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Review

Potential Applications of Yeast Biomass Derived from Small-Scale Breweries

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Abstract: Yeast biomass, a brewery by-product of the world's substantial alcohol beverage industry, finds successful applications in the fodder industry and food additive production. This is attributed to its rich nutritional profile that comprises high protein and vitamin content. Nonetheless, in small-scale breweries, yeast slurries present a significant challenge, as the quantities obtained are insufficient to attract the attention of the food industry. The disposal of yeast contributes substantially to the organic load of wastewater (approximately 40%) and elevates water consumption (3–6 hL/hL of beer), consequently escalating production costs and environmental impact. In recent years, diverse potential applications of products derived from yeast biomass have emerged, encompassing the substitution of sera in cell culture media, the fortification of animal feed with vitamins and selenium, the utilization of beta-glucan in low-fat food products, and the development of functional foods incorporating yeast-derived peptides. These peptides exhibit the potential to safeguard the gastric mucosa, prevent hypertension, and address neurodegenerative disorders. The rising demand for value-added products derived from yeast underscores the potential profitability of processing yeast from small breweries. Due to the high equipment costs associated with yeast biomass fractionation, the establishment of specialized facilities in collaboration with multiple small breweries appears to be the most optimal solution.

Keywords: yeast biomass; functional foods; yeast nutritional values; single-cell protein

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1. Beer Production Statistics

Beer stands as the most widely consumed alcoholic beverage globally. The global production of beer has remained relatively stable over the past decade, ranging between 1.91 and 1.97 billion hectoliters. China leads as the foremost beer producer, with its 2022 production exceeding 420 million hectoliters, constituting approximately 22% of the global output. In the Americas, North America and Central and South America contribute 222 million hectoliters and 360 million hectoliters, respectively. Within Europe, the annual production exceeds 496 million hectoliters. A breakdown of the share of beer production by continent is illustrated in Figure 1. In Europe, Germany emerges as the top producer, boasting a rich brewing tradition. In 2022, Germany produced 78 million hectoliters of beer, predominantly comprising lager and wheat beer. Following Germany in European production are the United Kingdom, Spain, and Poland. The pooled output for these four countries hovers at around 206 million hectoliters.

In global beer production, a significant portion of the market is dominated by several multinational corporations. In 2022, the largest player in this industry was AB InBev, responsible for 27.4% of the world's beer production, and the top 40 international brewing companies control up to 88.1% of the global market [1]. However, in recent years, there has been a noticeable increase in the share of craft breweries in the market, both in areas

with a strong brewing tradition and in areas where the industry is gaining importance. The terms "craft brewery", "microbrewery", and "local brewery" refer to small businesses that produce beer, often trying to create both traditional and innovative brews, and operating independently of large brewing corporations [2]. The scale of production of these breweries is not precisely defined, but they share a common philosophy based on the use of unconventional ingredients and continuous innovations in the production process. In the United States, small breweries hold a significant share of the market, accounting for 23.3% of total beer production in the country in 2017, with a total annual production of 224 million hectoliters [3]. According to reports prepared by Deloitte for the Polish Breweries Association, in 2017, small breweries accounted for about 3.5% of total beer production in Poland, with a total production of 39.9 million hectoliters [4]. However, in 2019 and 2020, small breweries increased their market share to 4%, with total production reaching 40.1 and 39.4 million hectoliters annually, respectively [5].

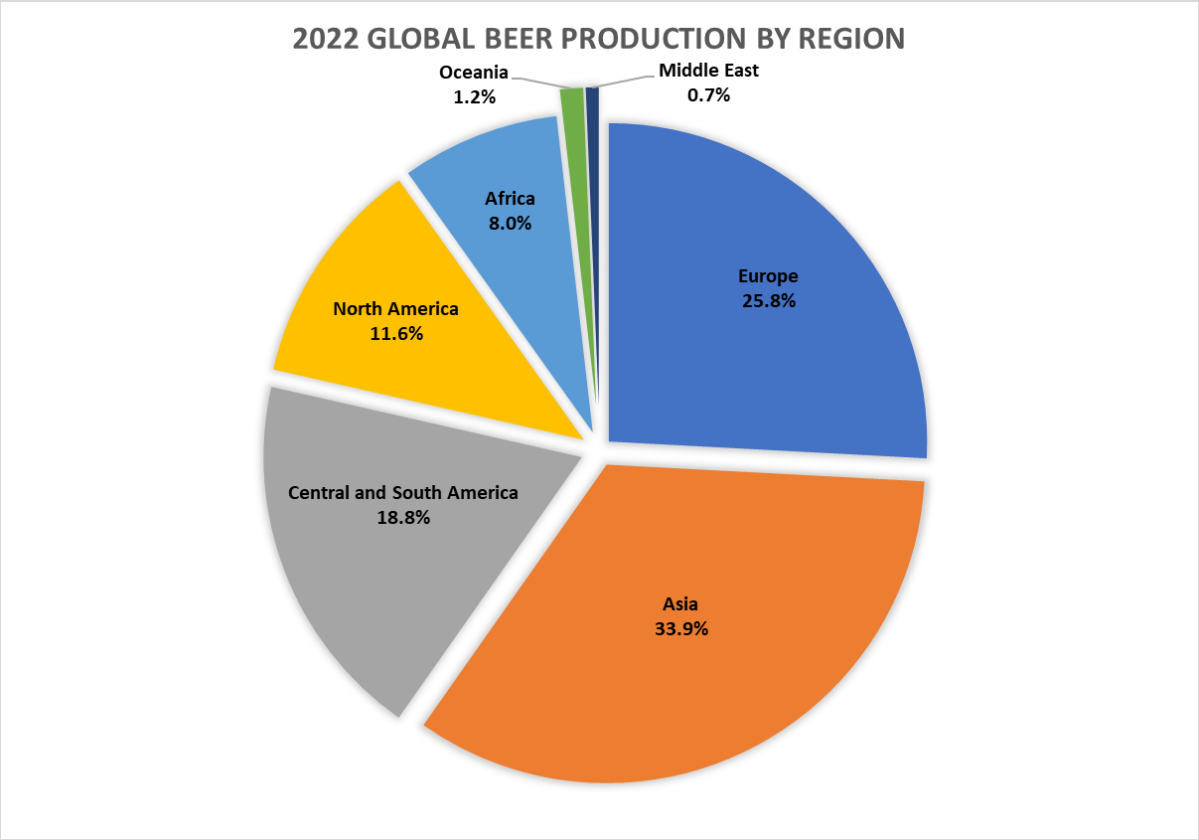

Figure 1. World beer production share in 2022 [1].

The presented data underscore the global distribution of beer production across diverse regions worldwide. As such, research endeavors focused on the manufacturing, consumption, health implications, and sustainable utilization of by-products generated in the beer production process are deemed essential. These aspects have the potential to impact a vast number of individuals on a global scale.

2. Brewery Industry Waste and By-Products

Over the past three decades, extensive research and practical implementations have introduced numerous technologies to the brewing industry, affording notable efficiencies in production, notably those that curtail the generation of by-products. Nevertheless, certain waste streams inherently associated with beer production are difficult to reduce. New strategies in brewery waste management should not only facilitate disposal but also aim to extract additional value from processing whenever feasible. Residual materials, including

spent grain, hot trub generated in the brewing process, and brewer's yeast, present disposal challenges owing to their complex physicochemical properties and microbiological activity. The disposal process itself is characterized by considerable costs [6]. According to data from the European Environment Agency in 2022, the average cost of waste disposal in Poland was EUR 60 per ton [7]. However, as indicated by the "Committed to the Environment" report, this value could increase by an additional EUR 533 per ton of waste by 2030 [8]. Taking into account only the spent grain (20 kg/hL) and yeast (3 kg/hL) in the calculations, the additional cost of waste disposal could reach up to EUR 13.64 per hectoliter of beer. These aforementioned waste components also contribute to a loss of up to 20 L of water per every 100 L utilized in the production process, and this is particularly pronounced in instances involving hot trub and brewer's yeast, where the water content can constitute up to 90% of the overall volume [9].

The disposal of brewing waste generates numerous ecological challenges due to its substantial nutritional value and elevated concentrations of organic compounds, thereby imposing a significant chemical and 5-day biochemical oxygen demand (COD and BOD5) during degradation [10]. Given various factors that encompass environmental policy, the advocacy for the "zero waste" philosophy, the presently observed scarcity of non-renewable resources, and challenges associated with the inappropriate utilization of renewable resources collectively necessitate the development of innovative technologies aimed at minimizing waste generation or facilitating the reutilization of produced waste in order to enhance the overall value added.

The largest beer companies, representing approximately 95% of the global beer production worldwide, possess an extensive infrastructure tailored for the efficient treatment of production-derived wastewater. In addition, these establishments yield substantial volumes of valuable by-products; thanks to their abundant supply, economic viability is realized through various processing methods, such as the incorporation of by-products into animal feed production. Conversely, the minority share of the market is served by small craft breweries, whose production outputs of most by-products are insufficient for economically viable utilization. Consequently, these smaller enterprises confront the challenge of disposing of by-products effectively, which, nonetheless, constitute valuable substrates for diverse sectors of the industrial landscape.

While the issue of spent grain presents a relatively minor challenge, with a supply ranging from 14 to 20 kg/hL deemed sufficient for small-scale farmers or modest processing facilities, more formidable difficulties arise in the context of yeast slurry [9]. The latter constitutes a significant environmental burden, and the prevailing methods of its disposal are economically infeasible in the face of limited supply levels.

Post-production yeast slurry is not added to brewing wastewater, because the wastewater is already heavily polluted with contaminants from different production stages, including cleaning. Yeast slurries are characterized by a high content of organic matter (BOD5: 76,000 mg/g of DW; 1250–1350 mg of COD/g of DW; COD calculation details in Table S3), nitrogen (up to 7.5% of DW), and phosphorus (around 1% of DW) [11,12]. The general characteristics of wastewater generated from breweries are presented in Table 1. The wastewater load attributed to the brewing industry, contingent upon the specific parameter measured, spans from 800 to an elevated 38,000 mg/L for COD, and from 1005 to an upper limit of 50,000 mL/g for biochemical five-day oxygen demand (BOD5). The total suspended solids (TSSs) exhibit values oscillating between 200 and 3000 mg/L (a detailed compilation is available in Table S1 in the Supplementary Materials Section). The introduction of yeast into the wastewater stream would escalate parameters, notably the COD and BOD5, beyond levels accepted by conventional treatment plants (see Tables 1 and S2 in Supplementary Materials). The latter generally accommodates wastewater parameters within the range of 160 to 540 mg/L for BOD5, from 497 to 1580 mL/L for COD, and from 177 to 1260 mg/L for TSSs. The cited data elucidate the recurrent transgression of established regulatory norms by wastewater generated within the brewing industry [13]. Hence, the imperative arises for the exploration of alternative methodologies

for the management of solid waste, with specific emphasis on yeast slurry emanating from the brewing industry.

Table 1. Parameters for wastewater streams.

Parameter	Generated in Brewery	Accepted by Treatment Plant	EUNorm for Purified Wastewater
BOD 5 [mg/L O_2]	1000–50,000	100–940	25
COD [mg/L O_2]	800–38,000	109–1668	125
TSS [mg/L]	200–3000	78–1260	35
Total P [mg/L]	4–100	2–40	1–2
Total N [mg/L]	20–100	20–210	10–15
Wastewater-to-beer ratio [hL/hL]	2.2–8.7	-	-
References	[13–15]	[16–18]	[19]

Ranges based on data presented in Tables S1 and S2 in Supplementary Materials.

3. Fermentative Yeast Growth

Yeasts represent a versatile group of fungi broadly used in the beverage industry, especially in beer production. The strains that are the most important to breweries belong to the species *Saccharomyces cerevisiae* and *Saccharomyces pastorineus* (a hybrid of *S. cerevisiae* and *S. bayanus*). Under the anaerobic conditions used in beverage production, *S. cerevisiae* ferments sugars to obtain energy for growth. In this metabolic pathway, a hexose molecule (preferentially glucose) is converted into two ethanol molecules and two molecules of carbon dioxide, and two ATP molecules are generated (Figure 2). Since only a small portion of energy is captured by the cells, the majority of sugar available in the growth medium is converted into ethanol, and the biomass gain is limited. In anaerobic conditions, the sugar-to-biomass conversion ratio is below 0.1, while during glucose-limited aerobic growth, this ratio is close to 0.5 [20]. Taking into account the initial sugar concentration in wort ranging between 10 and 15% (*w*/*v*), in a properly executed process utilizing *Saccharomyces cerevisiae*, approximately 1.5 to 3 kg of yeast slurry that comprises 85 to 90% water can be produced for every 100 L of beer [9]. In this scenario, biomass production in beer fermentation is around 0.15 to 0.75 kg of dry weight per 1 hL of beer produced.

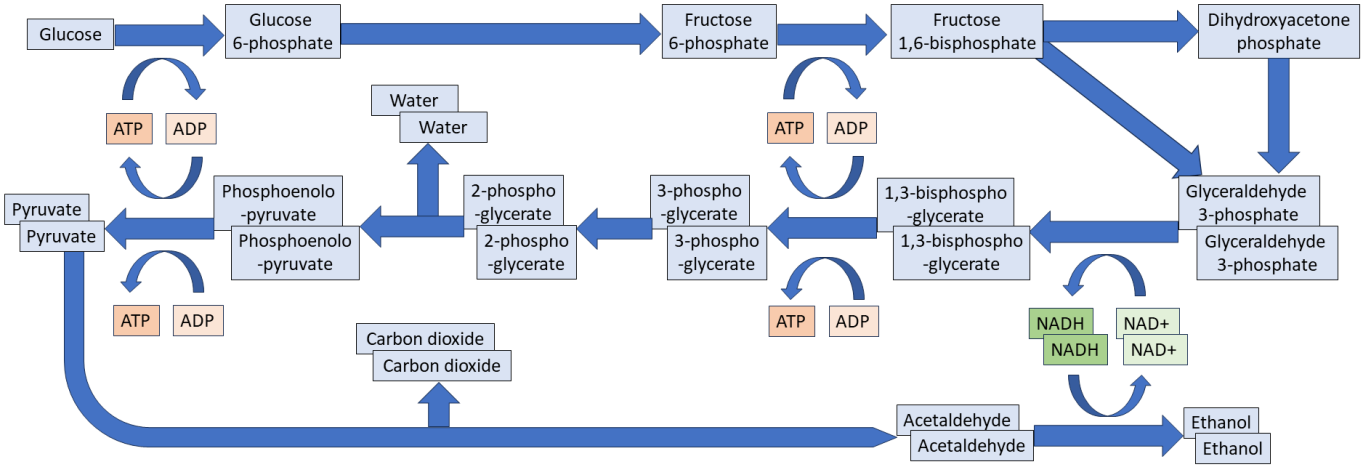

Figure 2. Main metabolic reactions leading to ethanol formation in brewery yeast.

Yeast cells also absorb nutrients such as free amino acids, short peptides, and vitamins from the wort. Amino acids may be divided into three groups according to the order and intensity of assimilation: rapidly assimilated (glutamic and aspartic acids, aspartate, glutamate, serine, threonine, lysine, and arginine), gradually absorbed (valine, methio-

nine, leucine, isoleucine, and histidine), and late assimilation (glycine, phenylalanine, tyrosine, tryptophan, alanine) [21]. Proline is incorporated into the yeast biomass to a very limited extent. For the majority of brewery yeast strains, biotin and pantothenate are required for proper growth, although strains requiring no biotin supplementation have been identified [22,23].

4. Brewery Yeast Biomass

Brewer's yeast represents an initially highly manageable by-product within the context of the beer production process, concurrently serving as a desired substrate across diverse industrial sectors. Not all batches of yeast obtained in the beer production process are considered waste due to the possibility of reuse in the fermentation of fresh wort. Following each inoculation event, the yeast population undergoes substantial augmentation, increasing by a factor of three to five. The frequency of reuse cycles is contingent upon several factors that involve the specific microbial species employed, the nature of the beer being produced, the composition of the brewing wort extract, and the meticulous adherence to hygiene practices. Conventionally, yeast is subjected to reuse cycles ranging from 3 to 10 iterations, or until such time that its application does not introduce noticeable alterations to the sensory attributes of the beer [24]. Consequently, with depleting recycling efficacy, the removal of yeast from the production process becomes imperative, thereby generating a waste stream that poses formidable challenges for disposal. Brewing yeast constitutes the second-largest by-product (after spent grain—20 kg/hL of beer [25]) in terms of mass within the framework of the beer brewing process. According to the previously provided data, the annual global beer production is approximately 1.9 billion hectoliters, with the mass of produced yeast exceeding 4.321 million tons (assuming an average biomass production of 2.25 kg per hectoliter of beer), resulting in about 864 thousand tons of dry biomass globally. The quantifiable production of biomass is contingent upon many factors, including the parameters governing fermentation (such as temperature, pH, and aeration), the taxonomy of microorganisms employed, the concentration and specifications of the inoculum, and the compositional intricacies of the nutrient milieu [26].

After fermentation is finished, yeast cells are set on the bottom of the fermentation tank and may be collected. Yeast sludge along the cells contains the remains of fermented wort (beer) and potentially some solid debris (such as hops). Beer usually contains the following components: ethanol (3–5%), carbohydrates (1–6%), hop-derived compounds (20–60 mg/L), and organic acids (50–250 mg/L). It also includes amino acids (mainly proline) and peptides (total nitrogen: 300–1000 mg/L) [27]. Moreover, yeast cell walls absorb bitter chemical compounds derived from hops (humulones and isohumulones) [28]. Depending on the further use of biomass, yeast cells may require additional washing to remove impurities.

The exemplary composition of biomass is presented in Table 2. The main groups of organic compounds constituting yeast cells are proteins (up to 49% DW) and carbohydrates (up to 54% DW).

Free amino acids constitute only a small part of the entire cell biomass (below 0.5% of DW). The amino acid composition of yeast biomass (including amino acids derived from protein) is presented in Table 3. The most abundant amino acids are glutamate/glutamic acid, aspartate/aspartic acid, alanine, leucine, and lysine. Notably, the proteins inherent in yeast slurry exhibit a noteworthy biological value, as evidenced by the quantity of essential amino acids within their structural composition, encompassing a range from 70 to 85% of the value observed in casein [9].

Table 2. Yeast biomass and yeast extract composition [% DW].

	Yeast Biomass	YE, Cell Mill	YE, Sonication	YE, Autolysis
Proteins/peptides	41–49	32.2	28.5	5.3
Free amino acids	0.2–0.4	11.6	15.5	30.6
Ribonucleic acids	1.9–7.5	7.5	7	5
Total sugars	22–54	31.2	31.4	28.8
Lipids	3.9	1	1.1	0.5
Ashes	1.7–8.5	13.1	13.3	13.2
Reference	[29]	[30]	[30]	[30]

Table 3. Amino acid composition for yeast biomass and protein concentrates (g/100 g total amino acids).

Amino Acid	Yeast Biomass	Yeast Extract (Total)	Yeast Extract (Free Amino Acids)	WHO Amino Acid Requirements
Lysine	7.13	7.5	3.2	4.5
Histidine	2.06	2.4	1.4	1.5
Threonine	6.16	4.6	2.7	2.3
Valine	6.2	6.2	4.0	3.9
Leucine	8.84	7.2	5.7	5.9
Isoleucine	5.64	5.0	3.4	3
Phenylalanine	5.3	4.6	3.5	3.8
Tyrosine	4.68	3.5	2.4	
Tryptophan	1.1	1.3	1.0	0.6
Methionine	2.5	1.6	1.2	1.6
Cystine	0.34	1.1	0.5	0.6
Glutamic acid + glutamine	13.15	15.8	5.2	
Aspartic acid + aspartate	11.98	10.5	4.0	
Serine	6.13	5.1	3.5	
Proline	4.45	5.8	3.3	
Alanine	7.07	7.6	6.0	
Glycine	4.93	5.2	2.2	
Arginine	4.11	5.1	3.0	
Source material type	Brewery yeast biomass	Yeast extract obtained from brewery yeast via lysis with papain and debittering		
Reference	[31]	[32]		[33]

Carbohydrates are mainly localized in the yeast cell wall and can be divided into the following groups: glucans (1–3 beta-glucans, 1–4 beta-glucans, and 1–6 beta-glucans), chitin, and mannans (bound to proteins localized in the cell wall). The share of individual polymers in the cell wall structure of yeast is presented in Table 4. The composition and structure of a brewery's yeast cell wall change depending on growth conditions and culture states [34]. Thicker cell walls seem to develop in rich culture media (like YPD) or under stress conditions such as low pH (4) or high temperature (37 °C) [35].

Table 4. Composition of yeast cell wall [35].

Cell wall	11–25	% cell DW
Chitin	1.4–6.9	% cell wall
Mannan	28–67	% cell wall
Total beta-glucan	32–57	% cell wall
1–6 beta-glucan	4.5–11.5	% cell wall

Yeast biomass also contains a considerable amount of water-soluble vitamins. The concentrations of individual vitamins in yeast biomass and yeast extracts are presented in Table 5. The most abundant vitamins in yeast biomass are niacin, thiamin, and pantothenic acid. The lowest concentrations are observed for biotin and cobalamin. The majority of *S. cerevisiae* strains cannot synthesize biotin, and draw it from the medium; thus, its intracellular level is limited by the biotin availability in the culture medium [23]. Since the concentration of biotin in wort is very low (0.1 mg/kg of DW), it may explain the low concentration of this vitamin in brewery yeast biomass [36].As already mentioned, after the fermentation process is finished, the yeast biomass may be collected and recycled for the production of beer from fresh wort. Prolonged storage time or inappropriate storage conditions (temperature above 15 °C) may result in a partial degradation of biomass due to autolysis [37]. This leads to reduced cell viability and attenuation, and may also result in increased concentrations of undesired compounds in beer, such as diacetyl. It may also lead to deterioration in the quality of products derived from such biomass.

Table 5. Vitamin content in brewery yeast biomass and yeast extracts [mg/g DW].

Vitamin	**Yeast Biomass**	**Yeast Extracts**				
Biotin (B7)	0.0013	0.5	1.14	1.28	1.38	6.19
Folic acid (B9)	0.0130	0.01	0.045	0.049	0.013	0.053
Niacin (B3)	0.3000	nd	0.94	1.04	0.68	0.79
Pantothenic acid (B5)	0.0700	0.08	0.2	0.19	0.16	0.43
Riboflavin (B2)	0.0400	0.13	0.002	0.002	0.001	0.011
Thiamine (B1)	0.1200	0.11	0.069	0.07	0.052	0.075
Pyridoxine (B6)	0.0280	1.76	0.049	0.051	0.031	0.059
Cobalamin (B12)	0.000001	0.17	0.0018	0.0012	0.011	0.0016
Source material type	Brewery yeast biomass	Dedicated yeast culture, autolyzed, supplied by Bio Springer AM Corporation	Brewery yeast biomass, cell mill	Brewery yeast biomass, sonication	Brewery yeast biomass, autolysis	*Saccharomyces* sp. biomass, industrial autolysis
Reference	[38]	[39]	[30]	[30]	[30]	[30]

5. Yeast Biomass Processing Technologies

Since brewery yeast sludge may contain different impurities, the initial processing focuses on removing undesirable particles. Large particles such as hops may be separated via mechanical sieving [32]. The remaining beer may be removed by centrifugation or filtration followed by washing with water [40]. Removal of hop-derived compounds bound to the yeast cell walls may be achieved with washing in basic solutions (pH > 9) [12].

Traditionally, yeast extract is prepared using an autolysis process. In this method, a yeast suspension containing around 10% of dry yeast biomass is heated up to around 50 °C in acidic conditions (pH = 5) and then incubated for 24 h [38]. During this process, biological membranes are disrupted and vacuolar proteolytic enzymes (such as proteinase

A and B, carboxypeptidase Y) are released and start to degrade proteins in the cell. This process produces a mixture of low-mass peptides and free amino acids. DNA and RNA also undergo hydrolysis, resulting in the production of oligonucleotides, nucleotides, and nucleosides. Enhanced production of monophosphate nucleotides, which are important flavor enhancers, may be achieved in altered autolysis conditions [41]. During autolysis, polymers present in the cell wall are partially hydrolyzed by the beta-glucanase enzyme, resulting in the disruption of cell walls. General degradation of cell elements finally leads to the release of hydrolysis products to the solution.

The efficiency of the autolysis process may be improved by the addition of sodium chloride (up to 3%), ethyl acetate, or isopropanol [38]. NaCl causes the separation of the plasma membrane from the cell wall due to reductions in cytoplasmic volume caused by differences in osmotic pressure. This process finally leads to the disruption of the lipid membrane and the release of hydrolytic enzymes. It may also be stimulated by the addition of Alcalase [42]. The hydrolysis of biopolymers derived from yeast may be enhanced by the addition of hydrolytic enzymes (proteases: trypsin, pepsin, papain, and others).

After autolysis is completed, the resulting fluid is heated up to 80 °C for 2 h to deactivate the remaining enzymes. In further steps, solid particles may be separated by centrifugation or filtration, and water-soluble compounds may be recovered in powder form after drying. Autolysis is considered to be a cost-effective process suitable to produce food additives. Water-soluble compounds may be further separated based on molecular mass using ultrafiltration techniques. Such approaches have been used in separating iron-interacting peptides from yeast extract [43].

The disruption of yeast cells may also be achieved with mechanical methods. Mechanical disruption includes cell mills (shaking with glass or steel balls), sonication, and French press [30]. These methods result in the fast disruption of cells and the release of intracellular content into the solution. Such approaches may be beneficial when separating biopolymers constituting cell wall or undigested proteins is considered.

Cell wall fractions may be further separated with the use of different methods. The earliest method for beta-glucan purification utilized incubation in an alkaline solution (3% NaOH), followed by incubation in an acidic solution (3% HCl) [44]. This protocol was later modified with additional steps including chromatography [45]. Later, a protocol based on hot water extraction was developed, leading to the isolation of mannoprotein fractions besides beta-glucan [46]. The authors claim that this procedure produces beta-glucan, which is unchanged in structure. If mannoproteins are the target, then protocols that rely on beta-glucan digestion with Zymolyase seem to be most efficient [47].

When yeast biomass is considered to provide a significant contribution to protein in human diets, reductions in nucleic acid content should be employed. This may be achieved through an enzymatic reaction following cell disruption or the separation of non-wall cell fractions. Hydrolyzed nucleotides may be separated from proteins via ultrafiltration technology.

The compositions of extracts vary depending on the biomass source and processing technology (see Tables 2, 4 and 5). Enzymatic lysis (including autolysis) generates more small molecules such as amino acids, nucleotides, and nucleosides. Autolysis also results in reduced vitamin extraction (except biotin); however, it does not cause major losses when compared to other methods. Surprisingly, yeast extracts seem to be substantially enriched in cobalamin when compared with raw yeast biomass. The highest yield of hydrolyzed proteins is obtained when mild cell disruption methods like cell milling are applied.

6. Spent Yeast Derivates—Potential Applications

The burgeoning global population, coupled with diminishing expanses of available land for agricultural pursuits, presents a formidable challenge. Concurrently, the escalating demand for portable water is accentuated by dwindling water reservoirs in numerous agricultural regions year after year. While plants stand as invaluable nutritional sources of protein, their cultivation necessitates extensive acreage and substantial water resources.

Conversely, animal protein entails a significantly prolonged acquisition period, and its amino acid profile does not markedly differ from that of yeast protein. In many regions, animal protein remains scarcely accessible, with its cost often surpassing the financial means of the populace. Recent years have witnessed a substantial decline in the supply of the most prevalent animal proteins, such as bovine milk protein, attributable to plummeting prices and protracted regional policies enforcing production constraints [48].

A potential solution to satisfying humanity's need for proteinaceous products lies in proteins synthesized by diverse microorganisms, including bacteria, yeast, algae, and fungi [49]. Among the economical protein sources is post-fermentative yeast biomass, a by-product of the brewing industry that boasts up to 49% protein content in dry matter. Proteins derived from brewing yeasts exhibit notable bioavailability, rendering them a sustainable protein alternative and a hypoallergenic choice for vegans and vegetarians. Given their origin as by-products, they also emerge as a superior alternative relative to plant or animal proteins, known for their more resource-intensive production.

Currently, grain meal (mainly soybean) dominates as the primary protein source in feed production. However, a proposition advocates for the substitution of plant-based feeds with microbial biomass in animal diets, including post-fermentative yeast biomass, potentially up to 100%. Owing to their vitamin and mineral content, high protein levels, and bioavailability, brewing yeasts find frequent applications as feed additives. Observable effects from studies encompass heightened immunity, enhanced milk production parameters, and a favorable influence on animal health attributable to an improved intestinal microflora [50,51]. Supplementing feed mixtures with brewing yeast (ranging from 1% to 5%) in broiler diets effectively ameliorated the adverse effects of diets characterized by vitamin and mineral deficiencies, culminating in enhanced bone health in these animals [52]. Consequently, brewing yeasts find extensive utility as food additives, not merely as an economical protein source but also in the form of yeast extract obtained through enzymatic processing. This extract is employed as a flavor enhancer, featuring components such as monosodium glutamate (MSG) and nucleotides like 5′-guanosine monophosphate (5′-GMP) and 5′-inosine monophosphate (5′-IMP). Such extracts, comprising monosodium glutamate and nucleotides, are deployed in the meat industry and the production of diverse food items such as sauces, soups, crackers, and chips [53].

The nutritional value of yeast protein is determined through its amino acid composition, with paramount importance ascribed to the group of eight essential amino acids—isoleucine, leucine, lysine, phenylalanine, methionine, threonine, tryptophan, and valine. These amino acids, unsynthesized by humans, are necessary in the diet [54]. In the amino acid profile of yeast protein, the quantities of individual amino acids, including isoleucine, leucine, lysine, phenylalanine, threonine, and valine, surpass the World Health Organization's (WHO) recommended standards for human diets (Table 4). The optimal protein content in the human diet exceeds 40% [48]. Consequently, 100 g of dry yeast biomass encapsulates up to 49 g of protein, precisely aligning with nearly 100% of the recommended daily intake for adults (50 g) [55]. Yeast protein comprehensively incorporates all essential amino acids in quantities adhering to FAO recommendations. It is imperative to underscore that protein deficiencies may manifest not only in the context of relative or absolute body protein deficiencies but also in scenarios where one or more essential amino acids are deficient [48]. Hence, yeast biomass offers protein of commendable quality, featuring a judicious balance of amino acids, including a complete complement of essential amino acids; this renders it suitable for both human and animal consumption. However, it is noteworthy that the elevated RNA content imposes constraints on the extensive utilization of yeast in human nutrition, as heightened concentrations of uric acid resulting from RNA metabolism may precipitate the onset of gout [32]. The concentration of nucleic acids in traditional sources of protein in the human diet is significantly lower (0.2% for beef muscle and 1% for soybean) [56,57].

7. Yeast Cell Walls and Their Derivates

The protein extracts derived from yeast post-beer production are not the only components that can be repurposed. Examples of yeast cell wall fraction applications are listed in Table 6. As previously highlighted, yeast cell walls encapsulate the majority of polysaccharides within the cellular matrix. Among these polysaccharide fractions, beta-glucan emerges as a significant component, possessing thickening and emulsifying capabilities, along with a notable efficacy in water retention [58]. Furthermore, glucans harbor the potential to serve as substitutes for fats in specific applications. Owing to their inherent structural characteristics, glucans are characterized by a low caloric profile, rendering them an exceedingly coveted supplementation in both pharmaceutical formulations and functional foods. Glucans sourced from by-products of beer production have demonstrated heightened apparent viscosity, increased water retention capacity, and superior emulsion-stabilizing properties when compared to commercially assessed products. Nevertheless, their oil-binding capacity remains unaltered [59].

Beta-glucans sourced from residues of beer brewing have also found utility in the realm of bakery products, exemplified by bread formulations. Bread produced by incorporating flour enriched with yeast beta-glucan at a concentration of 2.02 g of extract/100 g of flour exhibits a more uniform structure, concomitant with a heightened loaf volume [60]. This phenomenon may be ascribed to the stabilizing influence of glucans on gas cells within the dough, similar to the impact observed with other cereal-derived glucans.

An additional prospective application of the glucan fraction lies in its potential to serve as a substitute for fats and emulsifying agents. Extracts derived from yeast cell walls were employed to replace xanthan gum as an emulsifying and stabilizing agent in the production of mayonnaise. Sensory analyses revealed no deleterious effects on the sensory attributes of mayonnaise, even following prolonged storage [58]. This investigation underscored the potential of mannoproteins persisting post-beer brewing as synthetic emulsifying and stabilizing agents in the food industry. Furthermore, an endeavor was undertaken to utilize yeast glucans as a replacement for oil in reduced-fat mayonnaise formulations at levels of 25%, 50%, and 75% of the fat equivalent. Given the pivotal role of fat in numerous culinary preparations, particularly in high-fat products such as mayonnaise, a low-calorie substitute for oil would prove invaluable in the development of reduced-fat food products. In contradistinction to antecedent studies, the sensory characteristics of reduced-fat mayonnaise experienced marginal diminution. However, substitution levels of up to 50% of oil with glucan were deemed acceptable by the sensory panel [61].

Table 6. Examples of applications for products obtained from brewery spent yeast cell walls and extracts.

Product	Application	Advantages	Reference
	Food and fodder additives		
Whole yeast cell	Supplementation of broiler diet (1–5%)	Enhancement of bone health	[52]
Purified beta-glucan	Addition of beta-glucan to flour in bread production	Improved texture of bread	[60]
Purified beta-glucan	Low-fat mayonnaise	Reduced fat content, with acceptable changes in taste	[61]
Purified mannoprotein	Emulsifier in mayonnaise	Replacement of xanthan gum without deterioration of stability and taste	[62]
	Medical and special applications		
Peptide derived from yeast, fraction below 3 kDa	Orally applied solution (rats)	Blood pressure reduction comparable with captopril use	[63]
Selenium-rich peptide derived from yeast, fraction below 1 kDa	Intragastric applied solution (mice)	Protection against UV-induced skin damage	[64]
Yeast extract, yeast extract after additional enzymatic hydrolysis fraction below 3 kDa	Oral administration of water solution (rats)	Protection against stomach ulceration caused by 99.9% ethanol	[65]
Yeast extract	Addition to medium used in in vitro culture of Chinese hamster ovary cells	Growth enhancement	[39]
Yeast extract after additional enzymatic hydrolysis	Addition to serum-free medium used in in vitro culture of skeletal muscle cells	Restoration of cell culture growth	[66]

8. Other Potential Applications

The utilization of yeast protein in food production signifies one of its myriad potential applications. Peptides derived from brewer's yeast manifest a plethora of properties, rendering them compelling subjects for prospective utilization across industries, including pharmaceuticals and cosmetics [67]. Examples of specific uses of yeast-derived compounds are shown in Table 6. A particularly intriguing facet of the activity exhibited by proteins isolated from brewer's yeast pertains to their impact on hypertension—one of the major risks for cardiovascular diseases. Bioactive peptides sourced from these yeasts have been posited as potential alternatives to antihypertensive drugs, such as captopril and enalapril [63]. In experiments on rats, the required dose of yeast-derived peptides (fraction below 3 kDa) to obtain effects similar to captopril was around 300 mg/kg (six times higher than the captopril dose).

Yeast proteins emerge as a bountiful source of selenium, to which the heightened antioxidant activity of yeast extracts is ascribed. In vivo experiments conducted on mice that employed selenium-rich protein fractions have showcased exceptional antioxidant efficacy, culminating in a marked reduction in malonate levels within the liver and serum. Furthermore, an observed protective effect against skin damage induced by UVB radiation has been delineated, underscored by heightened activity in glutathione peroxidase, catalase, and augmented glutathione content [64]. Investigations into protein extracts from yeast have further unveiled salutary influences on processes associated with cellular aging, the mitigation of type 2 diabetes, and prophylaxis against neurodegenerative disorders. However, it is posited that these observed effects primarily emanate from the pronounced antioxidant activity inherent in the scrutinized protein fractions [67].

Amidst the array of properties exhibited by yeast extracts, noteworthy is their capability to sequester iron through the hydrolysates of yeast peptides, thereby augmenting iron bioavailability. This leads to the proposition that peptide extracts from yeast hold promise as integral components in supplements designed to bolster anemia treatment [43]. In vivo studies conducted on rats have substantiated that polypeptide fractions below 3 kDa, derived from brewer's yeast, confer cytoprotective attributes to the gastric mucosa. It is imperative to underscore that proteins and peptides extracted from brewer's yeast manifest diverse biological activities [65]. Each of these activities is characterized by its unique mechanism of action, and while certain peptides or protein extracts may be ascribed to specific activities, an individual peptide may exhibit a spectrum of activities within its structural milieu.

The findings arising from the aforementioned studies posit that selenium-rich peptides may represent a propitious constituent in the formulation of functional foods and cosmeceuticals, owing to their well-documented antioxidant attributes and their efficacy in safeguarding against skin damage.

A very promising application of yeast extract is in the supplementation of cell culture media. Traditionally, these types of media include animal-derived serum, which stimulates the growth of animal cells in vitro. High costs, along with ethical concerns, are the main obstacles to the applications of cell cultures on serum-containing media in food production. Yeast extract proved to be a growth enhancer in cultures of Chinese hamster ovary cells [39]. More importantly, yeast extract could restore the growth of a skeletal muscle cell culture in serum-free medium [66]. The effect was dose-dependent. Unprocessed yeast extract was shown to be toxic at a concentration of 10 mg/mL; however, the toxic effect could be reduced after the initial digestion of the extract with Alcalase enzyme (from Novozyme). These results suggest that the addition of yeast extract to industrial, in vitro cell cultures may significantly increase the demand for yeast extract, which is likely to become a very important application in the future.

9. Conclusions and Perspectives

For small breweries (with a production capacity of around 20 hL per batch), disposing of yeast slurry may be a considerable problem. Introducing yeast slurry to the wastewater

stream substantially increases its organic load. Taking into account that beer manufacturing generates around 750 g of COD/hL of beer, yeast disposal may increase this value by an average of 290 g of COD/hL of beer. Yeast slurry disposal into the wastewater system would also require significant dilution of the wastewater to meet parameter limits accepted by treatment plants, resulting in significant increases in water consumption (around an additional 3 hL to 6 hL of water per 1 hL of beer).

Yeast biomass processing seems to be a promising alternative for yeast slurry. Fractioning processes may produce value-added products such as yeast extract, cell wall fraction, protein concentrate, or beta-glucan, which could be further used in different branches of the food or pharmaceutical industry. Appropriately designed yeast biomass fractionation procedures should result in the conversion of all cell components into commercial products. One of the possible configurations may include the following products:

- Beta-glucan fractions (for food fortification or diet supplementation);
- Mannan fractions (suitable for animal fodder supplementation);
- Food-grade protein concentrates (with a reduced content of nucleotides);
- Low-mass metabolite fractions enriched with nucleotide salts derived from nucleic acid hydrolysis (suitable as a flavor enhancer).

Repurposing all yeast biomass-derived products is crucial for ensuring the profitability of processing facilities. It will also substantially reduce waste generation from the process.

Due to yeast biomass instability, processing should start as soon as possible. However, the equipment required for the complete processing of yeast biomass includes expensive elements such as centrifuges or high-pressure filtration systems, and their purchase in small-scale breweries would be economically unjustified.

A solution to this problem may be represented by partial processing in breweries, including autolysis. Biomass suspension after yeast autolysis is more stable than yeast slurry, and the autolysis process requires much simpler equipment than the full fractioning process. A small brewery (20 hL of production capacity) would require a heated vessel with a volume of only 100 L to run autolysis. Further yeast biomass processing could be performed in specialized facilities that collect biomass from several breweries. The production profile of such facilities would be flexible since procedures utilized in the purification of different fractions of yeast biomass rely on the same equipment.

The processing of yeast biomass derived from small breweries is possible only when favorable market situations emerge. This means that the price of yeast-biomass-derived products must be high enough to ensure profitability. Increasing the demand for vegetarian and functional food products may, along with developing industrial in vitro production, change the market in favor of yeast biomass processing in the near future.

Supplementary Materials: The following supporting information can be downloaded at https://www.mdpi.com/article/10.3390/app14062529/s1. Table S1: Brewery waste strem characterization. Table S2: Exemplary waste water parametews accepted in different treatment plants in Poland. Table S3: Calculation of yeast biomass COD. Table S4: Waste stream generated in beer productin process.

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