *18*, 64–73. [CrossRef]
86. Hendricks, A.J.; Eichenfield, L.F.; Shi, V.Y. The Impact of Airborne Pollution on Atopic Dermatitis: A Literature Review. *Br. J. Derm.* **2020**, *183*, 16–23. [CrossRef]
87. Kabashima, K.; Otsuka, A.; Nomura, T. Linking Air Pollution to Atopic Dermatitis. *Nat. Immunol.* **2017**, *18*, 5–6. [CrossRef]
88. Wesley, N.O.; Maibach, H.I. Racial (Ethnic) Differences in Skin Properties: The Objective Data. *Am. J. Clin. Dermatol.* **2003**, *4*, 843–860. [CrossRef] [PubMed]
89. Pesce, G.; Marcon, A.; Carosso, A.; Antonicelli, L.; Cazzoletti, L.; Ferrari, M.; Fois, A.G.; Marchetti, P.; Olivieri, M.; Pirina, P.; et al. Adult Eczema in Italy: Prevalence and Associations with Environmental Factors. *J. Eur. Acad. Derm. Venereol.* **2015**, *29*, 1180–1187. [CrossRef]
90. Nnoruka, E.N. Current Epidemiology of Atopic Dermatitis in South-Eastern Nigeria. *Int. J. Derm.* **2004**, *43*, 739–744. [CrossRef]
91. Silverberg, J.I.; Hanifin, J.M. Adult Eczema Prevalence and Associations with Asthma and Other Health and Demographic Factors: A US Population–Based Study. *J. Allergy Clin. Immunol.* **2013**, *132*, 1132–1138. [CrossRef] [PubMed]
92. Balić, A.; Vlašić, D.; Žužul, K.; Marinović, B.; Bukvić Mokos, Z. Omega-3 Versus Omega-6 Polyunsaturated Fatty Acids in the Prevention and Treatment of Inflammatory Skin Diseases. *Int. J. Mol. Sci.* **2020**, *21*, 741. [CrossRef] [PubMed]
93. Eriksen, B.B.; Kåre, D.L. Open Trial of Supplements of Omega 3 and 6 Fatty Acids, Vitamins and Minerals in Atopic Dermatitis. *J. Dermatol. Treat.* **2006**, *17*, 82–85. [CrossRef]

94. Ito, M.; Morita, T.; Okazaki, S.; Koto, M.; Ichikawa, Y.; Takayama, R.; Hoashi, T.; Saeki, H.; Kanda, N. Dietary Habits in Adult Japanese Patients with Atopic Dermatitis. *J. Derm.* **2019**, *46*, 515–521. [CrossRef] [PubMed]
95. Alfonso, F.; Goicolea, J.; Hernández, R.; Bañuelos, C.; Segovia, J.; Fernández-Ortiz, A.; Gonçalves, M.; Alonso, L.; Macaya, C. [Coronary angioscopy: Initial experience during coronary interventions]. *Rev. Esp. Cardiol.* **1995**, *48*, 798–806. [PubMed]
96. Park, S.; Bae, J.-H. Fermented Food Intake Is Associated with a Reduced Likelihood of Atopic Dermatitis in an Adult Population (Korean National Health and Nutrition Examination Survey 2012-2013). *Nutr. Res.* **2016**, *36*, 125–133. [CrossRef]
97. Silverberg, J.I.; Greenland, P. Eczema and Cardiovascular Risk Factors in 2 US Adult Population Studies. *J. Allergy Clin. Immunol.* **2015**, *135*, 721–728.e6. [CrossRef]
98. Lee, C.H.; Chuang, H.Y.; Hong, C.H.; Huang, S.K.; Chang, Y.C.; Ko, Y.C.; Yu, H.S. Lifetime Exposure to Cigarette Smoking and the Development of Adult-Onset Atopic Dermatitis: Smoking and Adult Atopic Dermatitis. *Br. J. Dermatol.* **2011**, *164*, 483–489. [CrossRef]
99. Pilz, A.C.; Schielein, M.C.; Schuster, B.; Heinrich, L.; Haufe, E.; Abraham, S.; Heratizadeh, A.; Harder, I.; Kleinheinz, A.; Wollenberg, A.; et al. Atopic Dermatitis: Disease Characteristics and Comorbidities in Smoking and Non-smoking Patients from the TREATgermany Registry. *Acad. Derm. Venereol.* **2022**, *36*, 413–421. [CrossRef]
100. Morra, D.E.; Cho, E.; Li, T.; Camargo, C.A.; Qureshi, A.A.; Drucker, A.M. Smoking and Risk of Adult-Onset Atopic Dermatitis in US Women. *J. Am. Acad. Dermatol.* **2021**, *84*, 561–563. [CrossRef]
101. Pilz, A.C.; Durner, V.; Schielein, M.C.; Schuster, B.; Beckmann, J.; Biedermann, T.; Eyerich, K.; Zink, A. Addictions in Patients with Atopic Dermatitis: A Cross-sectional Pilot Study in Germany. *J. Eur. Acad. Derm. Venereol.* **2022**, *36*, 84–90. [CrossRef]
102. Pondeljak, N.; Lugović-Mihić, L. Stress-Induced Interaction of Skin Immune Cells, Hormones, and Neurotransmitters. *Clin. Ther.* **2020**, *42*, 757–770. [CrossRef]
103. Seiffert, K.; Hilbert, E.; Schaechinger, H.; Zouboulis, C.C.; Deter, H.-C. Psychophysiological Reactivity under Mental Stress in Atopic Dermatitis. *Dermatology* **2005**, *210*, 286–293. [CrossRef]
104. Kodama, A.; Horikawa, T.; Suzuki, T.; Ajiki, W.; Takashima, T.; Harada, S.; Ichihashi, M. Effect of Stress on Atopic Dermatitis: Investigation in Patients after the Great Hanshin Earthquake. *J. Allergy Clin. Immunol.* **1999**, *104*, 173–176. [CrossRef] [PubMed]
105. Sanders, K.M.; Akiyama, T. The Vicious Cycle of Itch and Anxiety. *Neurosci. Biobehav. Rev.* **2018**, *87*, 17–26. [CrossRef] [PubMed]
106. Silverberg, J.I.; Garg, N.K.; Paller, A.S.; Fishbein, A.B.; Zee, P.C. Sleep Disturbances in Adults with Eczema Are Associated with Impaired Overall Health: A US Population-Based Study. *J. Investig. Dermatol.* **2015**, *135*, 56–66. [CrossRef] [PubMed]
107. Steinhoff, M.; Suárez, A.; Feramisco, J.; Koo, J. Psychoneuroimmunology of Psychological Stress and Atopic Dermatitis: Pathophysiologic and Therapeutic Updates. *Acta Derm. Venerol.* **2012**, *92*, 7–15. [CrossRef]
108. Yosipovitch, G.; Tran, B.; Papoiu, A.; Russoniello, C.; Wang, H.; Patel, T.; Chan, Y. Effect of Itch, Scratching and Mental Stress on Autonomic Nervous System Function in Atopic Dermatitis. *Acta Derm. Venerol.* **2010**, *90*, 354–361. [CrossRef]
109. Hurley, S.; Franklin, R.; McCallion, N.; Byrne, A.M.; Fitzsimons, J.; Byrne, S.; White, M.; O'Mahony, L.; Hourihane, J.O. Allergy-related Outcomes at 12 Months in the CORAL Birth Cohort of Irish Children Born during the First COVID 19 Lockdown. *Pediatr. Allergy Immunol.* **2022**, *33*, e13766. [CrossRef]
110. Singh, M.; Pawar, M.; Bothra, A.; Choudhary, N. Overzealous Hand Hygiene during the COVID 19 Pandemic Causing an Increased Incidence of Hand Eczema among General Population. *J. Am. Acad. Dermatol.* **2020**, *83*, e37–e41. [CrossRef]
111. Pecoraro, L.; Chiaffoni, G.; Piacentini, G.; Pietrobelli, A. The Need of an Updated Culture of "Occupational" Atopic Hand Dermatitis in Children at the Time of COVID-19. *Acta Biomed.* **2022**, *93*, e2022324. [CrossRef] [PubMed]
112. Rundle, C.W.; Presley, C.L.; Militello, M.; Barber, C.; Powell, D.L.; Jacob, S.E.; Atwater, A.R.; Watsky, K.L.; Yu, J.; Dunnick, C.A. Hand Hygiene during COVID-19: Recommendations from the American Contact Dermatitis Society. *J. Am. Acad. Dermatol.* **2020**, *83*, 1730–1737. [CrossRef] [PubMed]
113. Pereira, P.; Bašić, F.; Bogunovic, I.; Barcelo, D. Russian-Ukrainian War Impacts the Total Environment. *Sci. Total Environ.* **2022**, *837*, 155865. [CrossRef]
114. Riegleman, K.L.; Farnsworth, G.S.; Wong, E.B. Atopic Dermatitis in the US Military. *Cutis* **2019**, *104*, 144–147. [PubMed]
115. The Lancet Ukraine's Humanitarian Disaster: Priorities for Health. *Lancet* **2022**, *399*, 1023. [CrossRef]

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Is Dupilumab as Effective in Intrinsic Atopic Dermatitis as It Is in Extrinsic Atopic Dermatitis?

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Abstract: Atopic dermatitis (AD) can be subclassified into the more frequent extrinsic type (EAD), with elevated serum IgE levels and frequent association with other atopic conditions, and the less frequent intrinsic type (IAD), with normal IgE levels and no history of atopy. This retrospective study has the objective to compare the efficacy of dupilumab therapy in patients with IAD versus EAD in a real-life setting. We studied a group of 360 patients treated with dupilumab for moderate-to-severe AD of whom 49 had IAD (IgE < 200 kU/L and no history of other atopic conditions) and 311 had EAD (IgE $\geq$ 200 kU/L and/or history of atopy). There were no statistically significant differences in the achievement of EASI75 between IAD and EAD patients either at 16, 32, or 48 weeks (61% vs. 50%; 66% vs. 60%; and 53% vs. 65%, respectively). Similarly, there were no statistically significant differences in the achievement of EASI90 or the reduction in NRSpp, NRSsd, and DLQI at each timepoint. Additionally, mean absolute eosinophils and IgE values were significantly higher in the EAD group at all timepoints. This study confirms that dupilumab, targeting the Th2 pathway, which is known to be overexpressed in all AD phenotypes, appears to be equally effective in the two populations regardless of IgE levels.

Keywords: atopic dermatitis; dupilumab; intrinsic atopic dermatitis

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1. Introduction

Atopic dermatitis (AD) is a common inflammatory disease of the skin characterized by intense pruritus and chronic or relapsing eczematous lesions. The Global Burden of Disease study showed a prevalence of 15–20% among children and up to 10% among adults with a notable impact on the quality of life of patients and their families and with significant economic repercussions [1].

AD is frequently associated with a personal or family history of atopic conditions, such as bronchial asthma and allergic rhinitis, which is why the term "atopic dermatitis" was chosen [2,3].

In 2003, the Nomenclature Review Committee of the World Allergy Organization proposed a new nomenclature for allergies in which the term "atopy" refers to a genetic predisposition to become immunoglobulin E (IgE) sensitized to allergens commonly found in the environment to which everyone is exposed but to which most people do not produce a long-term IgE antibody response [4].

Most patients diagnosed with AD have high concentrations of total IgE, positive allergen-specific IgE levels, and positive skin prick tests with aeroallergens and/or food allergens. However, there are patients with similar clinical features of AD but without detectable sensitization to inhalant or food allergens [5]. For this reason, among patients with a clinical diagnosis of AD, a distinction into two subgroups has been proposed. Similar to the extrinsic and intrinsic types of asthma, the term "intrinsic AD" (IAD) and "extrinsic

AD" (EAD) have been suggested. For IAD, other similar terms such as "non-allergic AD" or "non-atopic eczema" have been introduced to describe AD patients without allergen-specific IgE [4,6]. Since these patients have no demonstrable atopy, the term "atopiform dermatitis" has also been coined to avoid the link with true atopy [7].

Since total serum IgE values are correlated with the allergen-specific IgE status, total IgE can be regarded as a clinically useful parameter to expectedly differentiate between the extrinsic type, with increased IgE levels, and the intrinsic type, with normal IgE values, in both adults and children [5,8].

The prevalence of IAD has been reported between 15% and 45% in different studies, and it is characterized by a female predominance, a later age at onset, and a more frequent distribution in the head and neck area (H&N) [5,9]. Instead, no histological differences were observed in the two groups. The typical findings are similar in both types of AD: epidermal spongiosis and dermal lymphocytic infiltrate in the acute-phase lesions and acanthosis in the chronic phase. Activated T cells predominate the dermal cell infiltrate, but other cells such as mast cells, eosinophils, and antigen-presenting dendritic cells are also present. In IAD and EAD, different cytokine patterns of involved T cells have been observed in peripheral blood, as well as in lesional skin biopsies. EAD patients have been found to express elevated levels of interleukin (IL)-4 and IL-13 with an increased production of IgE compared to patients with IAD [5]. In addition, patients with IAD were found to have a greater increase in the Th1 signal and more pronounced Th17/Th22 activation [10].

Taking this different cytokine production into account, we wondered whether dupilumab, an inhibitor of both IL-4 and IL-13 signaling, has less efficacy in patients with IAD.

In the literature, very few studies have evaluated the efficacy of dupilumab by stratifying patients according to normal versus increased IgE values, following a "one-size-fits-all" treatment approach [11,12]. With this study, we want to enrich the literature by comparing the efficacy of dupilumab in patients with IAD versus EAD in a real-life setting.

2. Materials and Methods

2.1. Study Population

We performed a retrospective observational study with the aim of comparing the efficacy of dupilumab therapy in patients with EAD versus IAD in a real-life setting.

Patients aged ≥12 years with moderate-to-severe AD who started treatment with dupilumab at the Dermatology Clinic of the Turin University Hospital between January 2019 and March 2022 were included in this study.

AD was diagnosed according to the Hanifin and Rajka criteria [13], and IAD was defined by total serum IgE levels < 200 kU/L and the absence of personal or family history of other atopic conditions such as bronchial asthma and allergic rhino-conjunctivitis according to Schmid (-Grendelmeier) et al. [5]. Adult patients had to have an EASI (eczema area severity index) ≥ 24, while adolescents required an EASI ≥ 24 or one of the following criteria: localization in sensitive or visible areas, an NRSpp (numerical rating scale peak of pruritus) ≥ 7, or a CDLQI (children's dermatology life quality index) ≥ 10.

All adult patients received an initial dose of 600 mg dupilumab and subsequently 300 mg every other week administered as subcutaneous injection. Adolescents (aged 12–17), if they weighed less than 60 kg, received an initial dose of 400 mg and then 200 mg every other week, whereas if they weighed more than 60 kg, the same dosage was used as for adults. Patients were examined before starting dubilumab therapy (T0) and were reevaluated at 16 (T1), 32 (T2), and 48 (T3) weeks. At T0, demographic characteristics, such as age, sex, BMI, age of onset, family history of atopy, predisposition to allergic conjunctivitis and recurrent herpetic recurrences, and history of parasitic infections, were collected. At each visit, blood count, leukocyte formula (with particular attention to eosinophils), IgE, and LDH were assessed; patients completed the DLQI (dermatology life quality index) or the CDLQI and a POEM (patient-oriented eczema measure) before each visit. Disease severity was assessed by EASI, with special attention to H&N EASI; pruritus

was assessed as NRSpp and NRSsd (sleep disturbance). The occurrence of adverse events (EAs) was also monitored at each visit.

2.2. Objectives

The aim of this study was the comparison of the efficacy of dupilumab therapy in patients with IAD versus EAD. These two groups were evaluated on the basis of improvements in EASI scores and H&N EASI at each T, the proportion of patients achieving EASI75 and EASI90 at each T, the percentage change from the baseline in worst pruritus NRS and NRSsd at each T, and improving quality of life by comparing DLQI and POEM scores at each T. LDH and eosinophil values were also compared in the two groups at each T.

2.3. Statistical Analysis

Quantitative variables were described by means with standard deviations (SD), while qualitative variables by percentages and absolute values.

Specifically, statistical analysis was performed using the Mann–Whitney U-test to compare independent variables with non-normal distributions.

For categorical variables, the Chi-square test was used, applying linear regression with Pearson's index for correlations. Statistical significance was considered as a p-value < 0.05.

All analyses were performed with STATA software version 10 (STATA Corporation, College Station, TX, USA).

3. Results

At the baseline, 360 patients were included in this study, of which 49 (14%) patients had IAD and 311 (86%) had EAD. Among the IAD patients, 25 (52%) were female and 24 (49%) were male, while the among the EAD patients, 133 (43%) and 178 (57%), respectively.

As shown in Table 1, in the group of patients with IAD, the mean EASI at baseline was 18.2 (SD ± 11.97) and 24.04 (SD ± 10.34) in the EAD group (p-value < 0.01); furthermore, in the IAD group, there was an H&N EASI of 2.6 (SD ± 2.09), while in the EAD group, it was 3.33 (SD ± 2.1) (p-value < 0.05). At T0, patients with EAD had significantly higher mean IgE values than those with IAD (3949.74 ± 5208.77 vs. 70.64 ± 60.05, p < 001), as expected according to the definition of the two groups. At the baseline, there was also no statistically significant difference between the mean DLQI of the IAD group (14.13 ± 7.56) and the mean DLQI of the EAD group (14.88 ± 7.07) (p-value 0.496). There were also no differences in the POEM; the mean value reported by IAD patients was 19.19 ± 6.56, while in the EAD group, it was 20.8 ± 6.31 (p-value 0.094).

Table 1. Patents' baseline characteristics.

	IAD (n = 49)	**EAD (n = 311)**	***p*-Value**
EASI score (mean ± SD)	18.2 ± 11.97	24.04 ± 10.34	<0.01
H&N EASI score (mean ± SD)	2.6 ± 2.09	3.33 ± 2.1	<0.05
DLQI (mean ± SD)	14.13 ± 7.56	14.88 ± 7.07	0.496
POEM (mean ± SD)	19.19 ± 6.56	20.8 ± 6.31	0.094
NRSpp (mean ± SD)	8.63 ± 1.44	8.56 ± 1.75	0.759
NRSsd (mean ± SD)	6.85 ± 2.83	7.07 ± 3.15	0.258
LDH (mean ± SD)	289 ± 125.42	327.12 ± 147.16	0.095
IgE kU/L (mean ± SD)	70.64 ± 60.05	3949.74 ± 5208.77	<0.001
Eosinophils × 10^9/L (mean ± SD)	0.31 ± 0.39	0.2 ± 9.07	<0.01

IAD: Intrinsic atopic dermatitis; EAD: extrinsic atopic dermatitis; SD: standard deviation; EASI: eczema area severity index; H&N = head and neck; DLQI: dermatology life quality index; POEM: patient-oriented eczema measure; NRSpp: numerical rating scale peak of pruritus; NRSsd: numerical rating scale sleep disturbance.

Among patients included in this study, 38 IAD patients and 234 EAD patients reached T1, at which time the mean EASI was 2.78 (SD ± 5.62) in the IAD group and 3.88 (SD ± 5.13) in the EAD group (*p*-value 0.028). At T2, 35 IAD patients had a mean EASI of 1.79 (SD ± 2.97), and 212 EAD patients had a mean EASI of 2.96 (SD ± 3.78) (*p*-value 0.011). At T3, 30 IAD patients had a mean EASI of 1.61 (SD ± 1.6), and 160 EAD patients had a mean EASI of 2.75 (SD ± 4.04) (*p*-value 0.222).

Thus, patients with IAD showed lower mean EASI values at T0, T1, and T2 than patients with EAD, whereas at T3, this difference nullifies.

There were no statistically significant differences between the two groups in the achievement of EASI75 at either T1, T2, or T3. The percentage of patients achieving EASI75 in patients with IAD versus patients with EAD was 61% vs. 50% (*p*-value 0.248) at T1, 66% vs. 60% (*p*-value 0.514) at T2, and 53% vs. 65% (*p*-value 0.224) at T3.

Similarly, there was no statistically significant difference in the achievement of EASI90 in patients with IAD and AED: 42% vs. 35% (*p*-value 0.352) at T1, 57% vs. 40% (*p*-value 0.059) at T2, and 40% vs. 47% (*p*-value 0.488) at T3.

Figures 1 and 2 show, respectively, the trend of EASI75 and EASI90 at various time-points. Patients with IAD show a faster response up to T2 and then reverse the trend compared to EAD.

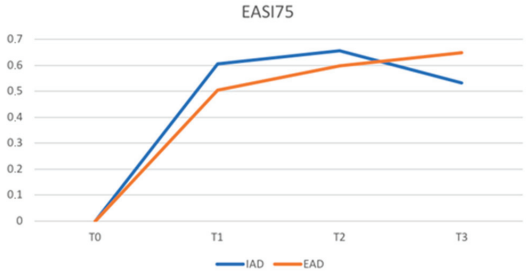

Figure 1. Proportion of patients achieving a 75% improvement in the eczema area and severity index (EASI75) (IAD = intrinsic atopic dermatitis; EAD = extrinsic atopic dermatitis).

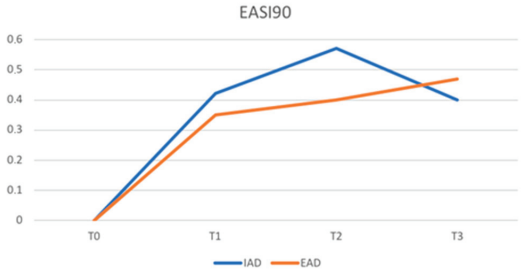

Figure 2. Proportion of patients achieving a 90% improvement in the EASI (EASI90) (IAD = intrinsic atopic dermatitis; EAD = extrinsic atopic dermatitis).

The mean H&N EASI in patients with IAD compared to those with EAD was 0.54 (SD ± 0.99) vs. 0.98 (SD ± 1.28) (*p*-value 0.17) at T1, 0.39 (SD ± 0.68) vs. 0.76 (SD ± 1.12) (*p*-value 0.46) at T2, and 0.38 (SD ± 0.62) vs. 0.73 (SD ± 1.02) (*p*-value 0.78) at T3.

With regard to itch scores, the mean NRSpp in IAD patients was 2.74 (SD ± 2.58) and 3.02 (SD ± 3.02) in EAD patients at T1 (*p*-value 0.50), 2.3 (SD ± 2.51) vs. 2.84 (SD ± 2.27) at T2 (*p*-value 0.112), and 2.54 (SD ± 2.10) vs. 2.66 (SD ± 2.33) at T3 (*p*-value 0.998).

At T1, the mean NRSsd in IAD patients was 1.06 (SD ± 2.03) and 1.15 (SD ± 2.2) in EAD patients (*p*-value 0.759). The mean NRSsd in IAD vs. EAD patients was 2.3 (SD ± 2.51) vs. 2.84 (SD ± 1.68) at T2 (*p*-value 0.781) and was 0.75 (SD ± 2.15) vs. 0.55

(SD ± 1.68) at T3 (*p*-value 0.781). The reduction trend in NRSpp and NRSsd in the two study groups is shown in Figures 3 and 4, respectively.

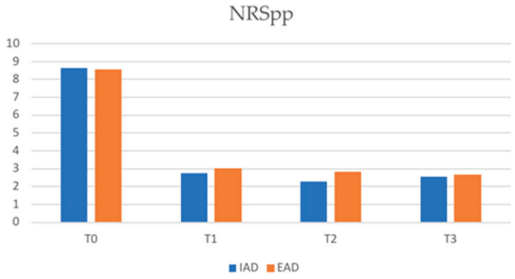

Figure 3. Numerical rating scale pick pruritus (NRSpp) in IAD and EAD patients (IAD = intrinsic atopic dermatitis; EAD = extrinsic atopic dermatitis).

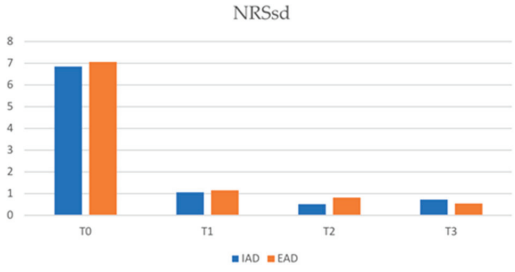

Figure 4. Numerical rating scale sleep disturbance (NRSsd) in IAD and EAD patients (IAD = intrinsic atopic dermatitis; EAD = extrinsic atopic dermatitis).

As for quality of life, the DLQI score was 4.79 ± 5.19 in the IAD group vs. 4.44 ± 5.01 in the EAD group at T1 (*p*-value 0.522), 3.88 ± 4.75 vs. 4.00 ± 4.99 at T2 (*p*-value 0.663) and 3.14 ± 2.86 vs. 3.24 ± 3.98 at T3 (*p*-value 0.679).

The percentage of patients with EAD who achieved a DLQI of 1/0 compared to those with IAD was: 75.24% vs. 77.55% at T1, 64.31% vs. 69.39% at T2, and 48.55% vs. 57.14% at T3 (Figure 5). No statistically significant differences were found in the achievement of a DLQI of 1/0 in the two study groups (*p*-value > 0.05).

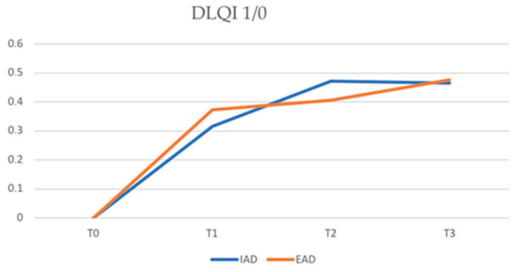

Figure 5. Proportion of patients achieving a DLQI of 1/0 in IAD and EAD. DLQI = dermatology life quality index (IAD = intrinsic atopic dermatitis; EAD = extrinsic atopic dermatitis).

Mean IgE values were significantly higher, although decreasing over time, in the EAD group than in the IAD group at all timepoints (2159.10 ± 3243.13 vs. 44.81 ± 40.30 at T1, 1348.92 ± 1816.90 vs. 38.85 ± 39.39 at T2, and 997.23 ± 1376.00 vs. 23.69 ± 16.72 at T3; $p < 0.001$).

The mean absolute values of eosinophils in the two groups were also compared and were statistically higher in patients with EAD than in those with IAD at all timepoints

(Figure 6). Values were 1.13 ± 9.07 × 10^9/L vs. 0.31 ± 0.39 × 10^9/L at T0 ($p < 0.001$), 0.68 ± 0.75 × 10^9/L vs. 0.45 ± 1.24 × 10^9/L at T1 ($p < 0.001$), and 0.65 ± 1.05 vs. 0.17 ± 0.11 × 10^9/L at T2 ($p < 0.001$).

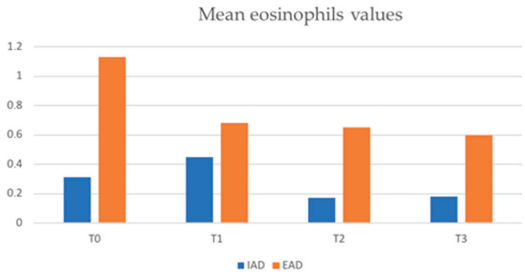

Figure 6. Mean eosinophils values (×10^9/L) in IAD and EAD patients (IAD = intrinsic atopic dermatitis; EAD = extrinsic atopic dermatitis).

With regard to mean LDH values, no statistically significant differences were found in the two groups at any timepoint.

4. Discussion

We evaluated the possible difference between the H&N EASI in the two groups as this parameter appears to have an important impact in measuring quality of life compared to other body area EASIs [14]. In contrast to what has previously been shown by other studies [5,9], our patients with EAD had a higher mean H&N EASI than patients with IAD at the baseline. During treatment with dupilumab, these values remained higher in the EAD group, although without a statistically significant difference and with an irregular trend, probably due to red face events.

The mean EASI values were significantly higher in patients with EAD at T0 as well as at T1 and T2; only at T3, after 48 weeks of treatment, was this difference no longer significant. Therefore, patients with EAD in our sample show a higher disease burden at the baseline that remains higher than patients with IAD in the first 32 weeks of dupilumab treatment. Nevertheless, the percentage of patients achieving an EASI75 is high in both the IAD and EAD groups (61% vs. 50% at T1, 66% vs. 60% at T2, and 53% vs. 65% at T3), and no statistically significant differences were found in the two groups. Patients with IAD showed a faster response up to T2 and then reversed the trend compared to EAD. Similarly, patients in the two groups achieved an EASI90 in comparable percentages, demonstrating equal efficacy of dupiluamb in patients with IAD and EAD.

It is well known that AD is a Th2-type inflammatory disease. However, patients with EAD present high levels of type 2 cytokines, including IL-4, IL-5, and IL-13, with an increased eosinophil count, whereas the levels of these cytokines are relatively lower, although increased compared to normal, in IAD. The latter is also characterized by increased Th1 signaling and more pronounced Th17/Th22 activation with an overproduction of IFN-g that may further downregulate IgE production [10].

These mechanisms may explain the higher, since baseline, serum eosinophil levels and the mean EASI values in EAD compared to IAD obtained in our study.

In our study, no statistically significant differences between the two groups of patients were recorded regarding the assessment of quality of life, both in terms of the reduction in DLQI and the achievement of a DLQI of 1/0 or in the reduction in NRSpp and NRSsd. This shows that dupilumab is responsible for an improvement in the patients' quality of life and a reduction in itching in both AD subtypes.

Since increased Th2 levels are a common feature across the AD spectrum, targeting this axis should theoretically be beneficial for all AD phenotypes [9].

This study confirms that dupilumab, by reducing IL-4 and IL-13 signaling, Th2-associated interleukins, appears to be equally as effective in the two study populations regardless of IgE levels, as previously shown by Hamilton et al. [11].

It can also be seen that the efficacy of dupilumab is not closely related to the reduction in IgE values; in fact, these remain above normal in patients with EAD despite an optimal clinical response in terms of both the EASI and pruritus reduction [15].

The main limitation of this study is the retrospective methodology. Certainly, larger prospective studies will be needed to confirm the equal efficacy of dupilumab in patients with IAD versus EAD and to further investigate such a topic.

The stratification of AD endotypes could be important for developing personalized medicine approaches that can potentially improve therapeutic outcomes. Indeed, patients with IAD also show significant Th17 and, in some cases, Th22 activation, with increased levels of the relevant cytokines (IL17/IL23 and IL22, respectively). Therefore, such patients could benefit from targeted treatment of these two cytokine axes [9,16].

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Informed Consent Statement: Informed consent was obtained from all subjects involved in this study.

Data Availability Statement: The data presented in this study are available on request from the corresponding author. The data are not publicly available due to privacy.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Laughter, M.R.; Maymone, M.B.; Mashayekhi, S.; Arents, B.W.; Karimkhani, C.; Langan, S.M.; Dellavalle, R.P.; Flohr, C. The global burden of atopic dermatitis: Lessons from the Global Burden of Disease Study 1990–2017. *Br. J. Dermatol.* **2021**, *184*, 304–309. [CrossRef] [PubMed]
2. Langan, S.M.; Irvine, A.D.; Weidinger, S. Atopic dermatitis. *Lancet* **2020**, *396*, 345–360. [CrossRef] [PubMed]
3. Mastorino, L.; Duò, V.C.; Vecco, C.; Gelato, F.; Giordano, S.; Roccuzzo, G.; Cavaliere, G.; Avallone, G.; Ortoncelli, M.; Ribero, S.; et al. Impact of comorbidities in the response of atopic patients treated with dupilumab: A real-life study up to 36 weeks. *J. Eur. Acad. Dermatol. Venereol.* **2022**, *36*, e1021–e1023. [CrossRef] [PubMed]
4. Johansson, S.G.; Bieber, T.; Dahl, R.; Friedmann, P.S.; Lanier, B.Q.; Lockey, R.F.; Motala, C.; Martell, J.A.; Platts-Mills, T.A.; Ring, J.; et al. Revised nomenclature for allergy for global use: Report of the Nomenclature Review Committee of the World Allergy Organization, October 2003. *J. Allergy Clin. Immunol.* **2004**, *113*, 832–836. [CrossRef] [PubMed]
5. Schmid-Grendelmeier, P.; Simon, D.; Simon, H.U.; Akdis, C.A.; Wuthrich, B. Epidemiology, clinical features, and immunology of the "intrinsic" (non-IgE-mediated) type of atopic dermatitis (constitutional dermatitis). *Allergy* **2001**, *56*, 841–849. [CrossRef] [PubMed]
6. Roguedas-Contios, A.M.; Misery, L. What is Intrinsic Atopic Dermatitis? *Clin. Rev. Allerg. Immunol.* **2011**, *41*, 233–236. [CrossRef] [PubMed]
7. Brenninkmeijer, E.E.A.; Spuls, P.I.; Legierse, C.M.; Lindeboom, R.; Smitt, J.H.S.; Bos, J.D. Clinical differences between atopic and atopiform dermatitis. *J. Am. Acad. Dermatol.* **2008**, *58*, 407–414. [CrossRef] [PubMed]
8. Ott, H.; Stanzel, S.; Ocklenburg, C.; Merk, H.; Baron, J.; Lehmann, S. Total Serum IgE as a Parameter to Differentiate between Intrinsic and Extrinsic Atopic Dermatitis in Children. *Acta Derm. Venerol.* **2009**, *89*, 257–261. [CrossRef] [PubMed]
9. Czarnowicki, T.; He, H.; Krueger, J.G.; Guttman-Yassky, E. Atopic dermatitis endotypes and implications for targeted therapeutics. *J. Allergy Clin. Immunol.* **2019**, *143*, 1–11. [CrossRef] [PubMed]
10. Tokura, Y.; Hayano, S. Subtypes of atopic dermatitis: From phenotype to endotype. *Allergol. Int.* **2022**, *71*, 14–24. [CrossRef] [PubMed]
11. Hamilton, J.D.; Suárez-Farinas, M.; Dhingra, N.; Cardinale, I.; Li, X.; Kostic, A.; Ming, J.E.; Radin, A.R.; Krueger, J.G.; Graham, N.; et al. Dupilumab improves the molecular signature in skin of patients with moderate-to-severe atopic dermatitis. *J. Allergy Clin. Immunol.* **2014**, *134*, 1293–1300. [CrossRef] [PubMed]

12. Beck, L.A.; Thaçi, D.; Hamilton, J.D.; Graham, N.M.; Bieber, T.; Rocklin, R.; Ming, J.E.; Ren, H.; Kao, R.; Simpson, E.; et al. Dupilumab Treatment in Adults with Moderate-to-Severe Atopic Dermatitis. *N. Engl. J. Med.* **2014**, *371*, 130–139. [CrossRef] [PubMed]
13. Hanifin, J.M.; Rajka, G. Diagnostic features of atopic dermatitis. *Acta Derm.* **1980**, *92*, 44–47. [CrossRef]
14. Ribero, S.; Puglisi, B.; Giura, M.T.; Viola, R.; Siliquini, N.; Quaglino, P.; Ortoncelli, M. Head and neck severity index is associated to a significant worsening of quality of life in atopic dermatitis patients. *Exp Dermatol.* **2021**, *30*, 1717–1718. [CrossRef] [PubMed]
15. Mastorino, L.; Rosset, F.; Gelato, F.; Ortoncelli, M.; Cavaliere, G.; Quaglino, P.; Ribero, S. Chronic Pruritus in Atopic Patients Treated with Dupilumab: Real Life Response and Related Parameters in 354 Patients. *Pharmaceuticals* **2022**, *15*, 883. [CrossRef] [PubMed]
16. Suárez-Fariñas, M.; Dhingra, N.; Gittler, J.; Shemer, A.; Cardinale, I.; de Guzman Strong, C.; Krueger, J.G.; Guttman-Yassky, E. Intrinsic atopic dermatitis shows similar TH2 and higher TH17 immune activation compared with extrinsic atopic dermatitis. *J. Allergy Clin. Immunol.* **2013**, *132*, 361–370. [CrossRef] [PubMed]

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Impact of the Family and Household Environment on Pediatric Atopic Dermatitis in Japan

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Abstract: Pediatric atopic dermatitis (AD) can negatively impact the family quality of life (QoL). We report data from the real-world Epidemiology of Children with Atopic Dermatitis Reporting on their Experience (EPI-CARE) study in Japanese pediatric patients, focusing on disease impact on family QoL. Children and adolescents aged 6 months to <18 years completed an online survey between September 2018–December 2019. The impact of disease severity on family QoL and its effect on parents' time were assessed using the dermatitis family impact (DFI) questionnaire. The impact of a family history of allergic conditions, current residency, second-hand smoke exposure, and household pets on AD prevalence and severity was also assessed. Family QoL decreased as AD severity increased, particularly in families with children aged <6 years; but had the greatest impact on sleep and tiredness in families with children aged <12 years. Parents spent at least 4.6 h/week caring for children <6 years, including those with mild symptoms. Most children (>80%) had a family history of allergic conditions; AD prevalence was increased in those exposed to second-hand smoke or household pets. This study demonstrated that pediatric AD in Japanese individuals has negative impacts on family QoL and that family and household environments can influence pediatric AD prevalence.

Keywords: atopic; child; dermatitis; family; quality of life

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1. Introduction

Atopic dermatitis (AD) is a chronic, relapsing skin condition characterized by pruritus and disruption of the epithelial barrier, with symptoms often appearing within the first 5 years of life and persisting into adolescence and adulthood [1,2].

The pathophysiology of AD involves a complex interaction between genetic epithelial skin barrier defects, dysregulated immune response, an altered microbiome, and environmental risk factors [3]. In particular, patients with atopic diathesis are genetically predisposed to develop one or more atopic diseases, including AD, hay fever, allergic rhinitis, and asthma [4]. In addition, the patient's exposome, a concept that encompasses all environmental exposure (e.g., chemical pollution, tobacco smoke, infectious agents, or lifestyle factors) encountered by an individual throughout their life, is thought to play an important role in AD development [5–9].

AD in childhood and adolescence has a negative impact on the quality of life (QoL) of the parents and caregivers of affected children [10–13]. Moderate-to-severe AD in children is often associated with a significant symptom burden, including pruritus, pain, and sleep disturbance, which may result in poor self-esteem, reduced school performance, and increased familial stress [14]. Parents of children with AD often report reduced QoL related

to sleep disturbances, daytime functioning, and financial problems [10–12]. A multinational survey on the impact of AD on family QoL reported that caring for a child with moderate-to-severe AD was associated with high levels of parental anxiety and exhaustion, as well as strained family relationships [10]. Life decisions for caregivers and families of children with AD are also negatively impacted, particularly in children with severe disease [15].

In addition to the negative impact of AD on families, the prevalence or severity of AD may be affected by several factors, including a family history of allergic conditions [16], second-hand smoke exposure [17], and the presence of domestic pets [18]. However, few studies have evaluated the impact of pediatric AD on family QoL or the impact of the family and household environment on AD prevalence and severity in Japan.

The Epidemiology of Children with Atopic Dermatitis Reporting on their Experience (EPI-CARE) study evaluated the prevalence and burden of AD among the pediatric population (aged 6 months to <18 years) and their caregivers in countries from several geographic regions worldwide, including Japan [19]. Data from the EPI-CARE study have indicated that parents and caregivers experience an impact of their child's AD on family QoL, particularly in children with higher disease severity [13]. In the Japanese EPI-CARE pediatric population (n = 5702), the 12-month prevalence of diagnosed AD (based on meeting the International Study of Asthma and Allergies in Childhood (ISAAC) criteria [20] and having a physician-diagnosed AD) was 10.7% overall, 12.9% in patients aged <6 years, 10.3% in those aged 6 to <12 years, and 9.1% in those aged 12 to <18 years [19].

Here, we report additional results of the EPI-CARE study in the Japanese pediatric population, focusing on the impact of pediatric AD on family QoL and the influence of the family and the household environment on its prevalence and severity.

2. Materials and Methods

2.1. Study Design

The design of the cross-sectional, epidemiologic EPI-CARE study has been described previously [19]. In addition, EPI-CARE was conducted across 18 countries to collect representative data from pediatric populations with AD. The study received ethical approval and was performed in accordance with the European Union General Data Protection Regulation, the European Society for Opinion and Marketing Research, the Insights Association, the European Pharmaceutical Marketing Research Association, the British Healthcare Business Intelligence Association, the US Health Insurance Portability and Accountability Act, and all international and local data protection legislation. All participants or their parents provided written informed consent before study entry. No personally identifiable information or medical data were collected.

A web-based survey was used for data collection, and the parents of participants were recruited via direct emailing, special interest websites, and broad-reach portals. Kantar Health was responsible for participant recruitment, administration of surveys, collation of responses, and data analyses. The survey was conducted in Japan between 26 September 2018, and 2 December 2019.

2.2. Study Population

The study included children and adolescents aged 6 months to <18 years. There were no other specific study inclusion/exclusion criteria.

Initially, parents of eligible children and adolescents were recruited via email and participated in an online panel; panel members were blinded to the research topic when invited and received points once they completed the survey that could be redeemed for items in a prize catalog (exact values unknown). After initial recruitment, the parents of children aged <12 years completed the survey on behalf of their children, and adolescents aged 12 to <18 years completed the survey themselves.

2.3. Questionnaire and Outcomes

The 30-min online, web-based survey included two sections. In the first section, a selection algorithm was used to determine which children were to be investigated (in cases where parents had multiple children), and demographic data were collected. Three definitions of AD were used: 'reported AD,' 'physician-diagnosed AD,' and 'diagnosed AD'. For 'reported AD' responders had to exclusively meet all three ISAAC criteria (Table 1) [20]. For 'physician-diagnosed AD' responders self-reported having ever been told by a physician that they had AD. For 'diagnosed AD' responders met all three ISAAC criteria and had self-reported having ever been told by a physician that they had AD.

Table 1. International Study of Asthma and Allergies in Childhood Criteria for atopic dermatitis [20].

Criteria	Description
1	An itchy rash that has come and gone for ≥6 months
2	The itchy rash has appeared at any time in the past 12 months
3	The itchy rash has affected any of the following places at any time: • Folds of the elbows • Behind the knees • In front of the ankles • Under the buttocks • Around the neck, ears, or eyes

In the second section of the survey, which was completed by responders with AD, disease severity was evaluated, and data on the impact of family and home life on the disease burden on the individual's family were collected. AD severity in the past week was evaluated by the patient-oriented eczema measure (POEM) [21], with total scores ranging from 0 (lowest severity) to 28 (highest severity). POEM scores of 0 to 7 indicate mild disease, 8 to 16 indicate moderate disease, and >16 indicate severe disease [22]. Disease severity was also assessed using the patient global assessment (PtGA) [23], with self-reported severity classified as clear/mild, moderate, or severe.

The impact of disease burden on the QoL of the parents and family of the individual with AD was assessed using the dermatitis family impact (DFI) questionnaire [24]. The DFI questionnaire assesses the impact of disease on (1) housework, (2) food preparation and feeding, (3) sleep of other family members, (4) family leisure activities, (5) time spent shopping for the family, (6) expenditure, (7) causing tiredness and exhaustion of parents/caregivers, (8) causing emotional distress of parents/caregivers, (9) relationships between the main caregiver and partner or other children, and (10) the main caregiver's life. Total scores range from 0 to 30, with higher scores indicating a greater impact on family life [24]. The impact of the disease on the time spent caring for the child and the number of workdays missed was also examined.

Additionally, the impact of a family history of allergic conditions, including AD, hay fever, or asthma, on AD severity was assessed. The impact of second-hand smoke exposure and the presence of domestic pets on the prevalence and severity of AD was also examined.

2.4. Statistical Analysis

The target population size was determined prior to data collection to ensure that the surveyed individuals were representative of the population in Japan for sex, age, geographic regions, and urban versus rural residence. A weighting adjustment was applied if this target was not met exactly, as previously described [19].

Descriptive statistics were used to present the data, with continuous data described by the mean, median, standard deviation (SD), and range. The numbers of individuals as a proportion of the sample, means, and medians were weighted, while the absolute numbers of individuals were unweighted.

3. Results

3.1. Study Population Diagnosed with AD

Of the 5702 pediatric patients in the Japanese EPI-CARE population, 1671 (29.3%) were aged 6 months to <6 years, 1989 (34.9%) were aged 6 to <12 years and 2042 (35.8%) were aged 12 to <18 years. The prevalence of diagnosed AD was 12.9% (n = 226) in children aged <6 years, 10.3% (n = 200) in children aged 6 to <12 years, and 9.1% (n = 182) among adolescents aged 12 to <18 years.

The proportion of patients with clear/mild AD, moderate AD, and severe AD based on the POEM score was 63.2%, 32.4%, and 4.4%, respectively, and based on PtGA scores, it was 72.3%, 25.5%, and 2.2%, respectively [19].

3.2. Impact on Family QoL

In all age groups, mean DFI scores increased as the severity of 'diagnosed AD' increased (Figure 1a). In patients with clear/mild AD based on POEM scores, mean (SD) DFI scores were numerically higher in children aged <6 years than in those aged 6 to <12 years or 12 to <18 years (3.98 (5.76) vs 2.47 (4.29) and 2.35 (4.21), respectively). The mean (SD) DFI scores were also numerically higher in children with moderate AD aged <6 years (7.52 (6.85)) than those aged 6 to <12 years (5.86 (6.28)) or 12 to <18 years (5.92 (6.95)). Among patients with severe AD, mean (SD) DFI scores were 9.36 (5.93) in children aged <6 years, 14.47 (8.22) in those aged 6 to <12 years, and 11.55 (9.86) in adolescents aged 12 to <18 years.

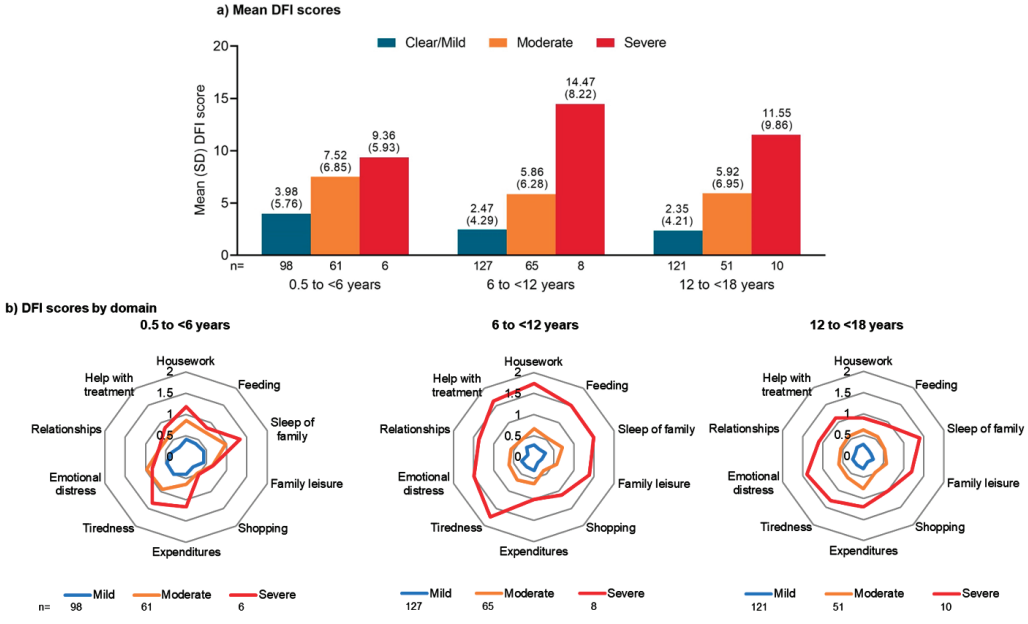

Figure 1. (**a**) Mean dermatitis family impact (DFI) scores according to POEM severity of atopic dermatitis in Japanese pediatric patients across age groups; (**b**) distribution of DFI domain scores across age groups. DFI, dermatitis family impact; POEM, patient-orientated eczema measure; SD, standard deviation.

When DFI scores were examined by individual domains in children aged <6 years, increased disease severity had the greatest impact on the 'sleep of family' and 'tiredness' domains (Figure 1b). In children aged 6 to <12 years, severe AD had a marked impact on all DFI domains, with the greatest impact being on 'tiredness.' In adolescents aged

12 to <18 years, moderate AD had the greatest impact on the 'expenditures' domain, and severe AD had the greatest impact on the 'emotional distress,' 'sleep of family,' and 'tiredness' domains.

3.3. Impact on Parents' Time

In general, the mean number of hours spent caring for a child with 'diagnosed AD' in the past week increased as AD severity increased (Figure 2a). In children aged <6 years, a mean of 4.62 h was spent by parents caring for those who had mild diseases. In all age groups, the length of time spent caring for those with moderate or severe AD was more than twice that needed for those with mild AD. In patients aged 6 to <12 years and 12 to <18 years, the time needed to care for those with severe AD was more than three times that required for those with moderate AD.

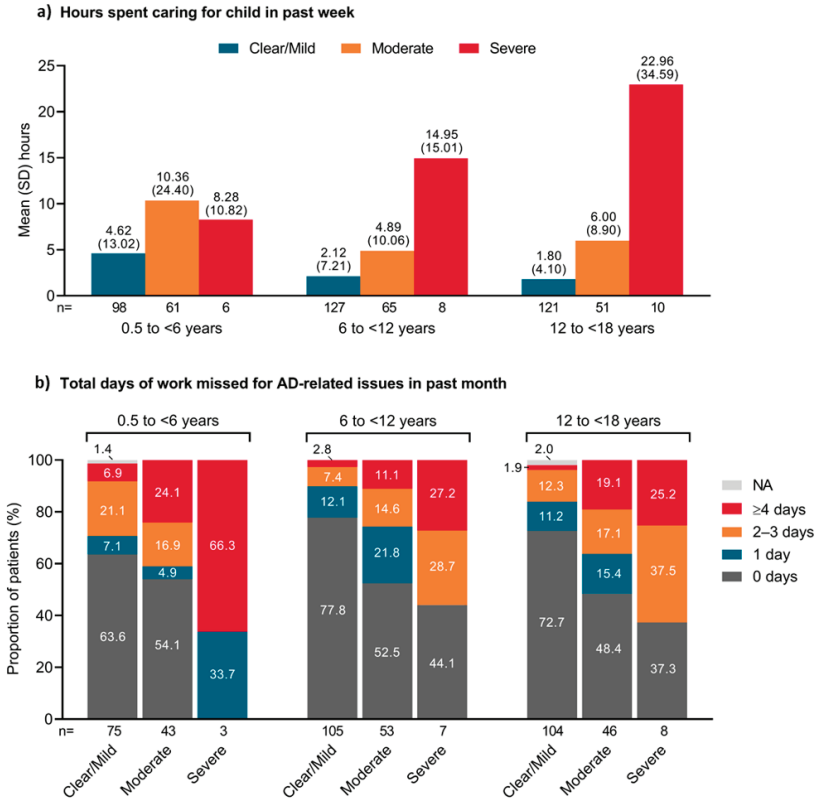

Figure 2. (**a**) Mean hours spent caring for child in the past week; (**b**) total days of work missed for atopic dermatitis-related issues in the past month for parents/caregivers of Japanese pediatric patients across age groups. AD, atopic dermatitis; NA, not applicable; SD, standard deviation.

The number of workdays missed for AD-related issues in the past month also increased as AD severity increased (Figure 2b). Among parents of children or adolescents with severe AD, at least 1 workday was missed by 100.0% of parents of children aged <6 years, 55.9% of those aged 6 to <12 years, and 62.7% of those aged 12 to <18 years.

3.4. Family History of Allergic Conditions

There was a family history of AD, hay fever, or asthma in 488/547 patients (89.1%) with 'diagnosed AD'. In children aged <6 years, 91.4% of those with clear/mild AD, 91.6%

of those with moderate disease, and 100.0% of those with severe disease had a family history of AD, hay fever, or asthma (Figure 3). Similar trends were observed in patients aged 6 to <12 years and 12 to <18 years.

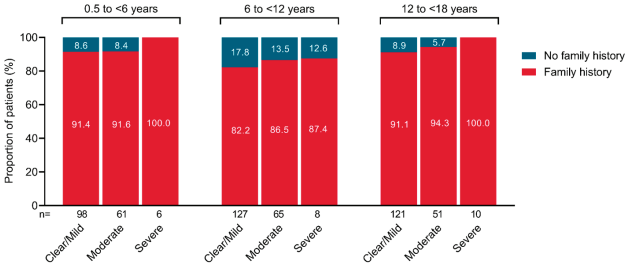

Figure 3. Proportion of patients with a family history of atopic dermatitis (AD), hay fever, or asthma according to POEM severity of AD in Japanese pediatric patients across age groups. POEM, patient-orientated eczema measure.

3.5. Current Residency

The majority of pediatric patients with AD lived in urban or suburban regions of the country across all age groups, regardless of AD severity (Supplementary Figure S1). Current residency did not appear to impact disease severity in any age group.

3.6. Second-Hand Smoke Exposure

The prevalence of 'reported AD,' 'physician-diagnosed AD,' and 'diagnosed AD' was higher in children or adolescents with a current or occasional smoker in the household than in those from a non-smoking household (Figure 4). Across age groups, the prevalence of 'reported AD' ranged from 12.9–24.2% in individuals living with non-smokers, from 25.5–31.8% in those living with a current smoker, and from 27.8–33.3% in those living with an occasional smoker. In young children aged <6 years, 83.0% of those with severe 'diagnosed AD' were living with a smoker in the household (Supplementary Figure S2).

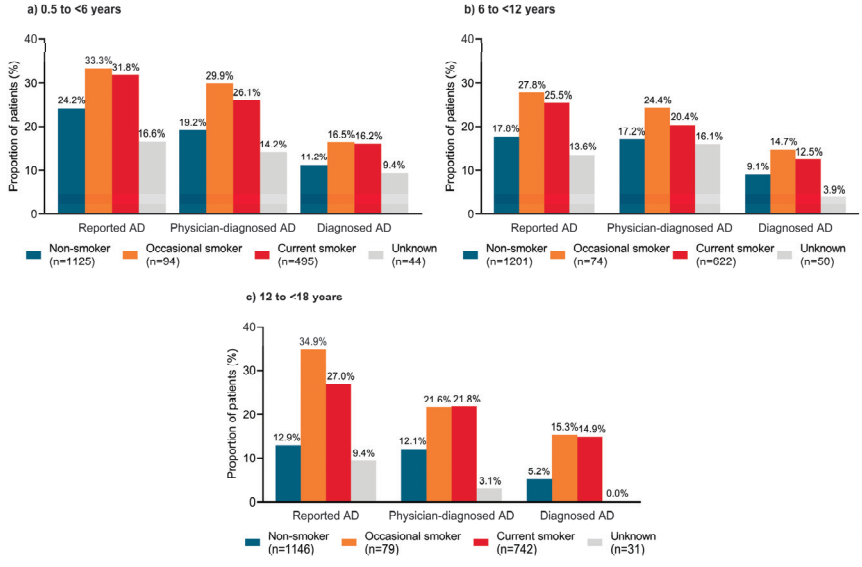

Figure 4. Prevalence of atopic dermatitis according to family smoking status in Japanese pediatric patients aged (**a**) 0.5 to <6 years; (**b**) 6 to <12 years; (**c**) 12 to <18 years. AD, atopic dermatitis.

3.7. Household Pets

The prevalence of 'reported AD,' 'physician-diagnosed AD,' and 'diagnosed AD' was higher among children or adolescents who lived with a household pet than in those without a household pet (Figure 5). Among those living with a household pet, 26.2–35.8% had 'reported AD,' 21.3–28.7% had 'physician-diagnosed AD,' and 13.8–18.5% had 'diagnosed AD'. In contrast, in children living without a household pet, 13–23.1% had 'reported AD,' 12.3–18.8% had 'physician-diagnosed AD,' and 5.9–10.7% had 'diagnosed AD'. However, the number of household pets did not appear to impact disease severity in any of the age groups (Supplementary Figure S3).

3.8. Parent Education and Employment Status

In general, a high proportion of the parents of affected children/adolescents had college, university, or graduate school education across all age groups, regardless of AD severity (Supplementary Figure S4). In addition, the majority of parents were employed (Supplementary Figure S5). Education and employment status did not appear to impact disease severity in any age group.

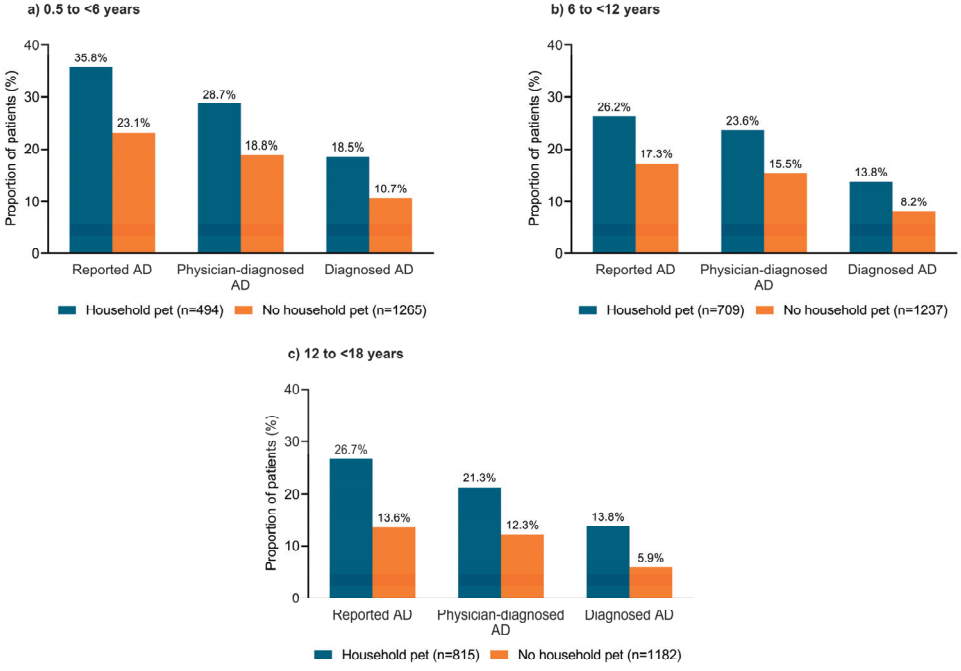

Figure 5. Prevalence of atopic dermatitis according to the presence of a household pet in Japanese pediatric patients aged (**a**) 0.5 to <6 years; (**b**) 6 to <12 years; (**c**) 12 to <18 years. AD, atopic dermatitis.

4. Discussion

In this Japanese pediatric population of the EPI-CARE study, 10.7% of patients had been diagnosed with AD within the last 12 months [19]. According to the POEM tool, the majority of those with diagnosed AD had a clear/mild disease (63.2%), while 32.4% and 4.4% had moderate or severe AD, respectively [19].

In the current analysis, the DFI questionnaire showed that family QoL decreased as the severity of their child's AD increased. The mean DFI scores ranged from 2.35–3.98 across age groups in patients with clear/mild AD, from 5.86–7.52 in those with moderate AD, and from 9.36–14.47 in those with severe AD. In patients with clear/mild, or moderate AD, mean DFI scores were numerically higher for families with children aged <6 years than for

families with older children. When the individual DFI domains were evaluated, disease severity appeared to have the greatest impact on the 'sleep of family' and 'tiredness' among families with children aged <6 years or 6 to <12 years.

These results are consistent with those of an international web survey [10] and the Avon Longitudinal Study of Parents and Children (ALSPAC) study in the UK [25]. In the international web survey (n = 235), parental sleep disturbance and fatigue both increased as the child's AD severity increased [10]. Similarly, the ALSPAC study, which included 11,649 mother–child pairs, showed that mothers of children with mild, moderate, or severe AD had difficulty falling asleep, subjectively insufficient sleep, and daytime exhaustion during the first 11 years of their child's life [25]. In Japanese families, young children and their parents often sleep in the same room, so parents will often experience sleep disturbances if their child has sleeping difficulties due to AD symptoms.

The current analysis showed a marked decrease in family QoL among children aged 6 to <12 years with severe AD, most likely because parents are solely responsible for managing skin care in this age group. Our results also showed that the burden on parents and families is greater in children aged <6 years than in those aged 6 to <12 years or 12 to <18 years for mild or moderate AD. This may be because younger children with AD are less able to cope with their symptoms and require more care from their parents/caregivers than older children or adolescents.

The time spent by parents caring for their children with AD in the past week increased in proportion to disease severity in children aged ≥6 years, being more than threefold higher for parents with children/adolescents with severe AD than for those with moderate AD. In children aged <6 years, parents spent an average of at least 4.6 h per week caring for their children, even those with mild symptoms. The increase in the time required to care for children aged ≥6 years was associated with an increase in the number of workdays missed by parents for AD-related issues in the past month, particularly when their child had severe AD. The increase in work absences may be caused by the child being absent from school. These findings are in line with those of a cross-sectional United States study that included 3132 children with AD and found a two-fold higher likelihood of chronic school absenteeism (i.e., ≥15 days missed per year) among children with severe AD versus mild-to-moderate AD. The same study found a significantly higher number of workdays missed among both fathers ($p = 0.03$) and mothers ($p < 0.0001$) of children with AD [26].

Among patients with 'diagnosed AD,' most (89.1%) had a family history of AD, hay fever, or asthma. The proportion of patients with a family history of allergic conditions was >80% across all age groups and all disease severity classifications, suggesting that a family history of allergies plays a role in the development of AD at any age, regardless of disease severity. This is consistent with the concept of atopic diathesis, whereby atopic diseases such as AD, asthma, and allergic rhinitis are genetically linked [4]. Data from the ALSPAC study indicated that a parental history of AD was a strong predictor of AD in their offspring, although a history of asthma or hay fever alone was only associated with childhood AD if both parents had these conditions [16]. In the Japan Environment and Children's Study, the lifetime prevalence of parental AD was 15.7% among mothers and 11.2% among fathers [27].

A correlation between second-hand smoke exposure, AD development, and AD severity has been observed in several studies, including a meta-analysis [17], Japanese studies [28–30], and studies from other countries [31–33]. In a Japanese survey of 4466 adolescents aged 13–14 years, household smoking was an important modifiable risk factor for AD disease severity [29]. In a cross-sectional Japanese study of 1177 parent–infant pairs, fetal smoke exposure after 28 weeks gestation was associated with significantly higher adjusted odds of AD syndrome in infants aged >6 months compared with unexposed infants (adjusted odds ratio (aOR), 5.21; 95% confidence interval (CI), 1.08–25.15; $p = 0.020$) [28]. Similarly, a Japanese prospective pre-birth cohort study of 1354 mother–child pairs showed higher adjusted odds of physician-diagnosed AD in children with prenatal smoking exposure compared with no prenatal smoking exposure (aOR, 7.11; 95% CI, 1.43–27.8). However,

there was no association between perinatal smoking exposure and AD defined according to ISAAC criteria [30]. In a meta-analysis of 86 studies from 39 countries, second-hand smoke exposure was associated with increased odds of AD in children aged <18 years (OR, 1.18; 95% CI, 1.01–1.38), but the odds of AD in childhood were not significantly increased with maternal smoking exposure during pregnancy [17]. The findings of the current analysis were generally in line with previous studies, with an increased prevalence of 'reported AD,' 'physician-diagnosed AD,' and 'diagnosed AD' among children or adolescents living with a current or occasional smoker compared with those living in a non-smoking household. However, statistical significance testing and logistic regression analyses were not conducted. In contrast, the Osaka Maternal and Child Health Study in 865 Japanese parent–child pairs showed that maternal smoking was not related to an increased risk of suspected AD [34].

Randomized studies on the association between household pets and pediatric AD are difficult to conduct, and data from observational studies are conflicting. A previous study of 3864 school children has shown a statistical association between having a pet rabbit and severe AD (adjusted prevalence ratio (aPR), 1.94; 95% CI, 1.02–3.71) or study-defined current AD (defined as physician-diagnosed AD and/or ever having a recurrent itchy rash for ≥6 months and having a current itchy flexural rash; aPR, 1.45; 95% CI, 1.07–1.20), and between having a pet cat and physician-diagnosed AD (aPR, 1.25; 95% CI, 1.03–1.50). In contrast, having a pet dog was not statistically associated with AD symptoms [18]. Alternatively, the Osaka Maternal and Child Health study found that exposure to indoor domestic pets (i.e., dogs, cats, birds, or hamsters) during pregnancy was not statistically associated with AD in 865 infants (aOR, 1.15; 95% CI, 0.55–2.25) [34]. Another Japanese study, which surveyed 35,242 schoolchildren aged 6 years, reported that cat ownership was associated with a significantly lower prevalence of AD (aOR, 0.79; 95% CI, 0.67–0.93) [35]. In the current analysis, AD prevalence appeared to be higher among children or adolescents who lived with a household pet than in those without pets, although there was no association between household pets and disease severity. The Japanese guidelines for AD recommend avoiding pets in households with individuals who test positive for specific immunoglobulin E antibodies in animals (e.g., dogs, cats, other mammals, birds, and hamsters) [36].

The apparent increase in AD prevalence with second-hand smoke exposure and household pets in the current study is consistent with the concept of the exposome, whereby environmental exposure to various factors leads to disruption of the epithelial skin and mucosal barriers and subsequent development of atopic diseases such as AD, allergic rhinitis, food allergies, chronic rhinosinusitis, and asthma [5–9]. In this context, aryl hydrocarbon receptor (AhR) signaling pathways are thought to be involved in regulating skin homeostasis in response to environmental exposure, which may have therapeutic implications for the pharmacologic management of AD [37,38].

The limitations of this study include its retrospective design, the small number of individuals with severe AD and the lack of long-term follow-up, sampling and nonresponse bias, potential recall bias due to the survey being completed by the parents of children aged <12 years or by adolescents aged 12 to <18 years, and the lack of statistical significance or regression analyses. In addition, the data were collected from Japanese patients only, which may limit the generalizability of these results to other ethnicities. However, the strengths of this study include the collection of country-specific representative data and the use of ISAAC criteria to identify individuals with reported or diagnosed AD, which allows for a consistent method of evaluating AD prevalence.

5. Conclusions

In conclusion, this population-based study of Japanese children and adolescents showed that AD had a negative impact on family QoL, especially the sleep of family members, tiredness, and the time spent caring for the child, which increased with increasing disease severity. A family history of allergic conditions was present in >80% of children or

adolescents with AD, and the prevalence of AD in the past 12 months appeared to be higher among children exposed to second-hand smoke and in those living with a household pet.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/jcm12082988/s1, Figure S1: Current residency of families of Japanese pediatric patients with atopic dermatitis by disease severity and age group; Figure S2: Proportion of households with or without smokers by the severity of atopic dermatitis in Japanese pediatric patients across age groups; Figure S3: Number of household pets by the severity of atopic dermatitis in Japanese pediatric patients across age groups; Figure S4: Parents' highest education level by the severity of atopic dermatitis in Japanese pediatric patients across age groups; Figure S5: Parents' employment status by the severity of atopic dermatitis in Japanese pediatric patients across age groups.

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Institutional Review Board Statement: The EPI-CARE study was conducted in accordance with the European Union General Data Protection Regulation, the European Society for Opinion and Marketing Research, the Insights Association, the European Pharmaceutical Marketing Research Association, the British Healthcare Business Intelligence Association, the US Health Insurance Portability, and Accountability Act, and all international and local data protection legislation. However, no personal data were collected from the survey and no medical information was abstracted from the patient's medical records, the submission and approval of the study by central and/or local ethical committees in each country were not needed. Therefore, there is no ethical code for this study.

Informed Consent Statement: Written informed consent was obtained from all participants and their parents prior to study entry. No personally identifiable information or medical data were collected from patients.

Data Availability Statement: The datasets generated/analyzed during the current study are available from the corresponding author upon reasonable request.

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References

1. Irvine, A.D.; Mina-Osorio, P. Disease trajectories in childhood atopic dermatitis: An update and practitioner's guide. *Br. J. Dermatol.* **2019**, *181*, 895–906. [CrossRef]
2. Lyons, J.J.; Milner, J.D.; Stone, K.D. Atopic dermatitis in children: Clinical features, pathophysiology, and treatment. *Immunol. Allergy Clin. N. Am.* **2015**, *35*, 161–183. [CrossRef] [PubMed]
3. Napolitano, M.; Fabbrocini, G.; Martora, F.; Genco, L.; Noto, M.; Patruno, C. Children atopic dermatitis: Diagnosis, mimics, overlaps, and therapeutic implication. *Dermatol. Ther.* **2022**, *35*, e15901. [CrossRef]
4. Novak, N.; Bieber, T. Allergic and nonallergic forms of atopic diseases. *J. Allergy Clin. Immunol.* **2003**, *112*, 252–262. [CrossRef] [PubMed]
5. Celebi Sozener, Z.; Özbey Yücel, Ü.; Altiner, S.; Ozdel Oztürk, B.; Cerci, P.; Türk, M.; Gorgülü Akin, B.; Akdis, M.; Yilmaz, I.; Ozdemir, C.; et al. The external exposome and allergies: From the perspective of the epithelial barrier hypothesis. *Front. Allergy* **2022**, *3*, 887672. [CrossRef]
6. Molina-García, M.; Granger, C.; Trullàs, C.; Puig, S. Exposome and skin: Part 1. Bibliometric analysis and review of the impact of exposome approaches on dermatology. *Dermatol. Ther.* **2022**, *12*, 345–359. [CrossRef]
7. Stefanovic, N.; Flohr, C.; Irvine, A.D. The exposome in atopic dermatitis. *Allergy* **2020**, *75*, 63–74. [CrossRef]
8. Stefanovic, N.; Irvine, A.D.; Flohr, C. The role of the environment and exposome in atopic dermatitis. *Curr. Treat. Options Allergy* **2021**, *8*, 222–241. [CrossRef] [PubMed]
9. Celebi Sozener, Z.; Ozdel Ozturk, B.; Cerci, P.; Turk, M.; Gorgulu Akin, B.; Akdis, M.; Altiner, S.; Ozbey, U.; Ogulur, I.; Mitamura, Y.; et al. Epithelial barrier hypothesis: Effect of the external exposome on the microbiome and epithelial barriers in allergic disease. *Allergy* **2022**, *77*, 1418–1449. [CrossRef]
10. Capozza, K.; Gadd, H.; Kelley, K.; Russell, S.; Shi, V.; Schwartz, A. Insights from caregivers on the impact of pediatric atopic dermatitis on families: "I'm tired, overwhelmed, and feel like I'm failing as a mother". *Dermatitis* **2020**, *31*, 223–227. [CrossRef]
11. Meltzer, L.J.; Flewelling, K.D.; Jump, S.; Gyorkos, E.; White, M.; Hauk, P.J. Impact of atopic dermatitis treatment on child and parent sleep, daytime functioning, and quality of life. *Ann. Allergy Asthma Immunol.* **2020**, *124*, 385–392. [CrossRef] [PubMed]
12. Yang, E.J.; Beck, K.M.; Sekhon, S.; Bhutani, T.; Koo, J. The impact of pediatric atopic dermatitis on families: A review. *Pediatr. Dermatol.* **2019**, *36*, 66–71. [CrossRef]
13. Barbarot, S.; Silverberg, J.I.; Gadkari, A.; Simpson, E.L.; Weidinger, S.; Mina-Osorio, P.; Rossi, A.B.; Brignoli, L.; Mnif, T.; Guillemin, I.; et al. The family impact of atopic dermatitis in the pediatric population: Results from an international cross-sectional study. *J. Pediatr.* **2022**, *246*, 220–226.e5. [CrossRef] [PubMed]
14. Huang, E.; Ong, P.Y. Severe atopic dermatitis in children. *Curr. Allergy Asthma Rep.* **2018**, *18*, 35. [CrossRef]
15. Capozza, K.; Schwartz, A.; Lang, J.E.; Chalmers, J.; Camilo, J.; Abuabara, K.; Kelley, K.; Harrison, J.; Vastrup, A.; Stancavich, L.; et al. Impact of childhood atopic dermatitis on life decisions for caregivers and families. *J. Eur. Acad. Dermatol. Venereol.* **2022**, *36*, e451–e454. [CrossRef]
16. Wadonda-Kabondo, N.; Sterne, J.A.; Golding, J.; Kennedy, C.T.; Archer, C.B.; Dunnill, M.G.; Team, A.S. Association of parental eczema, hayfever, and asthma with atopic dermatitis in infancy: Birth cohort study. *Arch. Dis. Child.* **2004**, *89*, 917–921. [CrossRef] [PubMed]
17. Kantor, R.; Kim, A.; Thyssen, J.P.; Silverberg, J.I. Association of atopic dermatitis with smoking: A systematic review and meta-analysis. *J. Am. Acad. Dermatol.* **2016**, *75*, 1119–1125.e1. [CrossRef]
18. AlShatti, K.A.; Ziyab, A.H. Pet-keeping in relation to asthma, rhinitis, and eczema symptoms among adolescents in Kuwait: A cross-sectional study. *Front. Pediatr.* **2020**, *8*, 331. [CrossRef]
19. Silverberg, J.I.; Barbarot, S.; Gadkari, A.; Simpson, E.L.; Weidinger, S.; Mina-Osorio, P.; Rossi, A.B.; Brignoli, L.; Saba, G.; Guillemin, I.; et al. Atopic dermatitis in the pediatric population: A cross-sectional, international epidemiologic study. *Ann. Allergy Asthma Immunol.* **2021**, *126*, 417–428.e2. [CrossRef]
20. Asher, M.I.; Keil, U.; Anderson, H.R.; Beasley, R.; Crane, J.; Martinez, F.; Mitchell, E.A.; Pearce, N.; Sibbald, B.; Stewart, A.W.; et al. International Study of Asthma and Allergies in Childhood (ISAAC): Rationale and methods. *Eur. Respir. J.* **1995**, *8*, 483–491. [CrossRef]
21. Charman, C.R.; Venn, A.J.; Williams, H.C. The patient-oriented eczema measure: Development and initial validation of a new tool for measuring atopic eczema severity from the patients' perspective. *Arch. Dermatol.* **2004**, *140*, 1513–1519. [CrossRef]
22. Charman, C.R.; Venn, A.J.; Ravenscroft, J.C.; Williams, H.C. Translating Patient-Oriented Eczema Measure (POEM) scores into clinical practice by suggesting severity strata derived using anchor-based methods. *Br. J. Dermatol.* **2013**, *169*, 1326–1332. [CrossRef] [PubMed]
23. Silverberg, J.I.; Chiesa Fuxench, Z.C.; Gelfand, J.M.; Margolis, D.J.; Boguniewicz, M.; Fonacier, L.; Grayson, M.H.; Simpson, E.L.; Ong, P.Y. Content and construct validity, predictors, and distribution of self-reported atopic dermatitis severity in US adults. *Ann. Allergy Asthma Immunol.* **2018**, *121*, 729–734.e4. [CrossRef]
24. Lawson, V.; Lewis-Jones, M.S.; Finlay, A.Y.; Reid, P.; Owens, R.G. The family impact of childhood atopic dermatitis: The dermatitis family impact questionnaire. *Br. J. Dermatol.* **1998**, *138*, 107–113. [CrossRef] [PubMed]
25. Ramirez, F.D.; Chen, S.; Langan, S.M.; Prather, A.A.; McCulloch, C.E.; Kidd, S.A.; Cabana, M.D.; Chren, M.M.; Abuabara, K. Assessment of sleep disturbances and exhaustion in mothers of children with atopic dermatitis. *JAMA Dermatol.* **2019**, *155*, 556–563. [CrossRef]

26. Cheng, B.T.; Silverberg, J.I. Association of pediatric atopic dermatitis and psoriasis with school absenteeism and parental work absenteeism: A cross-sectional United States population-based study. *J. Am. Acad. Dermatol.* **2021**, *85*, 885–892. [CrossRef] [PubMed]
27. Yamamoto-Hanada, K.; Yang, L.; Ishitsuka, K.; Ayabe, T.; Mezawa, H.; Konishi, M.; Shoda, T.; Matsumoto, K.; Saito, H.; Ohya, Y.; et al. Allergic profiles of mothers and fathers in the Japan Environment and Children's Study (JECS): A nationwide birth cohort study. *World Allergy Organ. J.* **2017**, *10*, 24. [CrossRef]
28. Shinohara, M.; Matsumoto, K. Fetal tobacco smoke exposure in the third trimester of pregnancy is associated with atopic eczema/dermatitis syndrome in infancy. *Pediatr. Allergy Immunol. Pulmonol.* **2017**, *30*, 155–162. [CrossRef]
29. Sugiyama, T.; Sugiyama, K.; Toda, M.; Yukawa, T.; Makino, S.; Fukuda, T. Risk factors for asthma allergic diseases among 13–14-year-old school children in Japan. *Allergol. Int.* **2002**, *51*, 139–150. [CrossRef]
30. Tanaka, K.; Miyake, Y.; Furukawa, S.; Arakawa, M. Pre- and postnatal smoking exposure and risk of atopic eczema in young Japanese children: A prospective prebirth cohort study. *Nicotine Tob. Res.* **2017**, *19*, 804–809. [CrossRef]
31. Jing, D.; Li, J.; Tao, J.; Wang, X.; Shan, S.; Kang, X.; Wu, B.; Zhang, Y.; Xiao, Y.; Chen, X.; et al. Associations of second-hand smoke exposure with hand eczema and atopic dermatitis among college students in China. *Sci. Rep.* **2020**, *10*, 17400. [CrossRef] [PubMed]
32. Kim, S.Y.; Sim, S.; Choi, H.G. Atopic dermatitis is associated with active and passive cigarette smoking in adolescents. *PLoS ONE* **2017**, *12*, e0187453. [CrossRef]
33. Lee, A.; Lee, S.Y.; Lee, K.S. Association of secondhand smoke exposure with allergic multimorbidity in Korean adolescents. *Sci. Rep.* **2020**, *10*, 16409. [CrossRef] [PubMed]
34. Miyake, Y.; Ohya, Y.; Tanaka, K.; Yokoyama, T.; Sasaki, S.; Fukushima, W.; Ohfuji, S.; Saito, K.; Kiyohara, C.; Hirota, Y.; et al. Home environment and suspected atopic eczema in Japanese infants: The Osaka maternal and child health study. *Pediatr. Allergy Immunol.* **2007**, *18*, 425–432. [CrossRef] [PubMed]
35. Kurosaka, F.; Nakatani, Y.; Terada, T.; Tanaka, A.; Ikeuchi, H.; Hayakawa, A.; Konohana, A.; Oota, K.; Nishio, H. Current cat ownership may be associated with the lower prevalence of atopic dermatitis, allergic rhinitis, and Japanese cedar pollinosis in schoolchildren in Himeji, Japan. *Pediatr. Allergy Immunol.* **2006**, *17*, 22–28. [CrossRef] [PubMed]
36. Katoh, N.; Ohya, Y.; Ikeda, M.; Ebihara, T.; Katayama, I.; Saeki, H.; Shimojo, N.; Tanaka, A.; Nakahara, T.; Nagao, M.; et al. Japanese guidelines for atopic dermatitis 2020. *Allergol. Int.* **2020**, *69*, 356–369. [CrossRef]
37. Furue, M.; Hashimoto-Hachiya, A.; Tsuji, G. Aryl hydrocarbon receptor in atopic dermatitis and psoriasis. *Int. J. Mol. Sci.* **2019**, *20*, 5424. [CrossRef]
38. Napolitano, M.; Fabbrocini, G.; Martora, F.; Picone, V.; Morelli, P.; Patruno, C. Role of aryl hydrocarbon receptor activation in inflammatory chronic skin diseases. *Cells* **2021**, *10*, 3559. [CrossRef]

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Definition of the Clinical Characteristics of Patients with Moderate and Severe Atopic Dermatitis for Whom Narrow-Band UVB (NB-UVB) and Medium-Dose UVA1 Phototherapies Are Still Valuable Treatment Options at the Age of Biologics

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Abstract: Narrow-band (NB) UVB and UVA1 have been successfully used for the treatment of atopic dermatitis (AD) since the 1980s, but the clinical indications for their use "at the age of biologics" remain to be assessed. From 2013 to 2017, 145 patients underwent a first treatment cycle with phototherapy. They achieved a median final EASI score of 9.90 with UVA1 and 13.70 with NB-UVB. The rates of patients achieving an IGA score of 0/1 persistent for at least 6 months were 33% with UVA1 and 28% with NB-UVB, and the rates with an EASI90 improvement were 10.9% with UVA1 and 11.0% with NB-UVB. The cut-off baseline EASI values for a good probability to achieve a 0/1 IGA were 24.4 with UVA1 and 24.7 with NB-UVB. A 0/1 IGA persistent for at least 6 months was more likely to be achieved by patients with a history of flares interspersed with periods of mild or no disease. From 2018, we only enrolled patients with the above-mentioned characteristics. The number of treated patients was lower, but the final EASI score, the rate of patients achieving IGA 0/1 persistent for at least 6 months, and EASI90 were significantly higher. Medium-dose UVA1 and NB-UVB phototherapies remain useful for the treatment of AD patients with a baseline EASI score lower than 24.4 and 24.7, respectively, and a medical history of flares followed by prolonged periods of complete or near-complete remission.

Keywords: narrow-band UVB phototherapy; ultraviolet A1 phototherapy; dupilumab; atopic dermatitis

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1. Introduction

Atopic dermatitis (AD) is a chronic inflammatory skin disease characterized by an impaired skin barrier, the upregulation of type 2 immune responses and increased Staphylococcus aureus colonization. Patients have dry skin, eczematous lesions and intense itching leading to excoriation and lichenification [1].

Emollients and topical drugs, i.e., corticosteroids and calcineurin inhibitors, are enough to manage patients with mild skin involvement and, until a few years ago, corticosteroids, cyclosporine, methotrexate, azathioprine and mycophenolate were the only systemic drug treatments for moderate and severe AD [1]. However, their efficacy is often poor and there are several contraindications and safety concerns, particularly if they are administered for a long time [2].

Phototherapies with exposures to narrow-band (312 ± 2 nm) UVB (NB-UVB) and medium-dose (30–60 J/cm^2) UVA1 (340–400 nm) radiation have represented therapeutic alternatives to drug treatments since the 1980s. Compared to placebo or no treatment, they

have been found to improve physician-rated signs and patient-reported symptoms without a difference in withdrawal due to adverse events [3]. Other main advantages include the low number of contraindications and the lack of toxicity on internal organs. However, randomized comparative controlled clinical trials (RCTs) with drug treatments have never been conducted [3], and they are not suitable for long-term and maintenance treatments because of their carcinogenic potential [4–6].

Recently, dupilumab, a fully human monoclonal antibody that binds specifically to the shared α-chain receptor subunit for interleukins (IL)-4 and IL-13, was found to be highly effective and with a good safety profile in two large RCTs [7,8]. Afterwards, other monoclonal antibodies, i.e., tralokinumab, and JAK inhibitors, i.e., baricitinib, upatacitinib and abrocitinib, were approved by the European Medicines Agency (EMA) for the treatment of AD [9]. These drugs represented a major breakthrough in the treatment paradigm for AD, which is now no longer the short-term improvement but the long-term complete or near-complete control of skin manifestations and the prevention of flares.

In this rapidly changing therapeutic landscape, the most recent guidelines and consensus papers [9–11] agree that phototherapies are still valuable treatment options for moderate and severe AD, but criteria for the assessment of the subgroup of AD patients who can preferentially benefit from phototherapies after the availability of new immunological treatments are not clearly established.

In 2018, which is when dupilumab became available for clinical use in Italy [8], we retrospectively reviewed the medical files of patients who underwent a first phototherapy cycle over the previous 5-year period from 2013 to 2017. We analyzed the outcome of the treatment in relation to two clinical criteria: the severity of the clinical manifestations at baseline, as measured with the EASI score; and the longitudinal medical history of the disease, by distinguishing patients with a continuous or almost-continuous course from those who present prolonged periods of absent or mild disease spontaneously or after treatments [12]. Based on the results of this first analysis (see later in Section 3), in the following 5 years (2018–2022) we only enrolled patients who had a baseline EASI score and a medical history that were associated with a good probability of remission and long-term control of the skin disease [13]. Finally, we verified the effect of these patient selection criteria by comparing the therapeutic results observed in patients treated in the period 2013–2017 with those of patients treated in the period 2018–2022.

2. Materials and Methods

In 2018, we reviewed the medical files of 187 patients who underwent a first treatment cycle with NB-UVB phototherapy and medium-dose UVA1 phototherapy from 2013 to 2017 at the Photodermatology Unit of the ASST Spedali Civili University of Brescia, a tertiary referral center for AD treatment in Northern Italy. All patients had chronic atopic dermatitis for at least 3 years before screening, and topical treatment provided inadequate control or was medically inadvisable. The patients were at least 12 years of age and suffering from AD, with a baseline Eczema Area and Severity Index (EASI) of $\geq$7 [14,15], without a maximum value of EASI score being established. Exclusion criteria were: other inflammatory skin diseases, absolute and relative contraindications to phototherapy [16], congenital or acquired immunodeficiency syndrome and an inability to understand and sign their informed consent. Before starting phototherapy, the patients discontinued systemic drug treatments for at least 1 month and topical corticosteroids and calcineurin inhibitors for at least 2 weeks.

At baseline, we also registered the individual longitudinal course of the disease in the previous years and dichotomized patients who reported a continuous course, i.e., skin involvement with small spontaneous variations over time and short periods of partial/good remissions after treatments, and patients who reported an intermittent course, i.e., flares interspersed with prolonged periods of absent or mild disease spontaneously or after treatments [12]. The distinction between continuous and intermittent forms is arbitrary and subject to many biases, but it was easily assessed in most cases. During the treatment

cycle, oral antihistamine drugs were allowed at night if pruritus and sleep disturbance were not tolerated. The application of emollient creams was allowed as needed.

After the end of the phototherapy cycle, we also recorded the number of treatment sessions and cumulative UV dose. The patients were followed up with visits every 3 months or sooner if there was a recurrence not controlled by local therapies alone. Patients achieving an IGA of 0/1 were only allowed to use emollient creams and a limited use of topical corticosteroids, if needed. If patients had an improvement lower than EASI75 and/or an IGA score of >1, and the disease was not controlled with only topical treatments, other systemic therapies were prescribed, including new monoclonal antibodies and anti-JAK small molecules if the EASI score was $\geq$24 [17].

The primary outcome measures were:

- The percentage of participants achieving an Investigator's Global Assessment (IGA) score of 0 (clear) or 1 (almost clear) [18];
- The percentage of patients achieving an IGA score of 0/1 without a relapse within 6 months from the end of treatment (EOT).

The secondary outcome measures were:

- The median final EASI score;
- The percentage of participants achieving EASI 75 ($\geq$75% improvement from baseline EASI), EASI 50 (>50% to <75% improvement) and EASI 90 (>90% improvement);
- The percentage of patients with an EASI improvement of <50%;
- The percentage of patients with phototherapy-related adverse events;
- The percentage of patients with phototherapy-related adverse events leading to treatment discontinuation.

Afterwards, we calculated the cut-off value of the baseline EASI score that was predictive of a high probability in order to achieve a final 0/1 IGA result and the correlation of an intermittent or continuous course with a final 0/1 IGA result. Finally, we looked at the probability of obtaining a final 0/1 IGA result persistent for at least 6 months on the bases of the baseline cut-off EASI score and the history of AD course.

After 2018, we only treated patients if they had a baseline EASI score lower than the cut-off value, as calculated above (see in Section 3), and they reported an intermittent skin involvement. We recorded the personal details, individual AD characteristics, the number of treatment sessions and cumulative UV dose of these patients, and the same primary and secondary outcome measures as described above for patients treated from 2013 to 2017.

For UVA1 exposures, we used a MediSun Xenia (Schulze & Bohm Gmbh, Bruhl, Germany) irradiation unit with UV emission strictly confined in the range from 340 to 400 nm. The radiation source of NB-UVB was a Waldmann 7001 cabinet (Waldmann Lichttechnik, Villingen-Schwenningen, Germany) equipped with 40 Philips TL-01/100W lamps (Philips, Eindhoven, Netherlands), with a peak in emission at 312 $\pm$ 2 nm. Irradiance was measured with portable broadband UV meters (Waldmann) after calibration with a Macam SR 9910 spectroradiometer (Macam Photometrics Ltd., Livingston, UK).

All patients treated with medium-dose UVA1 received a first dose of 30 J/cm^2 and, if well tolerated, fixed daily exposures of 50 J/cm^2 were delivered twice a week. The initial NB-UVB dose ranged between 0.1 and 0.4 J/cm^2, according to skin phototype. The patients were treated twice-weekly on non-consecutive days, and NB-UVB doses were adjusted at each session according to the erythema response. In short, 10%, 5% or 0% increments were delivered depending on the erythema response: none, a barely perceptible or a well-defined erythema, respectively, after 48 h. With both phototherapies, treatments were continued until complete clearing was obtained, or until partial or no improvement was seen without further amelioration despite 6 additional treatments [13,16].

The database was formatted through the Microsoft Excel TM vers. 365 software and later imported from the IBM-SPSS® software ver. 28.0.1 (IBM SPSS Inc., Chicago, IL, USA). The use of the Stata TM software ver. 17.0 (Stata Corporation, College Station, TX, USA) and the EpiInfo Statcalc TM software ver. 7.0 was also considered for comparisons or

implementations of test output. The normality of the distributions was assessed using the Kolmogorov–Smirnov test. Categorical variables were presented as frequencies or percentages and compared with the use of the Chi-Square test and the Fisher's exact test, as appropriate; associations of the crosstabs were verified using standardized adjusted residuals. Continuous variables were presented as means ± SD (in the case of a normal distribution), or medians and min/max (in the case of a skewed distribution), and compared with the use of a Student's *t*-test, ANOVA, or the Mann–Whitney and Kruskal–Wallis test; correlations among variables were identified by the Pearson's or Spearman's rank correlation test. A two-sided α level of 0.05 was used for all tests.

The authors had full access to and take full responsibility for the integrity of the data.

3. Results

The age, gender, Boston skin type, baseline EASI score and medical history (continuous/intermittent) of the AD lesions of 187 patients treated with UVA1 and NB-UVB, between 2013 and 2017, are reported in Table 1.

Table 1. Baseline main personal and clinical features and treatment results of patients who received a first treatment cycle with ultraviolet A1 (UVA1) phototherapy or narrow band-ultraviolet B (NB-UVB) phototherapy in the 2013–2017 and 2018–2022 time periods. AD = atopic dermatitis, NS = non significant.

	Patients Treated with Medium-Dose UVA1			**Patients Treated with NB-UVB**			**Comparisons of Patients' Groups Treated in 2013–2017 or 2018–2022 with the 2 Phototherapies**	
Column	**(a)**	**(b)**	**(a) vs. (b)**	**(c)**	**(d)**	**(c) vs. (d)**	**(a) vs. (c)**	**(b) vs. (d)**
	2013–2017	2018–2022	*p*	2013–2017	2018–2022	*p*	*p*	*p*
Number	46	16		99	26			
Age (years), median (range)	26.0 (7–67)	24.5 (10–52)	NS	31 (13–73)	20.5 (14–66)	NS	NS	NS
Gender			NS			NS	NS	NS
Male (%)	12 (41.3%)	5 (31.2%)		64 (64.6%)	11 (42.3%)			
Female (%)	27 (58.7%)	11 (68.8%)		35 (35.4%)	15 (57.7%)			
Boston skin phototype, n (%)			NS			NS	NS	NS
2 (%)	12 (26.1%)	5 (31.2%)		22 (22.2%)	7 (26.9%)			
3 (%)	21 (67.4%)	10 (62.5%)		67 (67.7%)	15 (57.7%)			
4 (%)	3 (6.5%)	1 (6.3%)		10 (10.1%)	4 (15.4%)			
Medical history of AD			<0.001			<0.001	NS	NS
continuous AD, n (%)	27 (58.7%)	0 (0%)		34 (34.3%)	0 (0%)			
intermittent AD *, n (%)	19 (41.3%)	16 (100%)		65 (65.7%)	26 (100%)			
Number of treatment sessions [median (range)]	30 (12–66)	18.5 (12–26)	<0.01	33 (16–51)	24 (12–30)	<0.001	NS	NS
Cumulative UV dose (J/cm^2) [median (range)]	1155.0 (360–3080)	770 (230–1750)	<0.01	16.3 (0.6–30.8)	11.3 (3.6–28.5)	0.02		
Patients with an IGA 0/1 result	21 (46%)	12 (75%)	0.014	42 (42%)	19 (73.1%)	0.037	NS	NS

Table 1. *Cont.*

	Patients Treated with Medium-Dose UVA1			Patients Treated with NB-UVB			Comparisons of Patients' Groups Treated in 2013–2017 or 2018–2022 with the 2 Phototherapies	
Column	**(a)**	**(b)**	**(a) vs. (b)**	**(c)**	**(d)**	**(c) vs. (d)**	**(a) vs. (c)**	**(b) vs. (d)**
Patients with an IGA 0/1 persistent for at least 6 months	15 (33%)	11 (69%)	0.012	28 (28%)	18 (69.2%)	0.016	NS	NS
Baseline median EASI (range)	28.7 (8.7–52.2)	18.7 (8.6–27.4)	<0.001	30.5 (7.9–52.2)	18.4 (9.1–26.3)	<0.001	NS	NS
Final median EASI (range) **	9.9 (0.6–50.1)	4.3 (0.3–19.0)	0.006	13.7 (0–39.2)	4.6 (1.4–34.4)	<0.001	NS	NS
Patients achieving EASI 90	4 (10.9%)	5 (31.3%)	0.027	11 (11%)	9 (34.6%)	0.041	NS	NS
Patients achieving EASI 75	15 (32.6%)	7 (43.8%)	0.02	28 (28.3%)	6 (30.8%)	NS	NS	NS
Patients with at least 1 adverse effect ***	8 (17.4%)	2 (12.5%)	NS	0	0	NS	NS	NS

Legend: * Intermittent AD indicates a course with flares followed by prolonged periods with no or mild disease, as it was reported by patients (POEM score <7); ** all differences of baseline versus final EASI scores were significantly different at a $p < 0.001$ level; *** Grade I burn, itching and dry skin only. Severe adverse effects or adverse effects leading to treatment discontinuation were never seen.

There were no statistically significant differences in the comparison of these features among the group of patients treated with the two phototherapies (Table 1). Both therapies were effective with a statistically significant lower median final EASI in comparison to the median baseline EASI score: final EASI score of 9.90 (0.6–50.1) with UVA1 and 13.70 (0–39.2) with NB-UVB. The measures of treatment outcome (the rate of patients achieving a 0/1 IGA result, the rate of patients with a 0/1 IGA persistent for at least 6 months, the rate of patients with EASI 90 and EASI 75 improvements) were good with both treatments (Table 1) without statistically significant differences when we compared the results obtained with medium-dose UVA1 phototherapy versus the results obtained with NB-UVB phototherapy. The median (range) number of treatment sessions was 30 (12–66) with UVA1 and 33 (16–51) with NB-UVB (p = NS).

A receiver-operating characteristic (ROC) analysis of the 2013–2017 findings was performed for an exploratory evaluation of the cut-off baseline EASI score for the achievement of an IGA 0/1 treatment result, and we found that it was 24.4 (area under the curve (AUC) = 0.925 sensitivity with $p < 0.001$) with UVA1 phototherapy and 24.7 with NB-UVB phototherapy (AUC = 0.923 sensitivity with $p < 0.001$) (Figure 1).

Patients achieving an IGA 0/1 remission persistent for at least 6 months numbered 14 of 19 (73.7%); 22 of 34 (64.7%) patients had a history of intermittent disease; and 1 of 27 (3.7%) and 6 of 65 (9.2%) patients had a history of continuous disease with UVA1 phototherapy and NB-UVB phototherapy, respectively, without statistically significant differences between treatments. Afterwards, we analyzed the treatment results on the bases of both the cut-off baseline EASI scores from the ROC curves and the medical history of continuous or intermittent course; we found that the rate of patients achieving an IGA 0/1 improvement persistent for at least 6 months was statistically significantly higher for patients with a baseline EASI score lower than the cut-off scores and a history of intermittent disease ($p < 0.001$) with both phototherapies (9/13 (69.2%) patients with medium-dose UVA1 and 19/26 (73.1%) with NB-UVB), in comparison to patients with a higher baseline EASI score and/or a history of continuous moderate or severe skin involvement (Table 2).

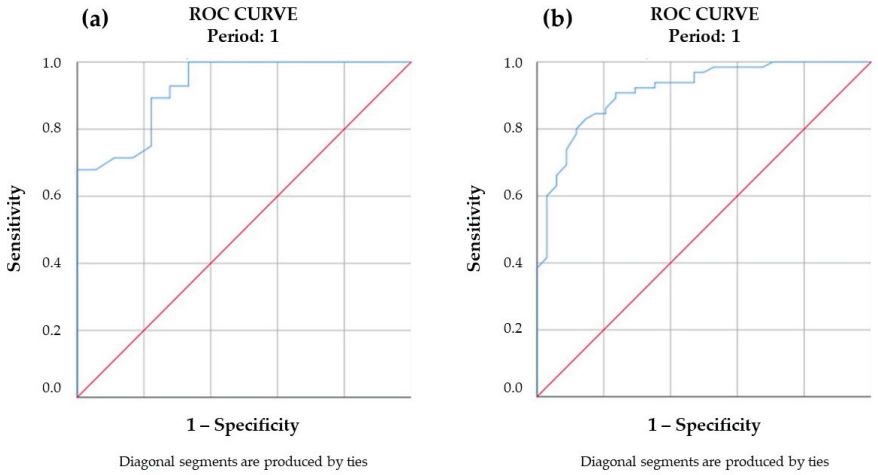

Figure 1. ROC curve (Receiver Operating Characteristic) of baseline EASI score (Eczema Area and Severity Index) and IGA (Investigator's Global Assessment) 0/1 result in patients treated with medium-dose UVA1 (ultraviolet A1) phototherapy (**a**) and NB-UVB (narrow-band ultraviolet B) phototherapy (**b**). The test result variable(s): baseline EASI score has at least one tie between the positive actual state group and the negative actual state group. Statistics may be biased. The cut-off values of the baseline EASI score are 24.4 with medium-dose UVA1 phototherapy and 24.7 with NB-UVB phototherapy.

Table 2. Patients achieving an IGA score (Investigator's Global Assessment) 0/1 improvement persistent for at least 6 months from 2013 to 2017. Treatment results were analyzed on the bases of the combination of the baseline EASI score (Eczema Area and Severity Index) and the clinical course of the skin lesions. UVA1 = ultraviolet A1; NB-UVB = narrow-band ultraviolet B.

Baseline EASI	Clinical Course	Treated Number of Patients	IGA 0/1 Persistent for at Least 6 Months	Rate (%) of Patients Achieving Persistent IGA 0/1	*p*
Patients treated with medium-dose UVA1 phototherapy (n = 46)					
<24.4	intermittent	13	9	69.2%	<0.001
<24.4	continuous	2	1	50%	
>24.5	intermittent	6	2	33.3%	
>24.5	continuous	25	3	12%	
Patients treated with NB-UVB phototherapy (n = 99)					
<24.7	intermittent	26	19	73.1%	<0.001
<24.7	continuous	6	1	16.7%	
>24.8	intermittent	8	2	25.0%	
>24.8	continuous	59	6	10.2%	

Therefore, from 2018, we only enrolled patients with a baseline EASI lower than 24.4 with UVA1 and 24.7 with NB-UVB, and an intermittent course of skin manifestations. Therefore, the patients who had begun a first treatment cycle after 2018 were fewer than the patients who were treated for the first time in the 5 years before with both UVA1 (16 versus 46 patients) and NB-UVB phototherapy (26 versus 99 patients) (Table 1). There was no statistically significant difference in age, gender and skin phototype (Table 1). The median (range) EASI scores of patients treated with both UVA1 and NB-UVB phototherapy were significantly lower: 18.7 (8.6–27.4) versus 28.7 (8.7–52.2) ($p < 0.001$) and 18.4 (9.1–26.3) versus 30.5 (7.9–52.2) ($p < 0.001$), respectively (Table 1). There were no significant differences between the baseline EASI scores of patients treated with the two phototherapies from 2018 to 2022 (p = NS).

Patients treated from 2018 to 2022 underwent a significantly lower median number of treatments (30 (12–66) versus 18.5 (12–26) ($p < 0.001$) with UVA1, and 33 (16–51) versus 24 (12–30) ($p < 0.001$) with NB-UVB) and received a lower cumulative UV dose (1155.0 (360–3080) J/cm^2 versus 770 (230–1750) J/cm^2 with UVA1, and 16.3 (0.63–30.8) J/cm^2 versus 11.3 (3.6–28.5) J/cm^2 with NB-UVB) than patients treated from 2013 to 2017 with both phototherapies (Table 1).

The rates of patients who maintained an IGA 0/1 remission for at least 6 months were higher in the group treated after 2017: 11/16 (68.7%) versus 15/46 (33%) ($p = 0.012$) with UVA1 phototherapy, and 18/26 (69.2%) and 28/99 (28%) ($p = 0.016$) with NB-UVB phototherapy. The comparisons of the rates of patients achieving a persistent IGA 0/1 result with the two phototherapies from 2018 to 2022 were never significantly different (Table 1).

The median EASI scores decreased significantly with both phototherapies from the baseline EASI score (Figure 2), but the final median EASI score was significantly lower in the patients treated between 2018 and 2022 in comparison to those treated between 2012 and 2017: 4.3 (0.3–19.0) versus 9.9 (0.6–50.1) ($p < 0.006$) with UVA1 phototherapy, and 4.6 (1.4–34.4) versus 13.7 (0–39.2) ($p < 0.001$) with NB-UVB phototherapy (Table 1). The comparison of the final EASI scores with the two phototherapies did not show differences at a statistically significant level (Table 1).

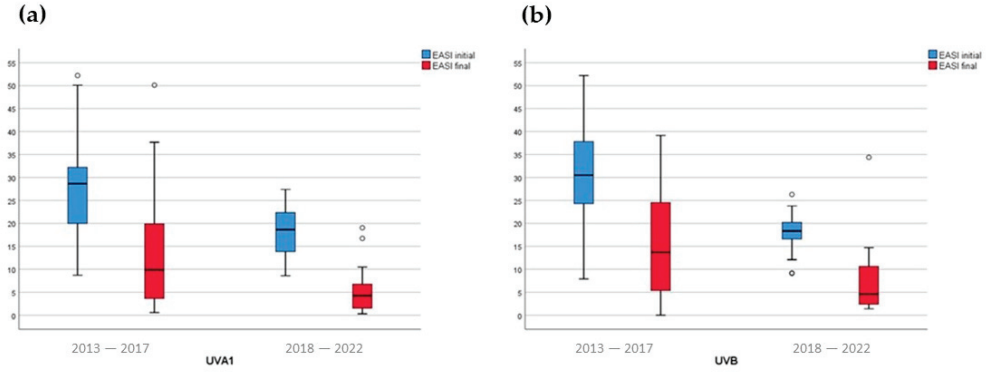

Figure 2. Baseline and final EASI scores (Eczema Area and Severity Index) with medium-dose UVA1 (ultraviolet A1) (**a**) and NB-UVB (narrow-band ultraviolet B) (**b**) phototherapies. The comparisons of median baseline and final EASI scores were always statistically different in all groups. The baseline EASI scores and the EASI scores of patients were significantly higher in the patients treated in the 2013–2017, in comparison to those treated in the 2018–2022 group with both phototherapies. There was no statistically significant difference comparing the baseline EASI scores and the final EASI scores of patients treated with UVA1 phototherapy or NB-UVB phototherapy in the periods 2013–2017 and 2018–2022.

The rates of patients achieving EASI 90 were higher in the group treated from 2018 to 2022 in comparison to the group treated from 2013 to 2017 (Table 1): 5/16 (31.3%) versus 4/46 (10.9%) ($p = 0.027$) with UVA1, and 9/26 (34.6%) and 11/99 (11%) ($p = 0.041$) with NB-UVB; and the rates of patients achieving EASI 75 were 7/16 (43.8%) versus 15/46 (32.6%) ($p = 0.02$) with UVA1, and 6/26 (30.8%) and 28/99 (28.3%) (p = NS) with NB-UVB.

The rates of patients with at least one mild adverse effect (burns, dry skin and itching) were always low, and they were not statistically different in the patients treated in the two time intervals with both phototherapies (Table 1). They were always quickly responsive to an emollient cream. We never registered withdrawals due to adverse events, and all patients completed the treatment cycle.

4. Discussion

The availability of biologics and small molecules has set a new paradigm of AD treatment that is not only a complete or near-complete clearance at the end of treatment, but also a durable remission over time with the prevention of acute flares. In this new treatment landscape, the most recent guidelines still recommend NB-UVB and medium-dose UVA1 phototherapies as valuable therapeutic options, but they do not specify for which patients their use is preferable to other treatment alternatives. In the present study, the analyses of the treatment findings of patients treated between 2013 and 2017 (Table 1) showed that patients with moderate and severe disease, with baseline EASI < 24.7 with NB-UVB and <24.4 with medium-dose UVA1 (Figure 1), and a history of an intermittent course with prolonged periods of no or mild disease between acute flares, frequently achieved a complete or near-complete (IGA 0/1) remission that persisted for more than 6 months. The duration of the remission is a particularly important issue because phototherapies cannot be repeated frequently, and maintenance treatments are discouraged, due to the risk of long-term side effects, namely skin carcinogenesis. Therefore, in the present study, we found that not only the assessment of baseline cut-off scores of clinical severity [11], but also the understanding of the individual longitudinal course of AD, may improve clinical phenotyping and prognostication and facilitate a personalized therapeutic recommendation [9,12]. Indeed, it has previously been underlined that a single, static (one point in time) measurement of severity may overestimate or underestimate the true AD severity experienced by the patient, given the characteristic fluctuating severity characterized by an infinite number of combinations of disease flares, long-term persistence, and quiescence [12,19,20].

These patient selection criteria (a baseline EASI score lower than the above-mentioned cut-off values and a medical history characterized by intermittent disease) led to a sharp decrease in the number of patients who began a first treatment cycle, but we observed a statistically significant improvement of all parameters of treatment efficacy with higher rates of patients achieving a IGA 0/1 result and maintaining it for at least 6 months, a lower median final EASI score, and statistically higher rates of patients achieving EASI90 and EASI75 with both phototherapies.

The comparison of the efficacy measures and safety of medium-dose UVA1 and NB-UVB in the two treatment periods did not show statistically significant differences. These results are in general agreement with the findings of previous meta-analyses showing that both phototherapies are effective, without clear differences, for improving physician-rated signs and patient-reported symptoms [3,6,21].

However, we emphasize that this study was not designed as a randomized controlled comparative clinical trial and the patients were not selected for one treatment or the other, according to precise enrollment criteria [22,23]. In our daily clinical routine, the choice was made on the basis of the practical aspects of two therapies, e.g., the duration of the exposures, the heat in the irradiation unit, the lying or standing position and the cost and the duration of the waiting list at the time [24].

The results of phototherapies can be influenced by differences in the treatment protocol. We used fixed UV doses (50 J/cm^2) for UVA1 because there is clinical evidence that medium-dose (30–60 J/cm^2) UVA1 is more effective than low-dose regimens (10–20 J/cm^2) and equally effective as high-dose (80–120 J/cm^2) regimens, with a lower risk of long-term adverse effects, such as photo-aging and carcinogenic potential [6]. The NB-UVB protocol, with an initial dose based on the skin type and cautious dose increments at each exposure, is a good compromise of high efficacy and a reduced risk of adverse effects [3,9,10,25]. Moreover, the treatment choice of one phototherapy or the other cannot be driven by the present knowledge of the action mechanisms.

Indeed, NB-UVB and UVA1 phototherapies have different photochemical and photobiological mechanisms, different penetration into the skin, intracellular targets and preferential target cell populations [26]. However, the relevant biological effects involved in the change of the pathophysiology of AD are largely similar, e.g., modulatory effects on both innate and acquired immunity with a reduction in the number and functionality of

IgE-bearing intraepidermal Langerhans cells and dermal mast cells; rapid proapoptotic activity on T and B lymphocytes; a reduction in the synthesis and release of proinflammatory cytokines, such as tumor necrosis factor-alpha (TNF-α), IL-3, IL-4, IL-5, IL-12 and IL-13; and the inhibition of calcineurin phosphatase and eosinophil cationic protein (ECP), as well as antipruritic, antifibrotic, pro-pigmentary and pro-prebiotic-effects [4,26–29].

5. Conclusions

In this study, we found that phototherapies remain a valuable treatment option for patients with moderate and severe AD provided that they simultaneously present two clinical criteria: (1) the baseline EASI is lower than 24.4 with UVA1 and 24.7 with NB-UVB; and (2) at medical history, they report prolonged spontaneous or treatment-induced periods of complete or near-complete remission. However, their use for patients with more severe AD and/or patients with continuous involvement and quick relapses after treatments should be carefully evaluated because of the more limited efficacy, the high risk of early relapses and the risk of long-term adverse effects, namely skin carcinogenesis and skin aging, if frequent and prolonged treatment cycles are delivered. Fortunately, these patients can now benefit from new immunotherapies which are highly effective and allow long-term control of the disease [2,9]. The main limitations of this study are the retrospective, uncontrolled design and the relatively small number of patients enrolled.

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Institutional Review Board Statement: The study was conducted in accordance with the Declaration of Helsinki and approved by the Ethics Committee of the ASST Spedali Civili di Brescia (protocol code NP3720).

Informed Consent Statement: Informed consent was obtained from all subjects involved in the study.

Data Availability Statement: The data presented in this study are available on request from the corresponding author.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Weidinger, S.; Novak, N. Atopic Dermatitis. *Lancet* **2016**, *387*, 1109–1122. [CrossRef]
2. Schneeweiss, M.C.; Perez-Chada, L.; Merola, J.F. Comparative Safety of Systemic Immunomodulatory Medications in Adults with Atopic Dermatitis. *J. Am. Acad. Dermatol.* **2021**, *85*, 321–329. [CrossRef]
3. Musters, A.H.; Mashayekhi, S.; Harvey, J.; Axon, E.; Lax, S.J.; Flohr, C.; Drucker, A.M.; Gerbens, L.; Ferguson, J.; Ibbotson, S.; et al. Phototherapy for Atopic Eczema. *Cochrane Database Syst. Rev.* **2021**, *10*, CD013870. [CrossRef]
4. Kemény, L.; Varga, E.; Novak, Z. Advances in Phototherapy for Psoriasis and Atopic Dermatitis. *Expert Rev. Clin. Immunol.* **2019**, *15*, 1205–1214. [CrossRef] [PubMed]
5. Sidbury, R.; Davis, D.M.; Cohen, D.E.; Cordoro, K.M.; Berger, T.G.; Bergman, J.N.; Chamlin, S.L.; Cooper, K.D.; Feldman, S.R.; Hanifin, J.M.; et al. Guidelines of Care for the Management of Atopic Dermatitis: Section 3. Management and Treatment with Phototherapy And Systemic Agents. *J. Am. Acad. Dermatol.* **2014**, *71*, 327–349. [CrossRef]
6. Meduri, N.B.; Vandergriff, T.; Rasmussen, H.; Jacobe, H. Phototherapy in the Management of Atopic Dermatitis: A Systematic Review. *Photodermatol. Photoimmunol. Photomed.* **2007**, *23*, 106–112. [CrossRef]
7. Beck, L.A.; Thaçi, D.; Hamilton, J.D.; Graham, N.M.; Bieber, T.; Rocklin, R.; Ming, J.E.; Ren, H.; Kao, R.; Simpson, E.; et al. Dupilumab Treatment in Adults with Moderate-to-Severe Atopic Dermatitis. *N. Engl. J. Med.* **2014**, *371*, 130–139. [CrossRef] [PubMed]
8. European Medical Agency. Available online: https://www.ema.europa.eu/en/medicines/human/EPAR/dupixent (accessed on 1 March 2023).

9. Wollenberg, A.; Kinberger, M.; Arents, B.; Aszodi, N.; Avila Valle, G.; Barbarot, S.; Bieber, T.; Brough, H.A.; Calzavara Pinton, P.; Christen-Zäch, S.; et al. European Guideline (EuroGuiDerm) on Atopic Eczema: Part I—Systemic Therapy. *J. Eur. Acad. Dermatol. Venereol.* **2022**, *36*, 1409–1431. [CrossRef] [PubMed]
10. Eichenfield, L.F.; Tom, W.L.; Berger, T.G.; Krol, A.; Paller, A.S.; Schwarzenberger, K.; Bergman, J.N.; Chamlin, S.L.; Cohen, D.E.; Cooper, K.D.; et al. Guidelines of Care for the Management of Atopic Dermatitis: Section 2. Management and Treatment of Atopic Dermatitis with Topical Therapies. *J. Am. Acad. Dermatol.* **2014**, *71*, 116–132. [CrossRef] [PubMed]
11. Simpson, E.L.; Bruin-Weller, M.; Flohr, C.; Ardern-Jones, M.R.; Barbarot, S.; Deleuran, M.; Bieber, T.; Vestergaard, C.; Brown, S.J.; Cork, M.J.; et al. When Does Atopic Dermatitis Warrant Systemic Therapy? Recommendations from an Expert Panel of the International Eczema Council. *J. Am. Acad. Dermatol.* **2017**, *77*, 623–633. [CrossRef] [PubMed]
12. Chovatiya, R.; Silverberg, J.I. Evaluating the Longitudinal Course of Atopic Dermatitis: Implications for Clinical Practice. *Am. J. Clin. Dermatol.* **2022**, *23*, 459–468. [CrossRef] [PubMed]
13. Calzavara-Pinton, P.G.; Sala, R.; Arisi, M.; Rossi, M.T.; Venturini, M.; Ortel, B. Synergism between Narrowband Ultraviolet B Phototherapy and Etanercept for the Treatment of Plaque-type Psoriasis. *Br. J. Dermatol.* **2013**, *169*, 130–136. [CrossRef] [PubMed]
14. Leshem, Y.A.; Hajar, T.; Hanifin, J.M.; Simpson, E.L. What the Eczema Area and Severity Index Score Tells Us about the Severity of Atopic Dermatitis: An Interpretability Study. *Br. J. Dermatol.* **2015**, *172*, 1353–1357. [CrossRef]
15. Hanifin, J.M.; Thurston, M.; Omoto, M.; Cherill, R.; Tofte, S.J.; Graeber, M.; Evaluator Group, T.E. The Eczema Area and Severity Index (EASI): Assessment of Reliability in Atopic Dermatitis. *Exp. Dermatol.* **2001**, *10*, 11–18. [CrossRef]
16. Calzavara-Pinton, P.; Zanca, A.; Arisi, M.; Rossi, M.T.; Zane, C.; Venturini, M.; Ortel, B. Prevalence of Phototherapy in the Age of Biologics. *Dermatology* **2018**, *234*, 166–172. [CrossRef]
17. Agenzia Italiana del Farmaco (AIFA). Available online: https://www.aifa.gov.it/-/attivazione-web-pubblicazione-registro-dupixent (accessed on 1 March 2023).
18. Futamura, M.; Leshem, Y.A.; Thomas, K.S.; Nankervis, H.; Williams, H.C.; Simpson, E.L. A Systematic Review of Investigator Global Assessment (IGA) in Atopic Dermatitis (AD) Trials: Many Options, No Standards. *J. Am. Acad. Dermatol.* **2016**, *74*, 288–294. [CrossRef]
19. Hong, M.R.; Lei, D.; Yousaf, M.; Chavda, R.; Gabriel, S.; Janmohamed, S.R.; Silverberg, J.I. A Real-World Study of the Longitudinal Course of Adult Atopic Dermatitis Severity in Clinical Practice. *Ann. Allergy Asthma Immunol.* **2020**, *125*, 686–692.e3. [CrossRef]
20. Zuberbier, T.; Orlow, S.J.; Paller, A.S.; Taïeb, A.; Allen, R.; Hernanz-Hermosa, J.M.; Ocampo-Candiani, J.; Cox, M.; Langeraar, J.; Simon, J.C. Patient Perspectives on the Management of Atopic Dermatitis. *J. Allergy Clin. Immunol.* **2006**, *118*, 226–232. [CrossRef] [PubMed]
21. Pérez-Ferriols, A.; Aranegui, B.; Pujol-Montcusí, J.A.; Martín-Gorgojo, A.; Campos-Domínguez, M.; Feltes, R.A.; Gilaberte, Y.; Echeverría-García, B.; Alvarez-Pérez, A.; García-Doval, I. Phototherapy in atopic dermatitis: A systematic review of the literature. *Actas Dermo-Sifiliogr.* **2015**, *106*, 387–401. [CrossRef]
22. Sastre, J.; Baldrich, E.S.; Armario Hita, J.C.; Herráez, L.; Jáuregui, I.; Martín-Santiago, A.; Ortiz de Frutos, J.; Silvestre, J.F.; Valero, A. Consensus on the Clinical Approach to Moderate-to-Severe Atopic Dermatitis in Spain: A Delphi Survey. *Dermatol. Res. Pract.* **2020**, *2020*, 1524293. [CrossRef]
23. Vermeulen, F.M.; Gerbens, L.A.A.; Schmitt, J.; Deleuran, M.; Irvine, A.D.; Logan, K.; Ouwerkerk, W.; Vestergaard, C.; Flohr, C.; Spuls, P.I. The European TREatment of ATopic Eczema (TREAT) Registry Taskforce Survey: Prescribing Practices in Europe for Phototherapy and Systemic Therapy in Adult Patients with Moderate-to-severe Atopic Eczema. *Br. J. Dermatol.* **2020**, *183*, 1073–1082. [CrossRef] [PubMed]
24. Krutmann, J. Phototherapy for Atopic Dermatitis. *Clin. Exp. Dermatol.* **2000**, *25*, 552–558. [CrossRef] [PubMed]
25. Rodenbeck, D.L.; Silverberg, J.I.; Silverberg, N.B. Phototherapy for Atopic Dermatitis. *Clin. Dermatol.* **2016**, *34*, 607–613. [CrossRef] [PubMed]
26. Vieyra-Garcia, P.A.; Wolf, P. A Deep Dive into UV-Based Phototherapy: Mechanisms of Action and Emerging Molecular Targets in Inflammation and Cancer. *Pharmacol. Ther.* **2021**, *222*, 107784. [CrossRef]
27. Tintle, S.; Shemer, A.; Suárez-Fariñas, M.; Fujita, H.; Gilleaudeau, P.; Sullivan-Whalen, M.; Johnson-Huang, L.; Chiricozzi, A.; Cardinale, I.; Duan, S.; et al. Reversal of Atopic Dermatitis with Narrow-Band UVB Phototherapy and Biomarkers for Therapeutic Response. *J. Allergy Clin. Immunol.* **2011**, *128*, 583–593.e4. [CrossRef]
28. Legat, F.J. The Antipruritic Effect of Phototherapy. *Front. Med.* **2018**, *5*, 333. [CrossRef]
29. Vieyra-Garcia, P.A.; Wolf, P. From Early Immunomodulatory Triggers to Immunosuppressive Outcome: Therapeutic Implications of the Complex Interplay Between the Wavebands of Sunlight and the Skin. *Front. Med.* **2018**, *5*, 232. [CrossRef]

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