Effects of hyaluronic acid injected using the mesogun injector with stamp-type microneedle on skin hydration

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Abstract

Introduction: The elasticity of the skin and its capacity to hold water decrease with aging because of the loss of hyaluronic acid (HA) in the skin. Therefore, there is an increasing interest in the use of HA fillers in skin rejuvenation beyond its conventional use which is supplementing decreased dermis volume and filling deep wrinkles.

Objective: We investigated the efficacy and safety of a novel device (Dermashine® balance™) that injects HA into the dermis using a stamp-type microneedle for maintenance of hydration and elasticity of the skin.

Methods: A single-center randomized double-blinded parallel-group clinical study was conducted, and 60 participants enrolled in this study. The subjects were randomized to receive HA injections or a placebo 3 times across the face using an automatic intradermal injector. At 4, 8, and 12 weeks after the treatment, skin hydration was measured using a corneometer.

Results: The patients who received HA showed significantly greater skin hydration than those who received the placebo. However, a significant difference was not noted in skin elasticity between the groups. No severe adverse event were reported.

Conclusion: Intradermal supplementation of HA using mesogun multi-needle injector may be a safe and effective treatment for improving skin hydration.

Keywords: hyaluronic acid, hydration, mesogun injector, skin rejuvenation

Introduction

Hyaluronic acid (HA) is present in high concentrations in the skin and plays an important role in aging [1,2]. The elasticity of the skin and its capacity to hold water decrease with aging owing to the loss of hyaluronic acid (HA) in the skin. Accordingly, the loss of HA with aging is associated with increased dehydration and wrinkling of the skin [3]. Therefore, there is an increasing interest in the use of HA fillers in skin rejuvenation beyond its conventional use which is supplementing decreased dermis volume and filling deep wrinkles.

Recently, skin rejuvenation with HA filler injections focuses on not only deep wrinkles, such as nasolabial folds, but also fine wrinkles. The automatic mesogun injector injects a fixed amount of HA gel (20 μl per point) to a certain depth in the skin. Injection of HA in this manner may aid in removing fine wrinkles. Previous studies have shown that a similar micropuncture technique was effective in delivering the HA gel into the skin. A pilot study using the same device identified the efficacy of HA delivered via automatic intradermal injection for skin hydration and elasticity in East Asian men [4]. The results showed that transepidermal water loss (TEWL) and corneometer values significantly improved after treatment. Herein, we conducted a single-center randomized double-blinded parallel-group clinical study to determine the effects of HA on skin hydration and elasticity when injected into the facial skin using an automatic mesogun multi-needle injector.

Materials and methods

Subjects

Sixty Korean female subjects were enrolled in this study. The inclusion criteria were as follows: age, 19–60 years; dryness on both cheeks; and a mean skin hydration level of <49 on both cheeks, measured using a corneometer. The exclusion criteria were as follows: male subjects, any cutaneous disease including atopic dermatitis, psoriasis, and infectious dermatitis on the face; a history of keloid or hypertrophic scar; a history of allergy or hypersensitivity to HA and local anesthetic agents; a history of cosmetic or surgical treatment including administration of fillers and botulinum toxin and fat transplantation on the face within the last 6 months; pregnancy; and lactation. The subjects who had used functional cosmetics for improving skin wrinkles and intense hydration cream on the face in the last 2 weeks were excluded.

The study protocol conformed with the guidelines of the Declaration of Helsinki and Korea Good Clinical Practice. The study was approved by the Institutional Review Board of Chung-Ang University Hospital (IRB-No. C2015271 (1729)). All subjects voluntarily participated in the study, and written informed consent was obtained from all participants after they received a complete explanation regarding the risks and benefits associated with the procedure.

Study design

This study was a 16-week randomized double-blinded placebo-controlled clinical study. The subjects were randomized to receive an injection of HA or placebo (normal saline) on their face using a mesogun injector with stamp-type microneedle. They received a total of three

treatment sessions (baseline; Week 0, Week 2, and Week 4) with 2-week intervals between each treatment session.

Treatment

The automatic intradermal injector, Dermashine Balance® (Huons, Seongnam, Korea), was used in this study. The automatic injector could be used in combination with a multi-needle applicator. The multi-needle applicator is composed of nine 32-gauge microneedles, and it delivers precise small quantities of the injectant to a constant depth up to the intradermal layer. The device injected 0.02 mL of the test sample at each injection point. The test sample was injected at 75 points across the face in each treatment session.

Before treatment, the topical anesthetic cream, EMLA® (Astra Pharmaceuticals, L.P., Wayne, PA, USA), a eutectic mixture of lidocaine and prilocaine, was applied on the entire face of each subject. A total of 1.5 mL of normal saline was injected across the face in the placebo group, whereas 1.5 mL of the HA filler (Elravie Balance®, Humedix, Anyang, Korea) was injected across the face in the HA group. The filler contained HA at a concentration of 20.0 mg/mL and 3.2 mg/mL lidocaine.

Efficacy and safety evaluation

Efficacy was evaluated at 4 week (Week 8), 8 weeks (Week 12), and 12 weeks (Week 16) after the last treatment session (Week 4). Standardized digital photographs were taken using consistent camera settings. Corneometer® CM825 (Courage and Khazaka Electronic Co., Germany) was used to evaluate the degree of skin hydration. The level of skin hydration was

measured on two points on both cheeks. Cutometer® MPA 580 (Courage and Khazaka Electronic Co., Germany) was used to evaluate the degree of skin elasticity. The R2 (gross elasticity), R5 (pure elasticity), and R7 (firmness) values were assessed. The level of skin elasticity was measured on the two points where the lateral canthus meets the alar of the nose bilaterally. We calculated the average value of skin hydration and elasticity at two points in each subject. Two independent dermatologists scored the investigator Global Aesthetic Improvement Scale (GAIS; 1, much improved; 2, improved; 3, no change; 4, worse; 5, much worse) at Week 8, 12, and 16. The subjects scored the level of aesthetic satisfaction through a patient survey. For safety evaluation, all adverse events including local injection site reactions were recorded during the complete follow-up period.

Statistical analysis

Statistical analyses were performed using SAS, Version 9.4. Paired t-tests were used to determine whether the differences within the groups were significant. Two sample t-tests and Wilcoxon rank sum tests were used to determine whether the differences between the groups were significant. The data are presented as mean ± standard deviation. P value of < 0.05 was considered statistically significant.

Results

A total of 60 Korean female subjects were enrolled and randomized into the HA and placebo groups in this clinical study. Three subjects dropped out, and 57 subjects completed the study (HA group, n=28; placebo group, n=29). The mean age of subjects in the HA group was

43.73 ± 6.19 years and that of subjects in the placebo group was 42.73 ± 6.82 years.

The average value of skin hydration measured using Corneometer® CM825 significantly increased in both the HA and placebo groups at Week 8, Week 12, and Week 16. The difference between Week 8 and baseline values was 43.32±16.14 in the HA group and 17.04±13.11 in the placebo group. The difference between Week 12 and baseline values was 45.21±14.24 in the HA group and 17.26±15.19 in the placebo group. The difference between Week 16 and baseline values was 44.68±14.37 in the HA group and 19.98±15.28 in the placebo group. Statistically significantly difference in the improvement of skin hydration were confirmed between the HA and placebo groups at Week 8, Week 12, and Week 16 (Figure 1, Table 1).

Among the values of skin elasticity measured using Cutometer® MPA 580, the R2 value decreased in both the HA and placebo groups at Week 8, Week 12, and Week 16. There was no statistically significant different changes between the HA and placebo groups at Week 16 (Figure 2, Table 1). The other values such as R5 and R7 did not show any meaningful changes.

In the analysis of investigator GAIS scores, significant differences were noted at Week 8, Week 12, and Week 16 between the HA and placebo groups (Table 2). Furthermore, 12 of 28 (42.86%) subjects in the HA group reported that the treatment efficacy lasted for more than 6 weeks, whereas only 5 of 29 (17.24%) subjects in the placebo group reported that the treatment efficacy lasted for more than 6 weeks after treatment.

Figure 3 shows the clinical photographs of subjects from the HA and placebo groups. No severe adverse events were noted during the complete study period. Among 28 subjects in the

HA group, 3 patients experienced local skin reactions including contusion, erythema, pruritus, pain, and irritation. All of these symptoms were transient and spontaneously resolved within 5 days. In one subject, pruritus occurred immediately after the first treatment session which subsided within 3 days. In another subject, pruritus and skin irritation occurred 6 days after the second treatment sessions which spontaneously subsided within 4 days. In the last patient, contusion, erythema and pain occurred the day after the second treatment session which spontaneously subsided within 5 days of treatment.

Discussion

Several studies have reported on skin rejuvenation through HA fillers injected using an automatic mesogun injector. Lim *et al*. reported a high degree of satisfaction and improved melanin index among subjects in whom the nasojugal groove was treated with stabilized HA-based gel of nonanimal origin (NAHSA) that was injected using a specialized injector [5]. Streker *et al.* also reported high subject satisfaction with the treatment outcomes after administration of NASHA gel using an injector device, with both subjects and blinded evaluators identifying aesthetic improvements in the face, dorsum of the hand, and décolletage [6]. Furthermore, facial skin roughness and texture improved after injecting stabilized HA in the superficial dermis [7]. In present study showed improvement in skin hydration after injection of HA in Korean women.

Although only male subjects were included in the preliminary study, we evaluated female subjects in this study. Skin hydration differs between men and women. In Asian women, the average skin hydration value is approximately 39, which is similar to the baseline hydration

value in our study (Figure 1) [8]. The difference in skin hydration value after the procedure was better for women than for men. In the preliminary study conducted on male subjects, the difference between skin hydration values before and after the procedure was 11.63 at 4 weeks after the last treatment session and 7.48 at 12 weeks after the last treatment session [4]. In this study conducted on female subjects, the difference between skin hydration values before and after the procedure was 43.32 at 4 weeks after the last treatment session and 45.21 at 12 weeks after the last treatment session. Hence, we suggested that HA supplementation using a multi-needle mesogun may aid in improving skin hydration in Korean men and women. This result also complemented the limitations of the preliminary study.

We noted that skin elasticity did not change despite the injection of the filler. This may be because Elravie Balance® is a monophasic HA filler, which has a lower elastic modulus and higher viscous modulus than the biphasic or particle-type HA filler. Different volumes of monophasic fillers can be used without a risk of lump formation because these fillers spread out well [9,10].

Both the rheological properties of the injected HA filler and the amount of HA filler injected may affect the treatment outcomes. Mechanical tension on fibroblasts exerted by the injected HA filler may play a critical role in stimulating collagen synthesis [11]. Trace amounts of HA injected across the entire face may not be suitable for significantly inducing collagen synthesis. We intended to detect skin elasticity based on the cutometer values. However, it is possible that minor changes in collagen synthesis after HA injection would not be reflected as significant changes in the cutometer values.

Recent studies have reported that injection of the HA filler into the intradermal layer may

aid in improving skin elasticity and alleviating roughness of the skin surface [7,11,12]. These effects may be due to increased collagen accumulation in areas surrounding the HA material [11]. However, we could not confirm the efficacy of HA injected using a microneedle intradermal injector for improved skin elasticity as compared to that of the placebo. Procollagen gene and protein expression remained elevated for at least 13 weeks, suggesting that the injected HA filler continually activated collagen synthesis pathways in the skin [11]. The duration of the study may not be adequate to accumulate new collagen in the intradermal skin layer, and long-term observation after such procedures is warranted to precisely detect the differences in skin elasticity. Additionally, we assume that the duration of collagen synthesis and amount of newly accumulated collagen after HA filler injection may be affected by rheological properties of the injected HA filler.

The demographic characteristics including age and individual skin status and factors may have affected the results of this study. Although this was designed as a randomized double-blinded parallel-group clinical study to minimize the effects of variables that could influence the prognosis of subjects, a long-term follow-up clinical study with a larger sample population is necessary to establish standard protocols for the use of the novel stamp-type microneedle intradermal injector.

In conclusion, this study showed that HA injection using an automatic intradermal multineedle injector is effective for improving skin hydration and maintaining good skin moisture status until 12 weeks after the treatment. Both the investigator improvement scale scores and subject satisfaction significantly improved, and severe adverse events were not reported throughout the study period. Therefore, administration of HA into the intradermal

layer using a stamp-type multi-microneedle device may be a safe and effective treatment strategy for improving skin hydration.

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Table 1. Skin hydration and elasticity (R2 value) changes

Biophysical parameters	Group	Baseline	Week 8	Week 12	Week 16
Skin hydration	HA group	31.12 ± 9.00	74.44 ± 12.63	76.34 ± 8.65	75.81 ± 9.77
	Placebo group	36.76 ± 5.65	53.79 ± 13.28	54.02 ± 14.27	56.74 ± 14.36
Elasticity (R2 value)	HA group	0.74 ± 0.06	0.72 ± 0.07	0.71 ± 0.08	0.68 ± 0.09
	Placebo group	0.77 ± 0.07	0.72 ± 0.08	0.69 ± 0.10	0.67 ± 0.11

Table 2. Investigator Global Aesthetic Improvement Scale (GAIS) score

	HA group	Placebo group	*p*-value*	*p*-value**
Week 8	1.68 ± 0.77	2.41 ± 0.57	0.0001	0.0004
Week 12	1.61 ± 0.69	2.52 ± 0.57	<0.0001	<0.0001
Week 16	1.57 ± 0.57	2.45 ± 0.57	<0.0001	<0.0001

HA: hyaluronic acid

*: compared between the groups; p-value determined using the two-sample t-test
**: compared between the groups; p-value determined using the Wilcoxon rank sum test

Figure Legends

Figure 1. Skin hydration measured using Corneometer® CM825

*, $p<0.0001$ for changes in values within the groups compared to baseline (Week 0) assessed using the paired t-test; **, $p<0.0001$ for differences in the improvement of values compared between groups assessed using the Wilcoxon rank sum test

Figure 2. Skin elasticity (R2 value) measured using Cutometer® MPA 580

*, $p<0.005$ for changes in values within the groups compared to baseline (Week 0) assessed using the paired t-test; **, $p<0.05$ for differences in the improvement of values compared between groups assessed using the Wilcoxon rank sum test

Figure 3. Serial photographs before treatment and at week 8, 12, and 16 (a) of a subject in the HA group, (b) another subject in the HA group, (c) and a subject in the placebo group.
HA: hyaluronic acid