

Article

The Comparative Efficacy and Safety of 250 μm versus 350 μm Long Microneedle Patch on Under-Eye Skin

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Abstract: Background: Microneedle patch (MNP) technology is now applied for many purposes, including transdermal drug delivery and percutaneous collagen induction in the cosmetic and dermatology fields. Previous research showed that a MNP effectively improved skin appearance, while treatments using larger or deeper microneedles were not easily tolerated by human subjects. Few studies have compared MNP designs in humans. Study Objective: To compare novel MNP designs with high length and low density versus low length and high density for rejuvenating skin wrinkles under the eyes. Methods: This non-randomized split-face clinical trial was conducted as a double-blind study with 36 Thai female participants. Each participant was treated with two different MNP designs, one on each side of the face. The microneedle lengths were 250 μm with a density of 945 needles/ cm^2 on the left side of the face under the eye and 350 μm with a density of 482 needles/ cm^2 on the right side of the face under the eye. The treatments were applied for 12 weeks, with the assessment outcomes evaluated at the baseline and 2, 4, 6, 8, 10, and 12 weeks. Results: The application of these two novel MNP designs successfully rejuvenated under-eye wrinkles with low pain level scores. Increasing the length of the needle or having a 350 μm long MN can better reduce under-eye wrinkles without statistical significance. During the study period, there was an improvement in skin surface roughness in both groups accompanied by a consistent reduction in under-eye skin wrinkles, without statistically significant differences observed between the groups when using the Antera 3D system. However, the 350 μm long MN also slightly increased the pain compared to the shorter needles (250 μm long MN) with a higher density of needles. There were no side effects associated with the two designs. Conclusions: The two novel MNPs gave favorable results as a safe non-invasive treatment for the rejuvenation of skin wrinkles under the eyes. Increasing the number of needles and increasing the length of the needles were both effective in safely reducing under-eye wrinkles without any adverse effects. Additionally, participants could self-apply them at home and were highly satisfied. However, increasing the length of the needles may result in slightly more pain compared to increasing the number of needles.

Keywords: microneedle patch; under eye wrinkle; rejuvenation; safety; efficacy



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1. Introduction

All people experience skin aging. The recent rise in demand for aesthetic facial procedures has increased public interest in dermatological consultations. Facial aging results in the appearance of abnormal lipid deposits and sagging skin. As we grow older, maintaining youthful-looking and well-cared-for skin becomes more important [1]. Reduced collagen and elastin levels in the skin lead to the formation of wrinkles and fine

lines [2]. Tear trough and periorbital wrinkles cause the eyes to look tired [3]. Nowadays, several treatments for skin rejuvenation are available, with favorable outcomes depending on the effectiveness, less downtime, and minimal pain levels.

Non-invasive microneedle patch (MNP) technology has recently gained popularity as an attractive mechanism for transdermal drug delivery by penetrating the skin's stratum corneum [4]. Microneedling is also used as a collagen induction therapy [1,5]. Microneedles have been extensively researched across various medical applications, spanning from diagnostics to therapeutic delivery in the dermatology field and others. This first publication on microneedle-mediated drug delivery was released in 1998. What is interesting and clinically beneficial is that it can help reduce pain compared to hypodermic needles [6–8]. During the COVID-19 pandemic, cosmetic home-use healthcare devices increased in demand; thus, safety points and benefits need to be evaluated for home use. Microneedling is typically performed by physicians or therapists and is not yet widely used as a cosmetic home treatment therapy. MNP facilitate the penetration of topical medications for individuals who suffer from trypanophobia or pediatric patients [9,10]. The choice of materials for microneedle fabrication determines the various applications and characteristics of the microneedles. Polymeric microneedle arrays penetrate the skin's stratum corneum barrier with minimal invasiveness due to their compatibility with biological systems, ability to degrade naturally, and hygienic properties [11,12]. Microneedles (MNs) are designed with different sizes, with lengths ranging from 25 to 2500 μm , widths from 50 to 250 μm , and tip diameters ranging from 1 to 25 μm . The classification of microneedles is often based on their overall shape and tip geometry such as rectangular, pyramidal, cylindrical, conical, or quadrangular. The needles' shapes and sizes are designed for specific needs, considering factors such as penetration depth, mechanical strength, and fabrication feasibility [13,14]. However, limited studies have compared MNP designs for skin rejuvenation in humans. This research determined the appropriate needle length and number for the production of MNP for home beauty enhancement purposes. Two designs, one with increased needle length and the other with increased needle density, were compared to assess their effectiveness for aesthetic skin rejuvenation in the area under the eye.

2. Materials and Methods

2.1. Subjects

Thirty-six healthy Thai females aged 20–60 years who had mild to moderate wrinkles under their eyes according to the wrinkles scale [15] were recruited. This study was approved by the Human Research Ethics Committee of Thammasat University (COA181/2022) and the Thai Clinical Trial Registry Committee (TCTR20231003005). All the participants provided written informed consent and the risks and benefits of the study were fully explained. The exclusion criteria included those who were pregnant and breastfeeding; those with a history of energy-based device treatment within 1 year, laser therapy within 1 month, chemical peeling within 1 month, mesotherapy injection within 1 month, botulinum toxin or filler injection under the eye area within 1 year, or allergy or hypersensitivity to polymethylmethacrylate (PMMA) and monomer plastic; and those with underlying health conditions that were impacted by the application of cosmetics.

2.2. Microneedle Patch

The MNP were designed and developed by the Nanorobotic System and Nanoneedle Research Team, Responsive Material and Nanosensor Research Group, National Nanotechnology Center, Thailand, and Spike Architectonics Company Limited, Thailand. The MNP was constructed in concordance with the European Committee specifications and was manufactured in the NANOTEC cleanroom under invention patent No. 2001004302, following ISO 13485 medical equipment quality control systems. The needles were made from polymethylmethacrylate (PMMA) and the monomer plastic used in patch production complied with European Medical Equipment Annex IX, Rule 5 standards (FDA Class I, IIA, IIB). These standards ensure contamination-free production, maintaining the sterility

of the microneedle part that penetrates the skin. The MNP portion that penetrates the skin was sterile and made from biocompatible polymers in the methacrylate group, such as PMMA, approved for clinical studies. The biocompatibility of the MN material has been approved in all aspects of cytotoxicity, skin irritation, and skin sensitization. Furthermore, microneedles that would penetrate the skin were sterilized by ethylene oxide. The patch has already been registered and approved by the Food and Drug Administration (FDA) in Thailand. The microneedle (MN) array was prepared on a fabric substrate via the photo-polymerization technique. Each microneedle was designed with a pyramidal shape, as shown in Figure 1a,b. The two microneedle lengths used in this research were 250 μm at a density of 945 needles/ cm^2 and 350 μm at a density of 482 needles/ cm^2 , with the base diameters (w) of the two different MNs being 160 μm and 200 μm , respectively. The distances between the tips of the microneedles (d) were 460 μm for the 250 μm long MNs and 644 μm for the 350 μm long MNs. The shape of the MNP for both sides (left and right) was designed identically to cover the area under the eye, as shown in Figure 1c. The microneedle penetration depth was evaluated at 70–80% of its length after MNP attachment under the eye on each side of the face, as shown in Figure 2a.

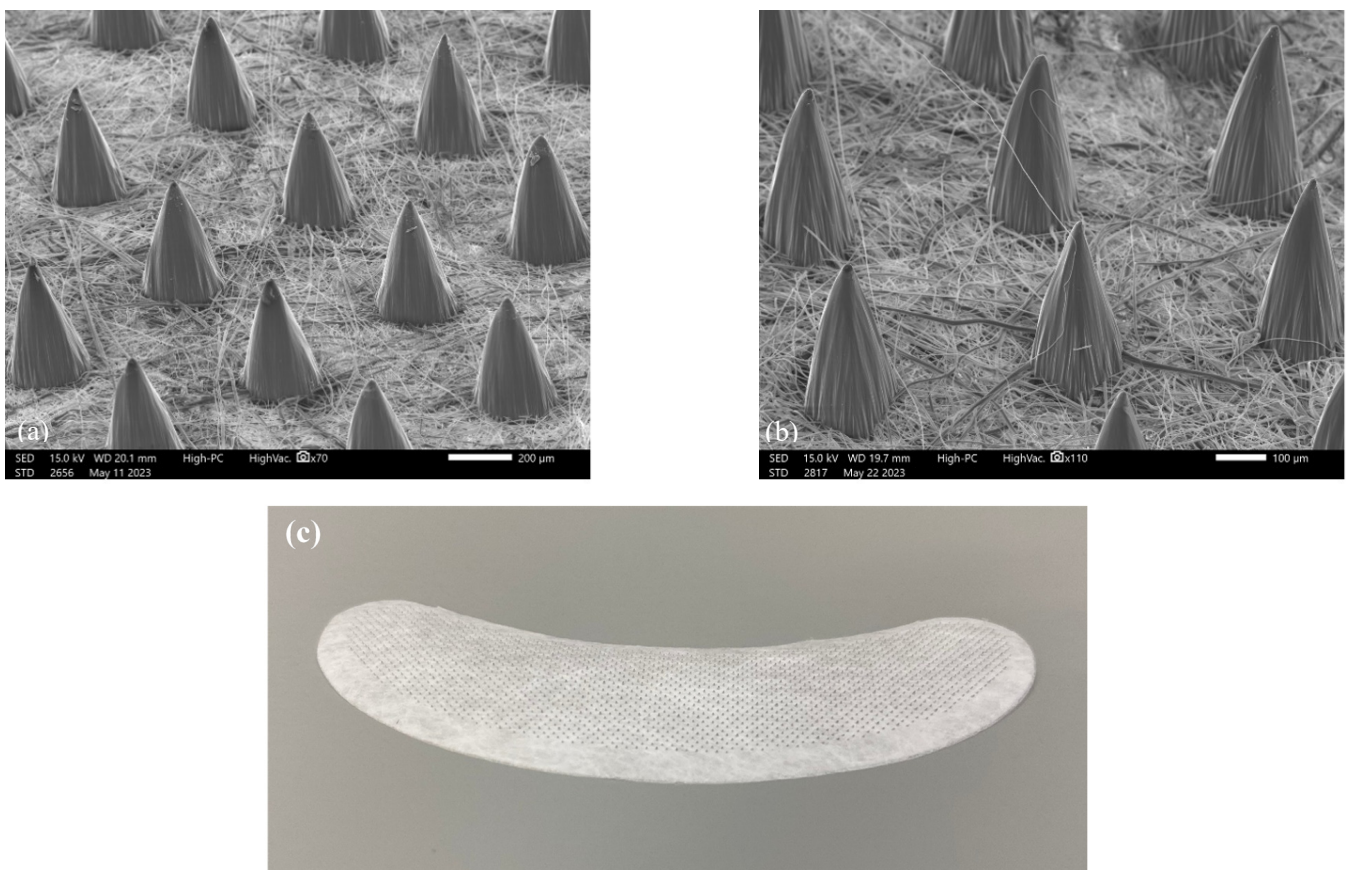


Figure 1. The microneedle arrays of (a) 250 μm length and (b) 350 μm length are shown in a side view of scanning electron microscope images of the pyramidal-shaped microneedle arrays situated on fabric substrates. (c) The shape of the microneedle patch.



Figure 2. (a) Placement of microneedle patch under eyes and the cover area and (b) the use of vibrator massaging for 5 min.

2.3. Study Design

This non-randomized double-blind controlled trial was conducted with a split-face design. Participants were directed to apply one of the MNP designs on one side of their face and the other MNP design on the other side. The microneedles with a length of 250 μm and density of 945 needles/ cm^2 were applied on the left side and those with a length of 350 μm and density of 482 needles/ cm^2 were applied on the right side of the face in the under-eye area for 5 min at night twice a week for 12 weeks. Clinic visits were scheduled at weeks 0 (baseline), 2, 4, 6, 8, 10, and 12 for assessment. All participants were informed of the risks and benefits of the study.

Before the experiment was conducted, a skin irritation test was performed by applying an MNP to the forearm for 5 min. The participants were asked to clean their faces with hibitane solution and dry the area well before applying the MNPs to the skin under their left and right eyes. During the application, the participants were asked to massage the patches for 5 min using a vibrator, as shown in Figure 2b. Moisturizers were supplied and all the participants were asked to apply the MNPs twice a week for 12 weeks, with instructions to continue using only the moisturizer without altering their routine cosmetics throughout the study duration. Any discomfort or adverse events experienced by the participants were promptly reported during the study period.

2.4. MNP Efficacy and Safety Evaluation

Treatment outcomes were assessed at the baseline and 2, 4, 6, 8, 10, and 12 weeks. At every visit, digital photographs of all the participants were taken. Two blinded dermatologists independently evaluated wrinkle grade by comparison with sample photographs of the grading scale of undereye wrinkles (0 = no wrinkles; 1 = a few distinct, fine wrinkles; 1.5 = fine wrinkles with one or two moderate wrinkles; 2 = numerous distinct, fine wrinkles with a deep wrinkle confined to the medial side; 2.5 = numerous distinct, fine wrinkles with a few moderate wrinkles; 3 = a deep wrinkle on both medial and lateral sides and/or indistinct bags under eyes; 3.5 = three or four deep wrinkles and/or distinct bags under eyes; and 4 = moderate wrinkles with numerous deep wrinkles) [15].

Skin roughness and wrinkles were measured using the Antera 3D system. The average roughness index (Ra) was derived from the saved images, with a lower Ra value implying a reduction in wrinkles in the under-eye area. The overall size (mm) of the wrinkles was derived from the saved images and the reduction in overall size was used to assess the improvement in wrinkles in the under-eye area.

The participants were asked to complete a survey using a 5-grade scale to assess the improvement in under-eye wrinkles as excellent (4), very improved (3), improved (2), no change (1), or worse (0).

2.5. Adverse Effects

Adverse effects including skin irritation, redness, and swelling were observed by the dermatologist at each visit.

The pain scores during each treatment session were rated using a visual analog scale (VAS) ranging from 0 to 10.

2.6. Statistical Analysis

SPSS version 19.0 (SPSS, Inc., Chicago, IL, USA) was used for the statistical analyses. All data are presented as mean $\pm$ SD, with statistical significance set at $p < 0.05$. Generalized estimating equations were used for both intragroup and intergroup comparisons with an exchangeable correlation matrix adjusted for the baseline value.

3. Results

3.1. Subjects

Out of the 39 female participants who enrolled, 36 completed the study, with 3 dropping out because of economic conditions. None of the participants showed any contraindications for participation. The average age of the participants was 34.25 ± 8.07 years (mean $\pm$ SD).

3.2. MNP Efficacy Endpoints

The results of the clinical assessment performed at the baseline by the two dermatologists using the grading scale of under-eye wrinkles were 1.61 ± 0.48 in the right eye group and 1.78 ± 0.52 in the left eye group (0 = no wrinkles; 1 = a few distinct, fine wrinkles; 1.5 = fine wrinkles with one or two moderate wrinkles; 2 = numerous distinct, fine wrinkles with a deep wrinkle confined to the medial side; 2.5 = numerous distinct, fine wrinkles with a few moderate wrinkles; 3 = a deep wrinkle on both medial and lateral sides and/or indistinct bag under eyes; 3.5 = three or four deep wrinkles and/or distinct bags under eyes; and 4 = moderate wrinkles with numerous deep wrinkles) [15]. The intraclass correlation coefficients of the two dermatologists were 0.971 and 0.962 for the 350 μ m MN group and the 250 μ m MN group, respectively.

The two MNP designs achieved favorable under-eye wrinkle scale results, with statistically significant differences between the baseline and weeks 8 to 12 ($p < 0.05$), as shown in Table 1 and Figure 3. The MNP group with the longer length demonstrated a statistically significant improvement in under-eye wrinkle reduction starting from week 6 compared to the MNP group that had increased density. Significant efficacy began to show in week 8.

Table 1. Mean grading of under-eye wrinkles.

	Baseline	Week 2	Week 4	Week 6	Week 8	Week 10	Week 12
	Mean $\pm$ SD	Mean $\pm$ SD	Mean $\pm$ SD	Mean $\pm$ SD	Mean $\pm$ SD	Mean $\pm$ SD	Mean $\pm$ SD
Grading reduction							
350 LMN	1.69 ± 0.46	1.67 ± 0.45	1.61 ± 0.45	1.58 ± 0.47	1.49 ± 0.44	1.48 ± 0.44	1.45 ± 0.45
250 LMN	1.78 ± 0.52	1.74 ± 0.51	1.73 ± 0.52	1.72 ± 0.50	1.63 ± 0.51	1.58 ± 0.48	1.54 ± 0.48

Table 1. Cont.

	Baseline	Week 2	Week 4	Week 6	Week 8	Week 10	Week 12
	Mean $\pm$ SD	Mean $\pm$ SD	Mean $\pm$ SD	Mean $\pm$ SD	Mean $\pm$ SD	Mean $\pm$ SD	Mean $\pm$ SD
Percentage change in grading	-						
350 LMN	-	-0.81 ± 9.18	-3.92 ± 11.55	-5.69 ± 12.97	-10.41 ± 16.85	-11.75 ± 14.28	-13.49 ± 14.25
250 LMN	-	-2.37 ± 6.28	-2.83 ± 6.70	-3.08 ± 6.76	-8.32 ± 13.00	-10.29 ± 13.98	-12.46 ± 15.11

Note: The data show the mean grading wrinkles of each design, represented as mean $\pm$ SD.

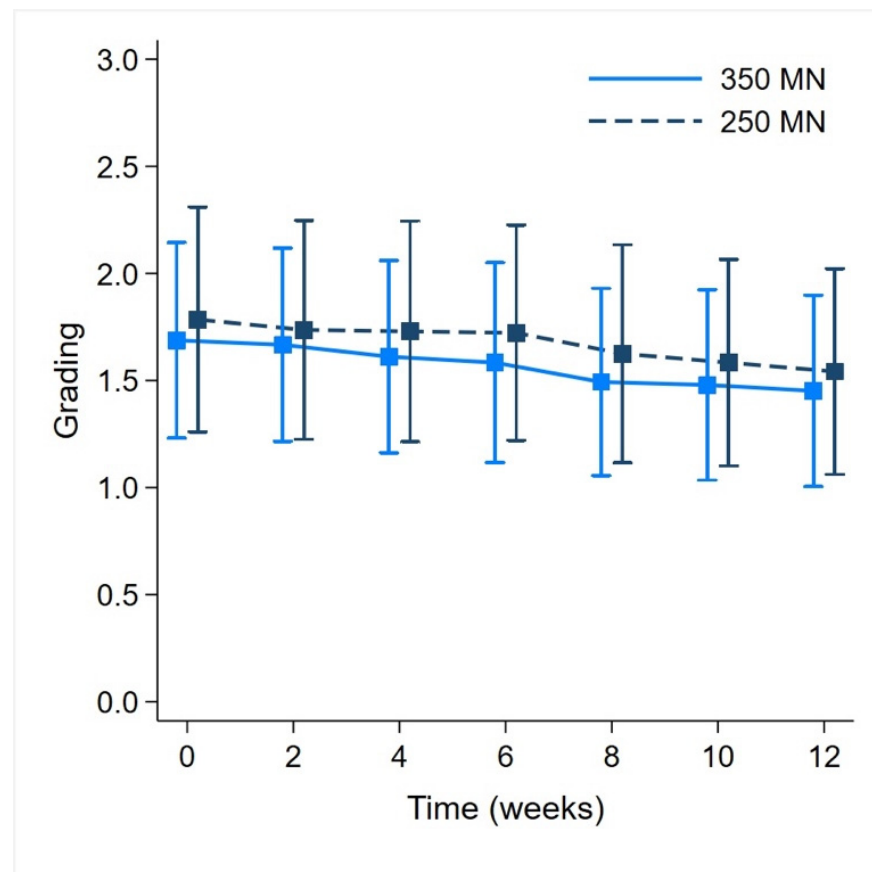


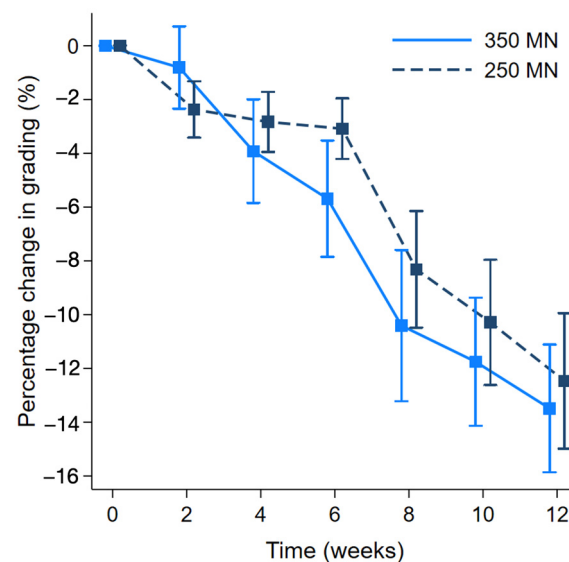
Figure 3. Under-eye wrinkle grade assessed by independent blinded dermatologists. The photographs were evaluated every 2 weeks.

The comparison between the 350 μ m MN group and the 250 μ m MN group regarding the change in severity of wrinkle grade over the study period from weeks 2 to 12 revealed an average difference of -0.012 , with a 95% confidence interval ranging from -0.103 to 0.08 . The statistical analysis indicated no significant difference between the two groups, with a p -value of 0.805 . Additionally, when examining the severity of wrinkle grade at each time point post-experiment, no statistically significant differences were observed, as shown in Table 1. From statistical calculations using percentage change comparison to clearly illustrate changes from baseline values, it is evident that the 350 μ m MN group demonstrated better wrinkle reduction compared to the 250 μ m MN group, with no statistical significance. The average difference in the percentage change in the severity of the wrinkle grade after the experiment from weeks 2 to 12, throughout the study period, between the 350 μ m MN group and the 250 μ m MN group, differed by an average of -0.39 (95% CI: -5.49 , 4.71) with no statistical significance (p -value = 0.881), as shown in Table 2 and Figure 4.

Table 2. Therapeutic efficacy of different designs of microneedle patches for skin rejuvenation in under-eye areas based on the grading scale of under-eye wrinkles.

Time	350 LMN			250 LMN			Difference between Groups (95%CI)		p-Value
	Change from Baseline (95% CI)	p-Value	Change from Baseline (95% CI)	p-Value					
Grading reduction									
Week 2	−0.021	(−0.088, 0.046)	0.543	−0.049	(−0.116, 0.018)	0.155	0.028	(−0.067, 0.123)	0.566
Week 4	−0.076	(−0.143, −0.009)	0.026	−0.056	(−0.123, 0.011)	0.104	−0.021	(−0.116, 0.074)	0.667
Week 6	−0.104	(−0.171, −0.037)	0.002 *	−0.063	(−0.130, 0.005)	0.068	−0.042	(−0.136, 0.053)	0.389
Week 8	−0.194	(−0.261, −0.127)	<0.001 **	−0.160	(−0.227, −0.093)	<0.001 **	−0.035	(−0.130, 0.060)	0.473
Week 10	−0.208	(−0.275, −0.141)	<0.001 **	−0.201	(−0.268, −0.134)	<0.001 **	−0.007	(−0.102, 0.088)	0.886
Week 12	−0.236	(−0.303, −0.169)	<0.001 **	−0.243	(−0.310, −0.176)	<0.001 **	0.007	(−0.088, 0.102)	0.886
Percentage change in grading									
Week 2	−0.81	(−4.59, 2.97)	0.674	−2.37	(−6.15, 1.41)	0.219	1.56	(−3.79, 6.9)	0.568
Week 4	−3.92	(−7.70, −0.14)	0.042	−2.83	(−6.61, 0.95)	0.142	−1.09	(−6.44, 4.25)	0.689
Week 6	−5.69	(−9.47, −1.91)	0.003 *	−3.08	(−6.86, 0.70)	0.110	−2.60	(−7.95, 2.74)	0.340
Week 8	−10.41	(−14.19, −6.63)	<0.001 **	−8.32	(−12.10, −4.54)	<0.001 **	−2.09	(−7.44, 3.25)	0.443
Week 10	−11.75	(−15.53, −7.97)	<0.001 **	−10.29	(−14.07, −6.51)	<0.001 **	−1.46	(−6.81, 3.88)	0.592
Week 12	−13.49	(−17.27, −9.71)	<0.001 **	−12.46	(−16.24, −8.68)	<0.001 **	−1.02	(−6.37, 4.32)	0.708

Note: The data show the percentage change in wrinkle grading for each design. * $p < 0.05$. ** $p < 0.001$. Analyses were conducted with the use of a generalized estimating equation (GEE) with an exchangeable correlation matrix adjusted for baseline value.

**Figure 4.** Percentage change in under-eye wrinkle grade assessed by independent blinded dermatologists.

However, the MNP design with the higher length showed superior improvement compared to that with the higher density without statistical significance. Based on these findings, it can be concluded that there is no significant difference between the groups with 350 μm length and 250 μm length in the effectiveness of the treatments in reducing the severity of wrinkles over the study duration. The clinical photograph is shown in Figure 5.

The roughness refers to the average roughness measurement recorded using an Antera3D camera (Figure 6). The baseline average roughness values were 13.29 ± 3.87 and 15.4 ± 5.49 for the 350 μm MN group and 250 μm MN group, respectively. At weeks 2, 4, 6, 8, 10, and 12, the averages were 13.41 ± 4.73 , 13.8 ± 4.47 , 11.71 ± 3.39 , 11.45 ± 2.72 , 12.49 ± 4.54 , and 11.89 ± 4.42 in the 350 μm MN group and 15.39 ± 6.17 , 14.75 ± 5.72 , 14.04 ± 5.42 , 13.65 ± 5.65 , 13.99 ± 5.91 , and 13.38 ± 5.15 in the 250 μm MN group, respectively. The skin roughness was slightly improved in both groups without statistical significance. The comparison of roughness in the 350 μm MN group and 250 μm MN group showed no statistically significant differences throughout the 12 weeks.



Figure 5. Representative participants that applied microneedle patches from week 0 until week 12. The 45-degree angle photographs were evaluated every 2 weeks.

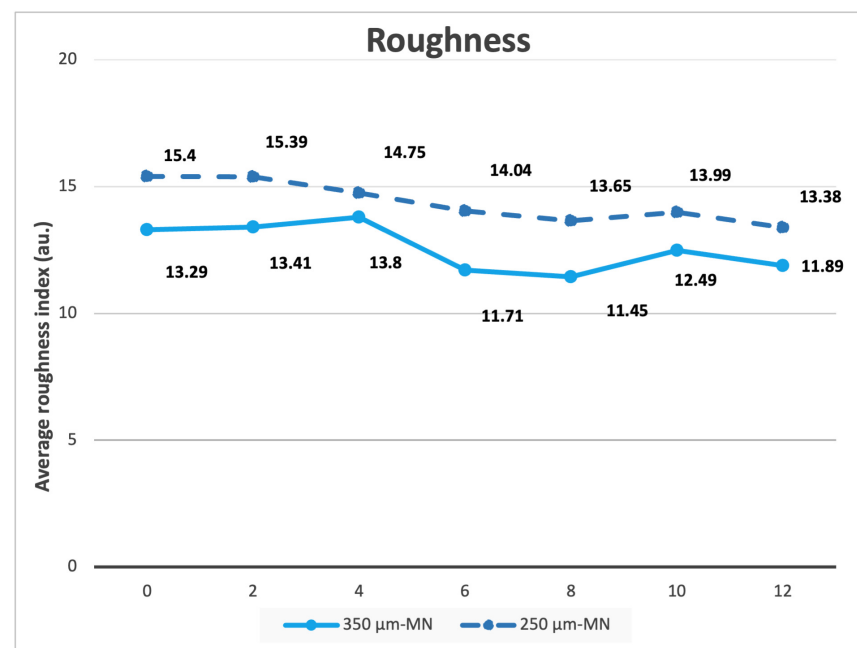


Figure 6. Average roughness indexes every 2 weeks. Both microneedle patch designs reduced skin roughness, with the *p*-value interpreted as the comparison of the average wrinkle cross-section in each group.

The wrinkles were objectively evaluated by comparing the 350 µm MN group and 250 µm MN group using an Antera3D camera. The overall wrinkle size refers to the average cross-section of the wrinkles in millimeters. The baseline average cross-sections of the wrinkles were 9.83 ± 3.62 mm and 11.12 ± 5.05 mm for the 350 µm MN group and 250 µm MN group, respectively. At weeks 2, 4, 6, 8, 10, and 12, the average wrinkle cross-sections were 10 ± 3.99 , 10.68 ± 4.51 , 8.38 ± 2.6 , 8.42 ± 2.42 , 9.39 ± 3.96 , and 8.68 ± 3.58 mm in the 350 µm MN group and 11.03 ± 5.15 , 11.09 ± 6.05 , 10.56 ± 5.12 , 10.19 ± 4.85 , 10.03 ± 5.42 , and 10.01 ± 5.01 mm in the 250 µm MN group, respectively. At the end of the study, there was an improvement in the overall size of the wrinkles on both sides but it was not statistically significant (*p*-values = 0.055 and 0.051). No statistically significant differences between the two groups were recorded. The details are shown in Figure 7.

The skin surface roughness significantly improved over the study period in both groups, with skin wrinkles under the eyes decreasing at all the follow-up weeks. No statistically significant differences were found between the groups. Representative Antera images and digital photographs are shown in Figure 8.

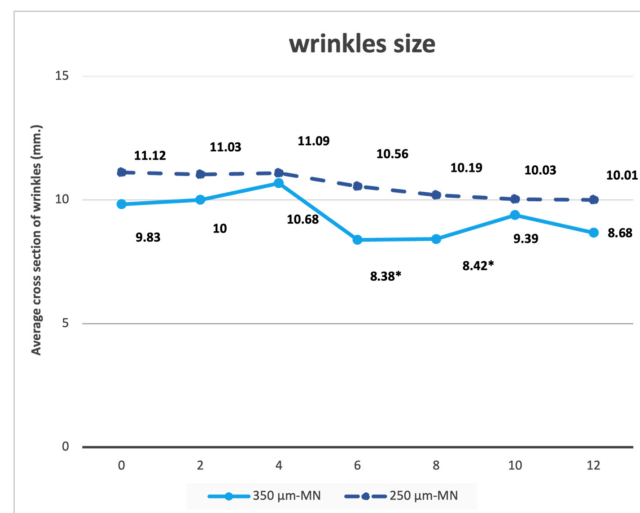


Figure 7. Average cross-section of wrinkles every 2 weeks. Both microneedle patch designs reduced the wrinkle size, with the p -value interpreted as the comparison of the average wrinkle cross-section in each group (* $p < 0.05$ compared within groups).

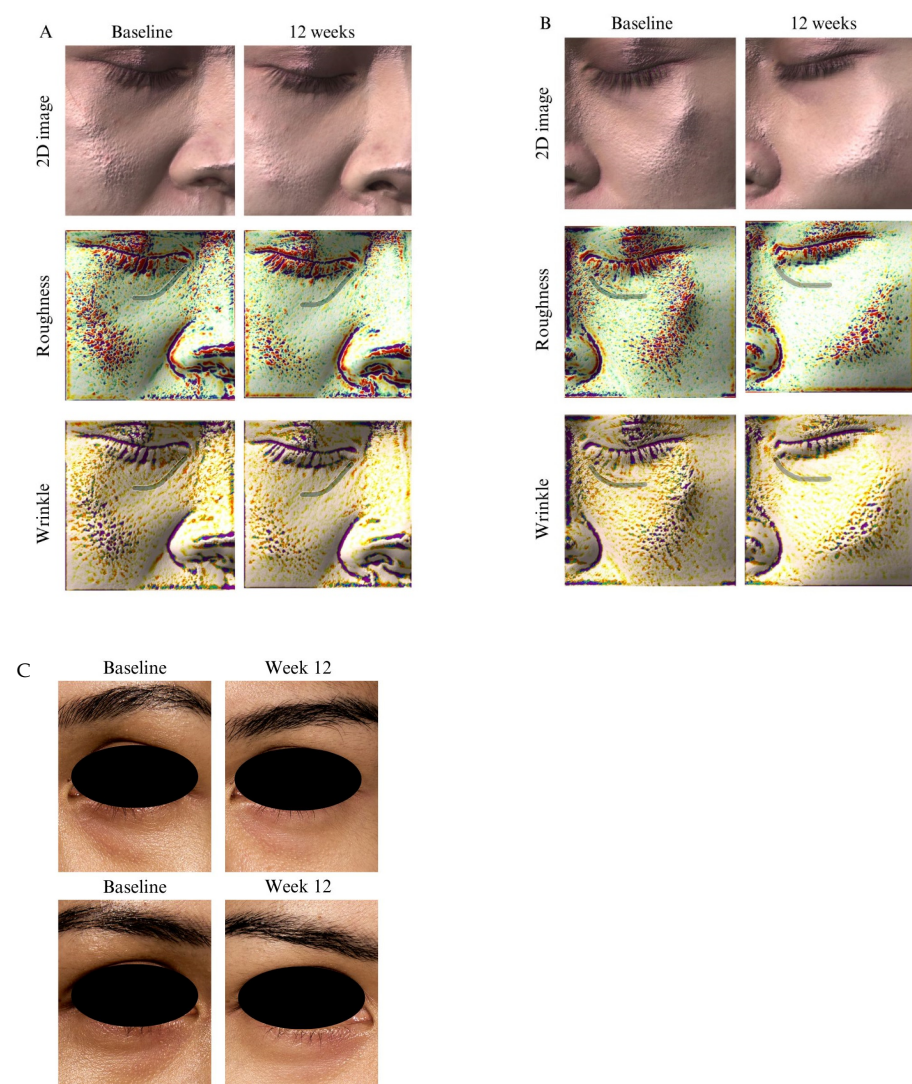


Figure 8. Antera 3D picture showing improvement in under-eye skin at the baseline and at week 12: (A) right side (350 µm MN); (B) left side (250 µm MN); and (C) front view digital photograph.

The baseline scores for both groups were 1. The improvement rates of the participants were assessed on the following scale: 0 = reduced score, 1 = no improvement, 2 = improvement of 25% to 50%, 3 = improvement of 50% to 75%, and 4 = improvement of 75% to 100%. The results suggested statistically significant improvements from weeks 2 to 12 in both treatment groups. At 12 weeks, the scores in the 350 μm MN group showed an improvement of 79% ($3.19 \pm 0.58/4$, p -value < 0.001 *), while the improvement in the 250 μm MN group was 77% ($3.08 \pm 0.6/4$, p -value < 0.001 *). No significant differences were recorded between the groups, as shown in Figure 9. Furthermore, looking at the patient satisfaction score, it is consistent with both subjective and objective outcomes, indicating that longer needles are slightly more effective, without statistical significance.

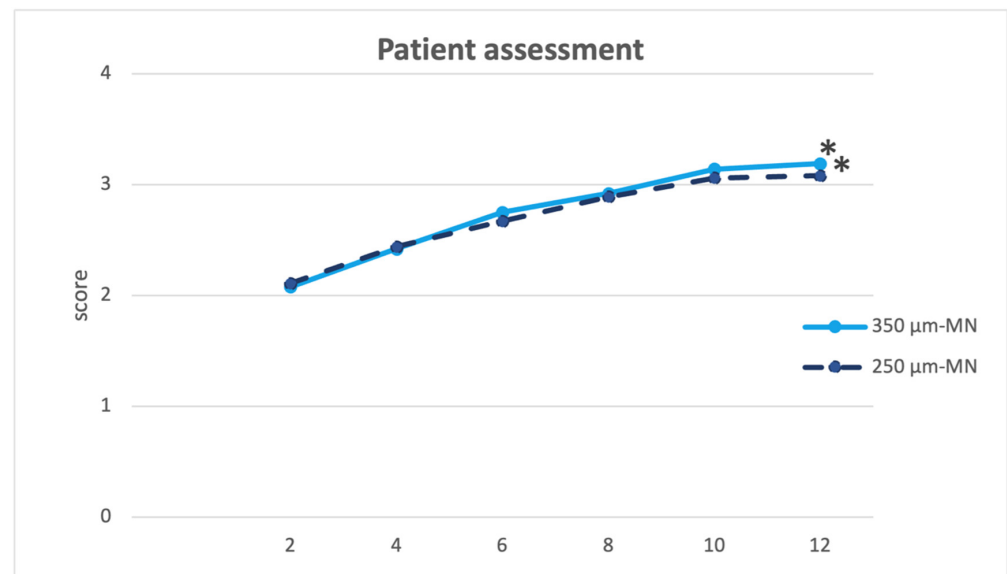


Figure 9. Under-eye wrinkle grades were assessed by participants (* $p < 0.05$ compared within groups).

The participants evaluated pain scores using a visual analog scale, with the results showing that the 350 μm MN design was slightly more painful than the 250 μm MN design. At weeks 2, 4, 6, 8, 10, and 12, the average wrinkles were 2.78 ± 2.23 , 2.81 ± 2.19 , 2.72 ± 2.15 , 2.11 ± 2.23 , 2.11 ± 2.12 , and 1.97 ± 1.99 in the 350 μm MN group and 2.17 ± 1.83 , 2.17 ± 1.75 , 2.28 ± 2.05 , 2.03 ± 2.2 , 1.92 ± 1.99 , and 1.67 ± 1.8 in the 250 μm MN group, respectively. There were some significant differences in pain scores between groups in weeks 2, 6, and 12 (p -value = 0.002 *, 0.009 *, and 0.032 *, respectively).

3.3. Safety Endpoints

Most of the participants experienced mild erythema, with the 350 μm group and the 250 μm group averaging 79.2% and 76.4%, respectively. The results revealed that skin reaction to the patches was similar to a previous study and disappeared completely within a few minutes or hours following application. Skin reactions were not associated with the MNP design. No adverse effects were reported and no skin abnormalities were observed by the dermatologists.

4. Discussion

The longer MN group (350 μm) and the high-density MN group (250 μm) exhibited excellent results on the under-eye wrinkles scale. Both groups recorded significant wrinkle improvements but with no significant differences between them after the study ended. The 350 μm MNP was shown to have a faster effect in reducing wrinkles starting from week 6, possibly because the longer needle length may stimulate collagen production more effectively. Increasing the length and number of needles has an impact on the

effectiveness of reducing under-eye wrinkles, with the trend of increasing needle length possibly being slightly more effective. Our study is consistent with previous studies in which needle length also had an impact on the efficacy of drug delivery. A prior study aimed to develop a transdermal microneedle (MN) patch for delivering naloxone, an FDA-approved opioid inhibitor. Patches of varying dimensions were fabricated to assess the impact of increasing MN length and density on drug release. MN patches demonstrated a reduced lag time and significantly higher drug flux, particularly with longer needles and higher needle density [16]. Another prior study also comparing skin patches found that shorter microneedles (MNs) were more effective in delivering doses due to the “bed of nails effect”. This study highlighted the importance of microneedle spacing (MN–MN spacing) in determining dose delivery efficiency for 1000 μm long MNP. Increasing the spacing between microneedles generally improved dose delivery efficiency by reducing the bed of nails effect [17]. However, our study used microneedles shorter than 1000 μm , potentially reducing the impact of the “bed of nails effect” compared to the length range used in previous research. Additionally, satisfaction scores improved in both groups when evaluated by the participants themselves, attributed to the ability of the MNP to stimulate the natural healing process of the skin and collagen production. At the end of the study, improvements in skin surface roughness and wrinkles were recorded from the baseline, resulting from the mechanism of the wound-healing process from microinjury.

Several previous studies showed similar results to our study. In a recent study in Thailand, 23 female Thai participants were treated with MNP and 1.8% hyaluronic acid (HA) to the right and left nasolabial folds. Patients treated with the MNP alone experienced significant improvements in their scores [18], with no differences reported in the improvement in wrinkles between the two groups. Similarly, treatment with 0.23 mm non-absorbable magnesium MNP under the eyes showed improvement in the grade of skin wrinkles and the index of dermal thickness after 12 weeks [15]. In another study, the application of HA MNP improved crow’s feet wrinkles and increased skin elasticity. After 8 weeks, the HA MNP showed a greater improvement compared to the HA essence [19]. Kim and colleagues noted that the use of MNP improved the appearance of various skin conditions [20]. Previous studies have also used absorbable and soluble MNs. Most of the recent studies have focused on the trans-epidermal drug delivery mechanism, while this study investigated the effects of non-absorbable patches on the appearance of wrinkles under the eyes by the microinjury mechanism.

Aggregation growth factors include vascular endothelial growth factor, transforming growth factor- α (TGF- α), TGF- β (TGF- β), and platelet-derived growth factor. These factors also impact the upregulation of collagen type 1, fibril, and collagen type 3 [1,3,5,21]. Microneedling facilitates the wound-healing process by delivering a current that stimulates the production of proteins at the DNA level [4,22]. Microneedling can also stimulate collagen production by initiating the wound-healing cascade process [23]. In one study, the histopathology of skin treated with microneedles showed a slight thickening of the dermis, characterized by an abundance of collagen bundles, compared to areas not treated with microneedles across all experimental groups [24]. Microneedles stimulate the release of various growth factors from platelets and neutrophils, which in turn triggers the production of collagen and elastin in the papillary dermis. This process, known as collagen induction, promotes the regeneration of skin tissue [25–27].

In this study, the participants evaluated pain scores using a visual analog scale. The results showed that the 350 μm MN design was slightly more painful than the 250 μm MN design. Even though the needle lengths differed by only 100 μm , it is evident that volunteers could discern the difference and felt more pain with the longer needles. This finding concurs with previous reports, indicating that longer MNs cause more discomfort than shorter ones. In a randomized double-blinded placebo-controlled study, 10 healthy individuals with active volar forearms were subjected to varying sizes and thicknesses of single microneedles, with lengths ranging from 480 to 1450 μm . The impacts of microneedle length on pain were the strongest, with a threefold increase in length resulting in a sevenfold

increase in pain. By contrast, the number of microneedles affected the pain score by a factor of over twofold [28]. Pain perception with microneedles is directly related to their length, with longer microneedles having a greater tendency to reach and stimulate nociceptors within the viable epidermis [29]. Our results concur with the literature review and suggest that increasing the needle length tends to increase discomfort more than increasing the number of needles on a MNP does.

The most common side effect of microneedle use was transient erythema, and severe adverse effects were rare. The material used for fabricating microneedles is the primary factor affecting the likelihood of skin irritation development [25,30]. In our study, we used PMMA, which possesses excellent properties that allow it to penetrate through the stratum corneum layer and exhibit relatively low hypersensitivity [31]. Polymers are favored for their cost-effectiveness, biocompatibility, biodegradability, and hygienic properties [12]. In this study, the side effects were minimal, with only transient erythema observed. Moreover, the same type of MNP has also been developed for combined use with drug delivery and LED light therapy. In previous studies, it was found to be effective and free of adverse effects [18,24].

5. Conclusions

Novel microneedle patching is effective for under-eye wrinkle rejuvenation and beneficial for needle-phobic patients as a minimally invasive procedure. The two novel MNP treatment designs tested in this study are safe and effective for home use. Increasing the length of the needles or increasing the number of needles can improve the effectiveness of reducing under-eye wrinkles. However, lengthening the needles may slightly increase the pain experienced compared to increasing the number of needles. The study scope was limited to Thai female participants and utilized only two MNP designs. To broaden the applicability of the findings, future clinical trials should encompass a larger and more diverse population, including a variety of skin types and different designs, or even explore drug delivery mechanisms. Conducting a long-term follow-up study would also be valuable to assess the durability of the observed effects over time.

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