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ORIGINAL ARTICLE GENITAL SURGERY

360 Genital Fat Transfer

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Abstract

Background This study was designed to evaluate utility of transferring autologous adipose-derived mesenchymal stem cells with high regenerative capacity and adipose tissue derived-stromal vascular fraction, so-called 360 Vaginal Beautification technique, in labia majora augmentation and vaginal tightening operation.

Methods A total of 97 female patients who underwent labia majora augmentation and vaginal tightening operation with 360 Vaginal Beautification technique were included. Post-discharge early (3rd and 7th postoperative day) and late (1st and 3rd postoperative month) surgical complications were assessed , while the Female Genital Self-Image Scale (FGSIS) was applied before surgery and also during postoperative 6–12 months.

Results All complications noted on postoperative 3rd day (ecchymosis of labia majus, ecchymosis of clitoral hood, tenderness in the pubic area and pain at the vaginal entrance points) regressed on postoperative 7th day with no infection, edema, lipoma or granuloma formation in any patient. Total mean FGSIS score was 17.7 ± 1.6 in the preoperative period, and increased significantly to 20.9 ± 1.4 and 22.2 ± 1.8 in the postoperative 6th month ($p < 0.001$) and 12th month ($p = 0.013$), respectively.

Conclusions The use of autologous fat, called 360 vaginal beautification, in the labia majora augmentation and vaginal tightening appears to be a safe technique due to use of autologous tissue transfer and to be associated with high satisfaction rate and an advantage of being more minimally invasive than surgical labia majora augmentation and vaginal tightening.

Level of Evidence IV This journal requires that authors assign a level of evidence to each article. For a full description of these Evidence-Based Medicine Ratings, please refer to Table of Contents or online Instructions to Authors www.springer.com/00266.

Keywords 360 Vaginal beautification technique · Labia majora augmentation · Vaginal tightening operation · Complications · Female genital self-image scale

Abbreviations

HA	Hyaluronic acid
PRP	Platelet rich plasma
RPM	Rounds per minute
FGSIS	The Female Genital Self-Image Scale

Introduction

The popularity of genital rejuvenation and related applications in the field of genital rejuvenescence has increased gradually in recent years. According to plastic surgery statistics report by the American Plastic Surgery Association in 2019, the frequency of labiaplasty operations increased by 9% in 2019 compared to previous year [1].

New studies in the field of genital rejuvenation with a wide spectrum of treatments are intended for introducing more long-lasting, practical, economical and safer

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applications with shorter preparation time in the clinical practice.

The decrease in fatty tissue and collagen content in the genital area secondary to aging, childbirth, or environmental factors leads to sagging, wrinkling and volume reduction of labia majora, and to enlargement of the vaginal opening. This study was designed to evaluate utility of transferring autologous adipose-derived mesenchymal stem cells with high regenerative capacity and adipose tissue derived-stromal vascular fraction, so-called 360-Vaginal Beautification technique, in labia majora augmentation and vaginal tightening operation.

Materials and Methods

A total of 97 female patients who admitted to our clinic with complaints of deformity, sagging and volume loss in the labia majora, vaginal enlargement, and vaginal gas causing noise during sexual intercourse participated in this study conducted between 2018 and 2020. Patient who had labia majora augmentation operation with any filling material including fat transfer or surgery, laser or thread lift therapy for vaginal tightening in the last year were excluded from the study. Written informed consent was obtained from all participants. This study was conducted according to the standards of Good Clinical Practice (ICH-E6) and the principles of the Declaration of Helsinki.

A diagram of procedure is provided in Fig 1. The liposuction part of the 360 vaginal fat technique procedure was performed under local infiltration anesthesia. For infiltration anesthesia, 1 ml Bupivacaine hydrochloride (0.5%), 1 ml Lidocaine hydrochloride (20 mg/ml), and 0.1 Adrenaline (1 mg/ml) were mixed in 100 ml Lactated Ringer's solution and applied to the abdominal area. After waiting for 5 minutes, lipoaspiration was performed from the abdominal region with lateral multi-hole aspiration cannulas with 3 mm diameter and 2 mm hole width, and 100 ml lipoaspirate was obtained from each participant. After ten minutes of decantation, 20-25 ml of infranatant fluid was removed. 55–60 ml oil is thinned with 400-micron Adinizer and at the end of this process; 55–60 ml nanofat is obtained. The remaining 20 ml of fat is reserved for preparation of fat juice (Fig 2).

Doctor B Fat Juice Kit® (Smart Kit Adinizer Bio Solution Co., Ltd. Company, KOREA) was used to micronize, homogenize and filter adipose tissue. 20 ml of lipid reserved for fat juice was taken into a special 20 ml injector. Adinizer® filtering and micronize kit was attached to the injector tip, and a special 20 ml injector was attached to the other exposed end of the kit, allowing Adinizer® to remain between two injectors (Fig 3).

The process was started with 2400 micron Adinizer® and 20 ml fat was filtered, micronized and homogenized by passing through the kit between two injectors 20 times each time. Adinizer®, with its patented design, bladed and micro-porous structure of different sizes and shapes, enables to reach the stromal vascular fraction non-enzymatically without damaging the extracellular matrix in the adipose tissue. The same process sequence was carried out

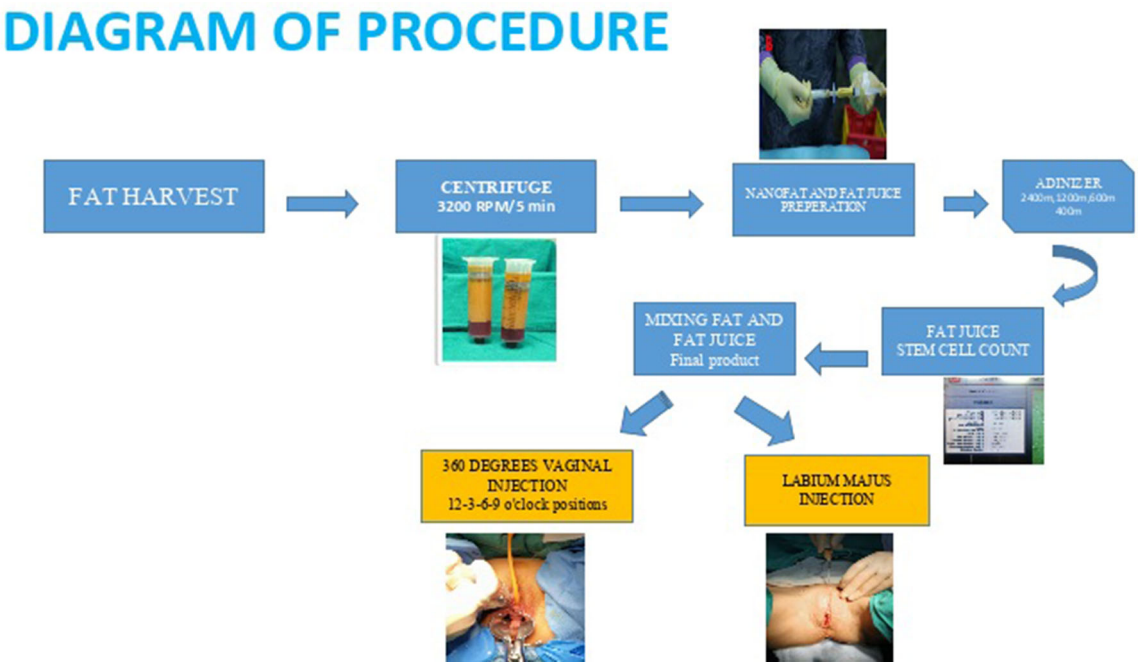

Fig 1. Diagram of the procedure

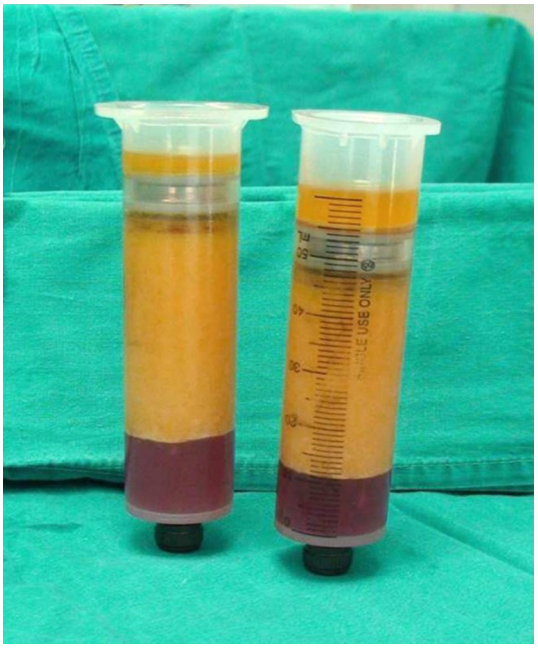

Fig 2. Decantation

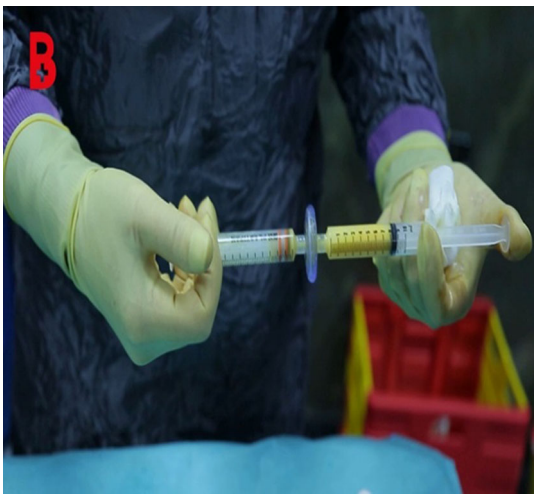

Fig 3. Fat juice preparation

by switching from injector to injector 20 times with 1200 micron, 600 micron and 400 micron Adinizer®.

During the procedure, 2 ml of adipose tissue remained in the kits during the Adinizer® transitions of different sizes and was mechanically lost as a residual adipose tissue. The remaining 18 ml of adipose tissue was divided into two sections as 9 ml each, taken into 10 ml locked injectors and centrifuged for 4 minutes with 1800 RCF (Relative Centrifugal Force).

After centrifugation, 3 ml of reddish colored liquid was obtained in the upper part of each injector and 6 ml residual adipose tissue was obtained in the lower part. This reddish colored liquid was equivalent to stromal vascular fraction obtained enzymatically. This reddish liquid obtained by this process, having high regenerative capacity, that is rich in mesenchymal stem cells of adipose tissue origin, growth factors and interleukins was named as fat juice.

To prepare the nanofat from 60 ml liposuction material, it was first centrifuged at a 3200 RPM for 5 minutes. The nanofat was obtained by passing the infranatant fluid from the injector to the injector 20 times with 2400 micron, 1200 micron, 600 micron and 400 micron Adinizer®, respectively.

Before applying to genital area, FAT JUICE was counted using *LUNA-STEM*^{TM} Automated Fluorescence Cell Counter For Stem Cells & Svf device and total cell count, nuclear cell count and viability rate were calculated (Fig 4).

For the counting, 1.8 microliter FAT JUICE was mixed with 0.2 microliter LOGOS Biosystems Acridine Orange & Propidium Iodide staining and 1 microliter sample was placed into *LUNA-STEM*^{TM} device. While Acridine Orange penetrates into live nuclear cells and stains them green, Propidium iodide penetrates into dead nuclear cells and stains them red. In the counting performed with Dual Fluorescence technique, it was found that the mean total cell count was 2.85 x 10^6 cell/cc, the mean number of nuclear cells was 1.83 x 10^6, and viability was 100% (n:97).

The 6 ml of fat juice was mixed with 60 ml of nanofat. Nanofat enriched with fat juice was used in all procedures.

Vaginal tightening surgery was performed first with the obtained nanofat. Entry points were determined with our special design tool that creates the entry point for the fat cannula, which we call the Punch tool, at the 12-3-6-9 o'clock positions [2]. Afterward, these points were entered with thin fat transfer cannula. The 4 ml of nanofats was applied under the mucosa with the retrograde injection technique, which was entered from each point until 1 cm

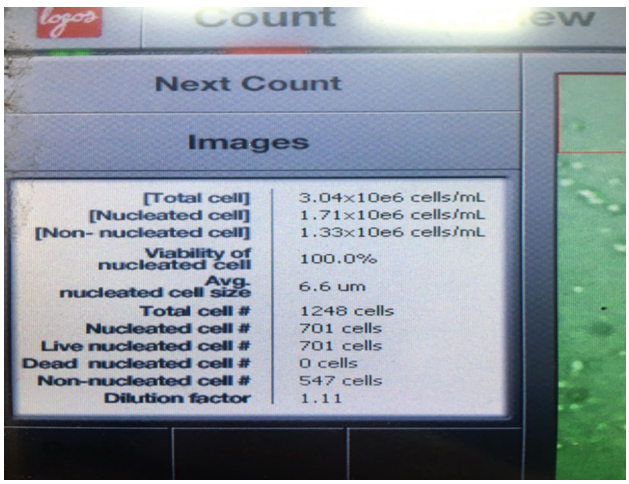

Fig 4. Stem cell count

left to the cervix. A total of 16 ml of nanofat was injected under the entire vaginal mucosa, covering the entire vagina around 360 degrees. Then, it was aimed to spread the nanofat 360 degrees, which was transferred by massaging it into the vagina in a clockwise direction with the right index finger (Fig 5).

Afterward, the fat transfer process for labium majus was started. A single entry point was created with the punch tool at the pubic area. Nanofat was transferred to the upper 2/3 of each labium majus by entering from this entry point with a fat transfer cannula, in such a way that a pudendal cleft was formed between the labium majus. Fat transfer procedure was applied to multiple superficial and deep layers to cover deeper tissues under the skin and under the dartos fascia. Nanofat transfer was performed so that both labium majus were in equal distance and at equal height when the pudendal cleft was considered as the origin. Nanofat was transferred to the pubic area and the sulcus between the clitoral hood and labium majus in order to keep the clitoris and clitoral hood hidden within the pudendal cleft in an upright position when viewed from the front. Nanofat was transferred between 40-60 ml to the labium majus and mons pubis area (Fig 6).

All patients were discharged on the same day after 8-hour monitoring vital signs, and post-discharge follow-up was based on postoperative early (day 3 and day 7) and late (month 1, month 3, month 6, and month 12) period control visits.. Early surgical complications were assessed at 3rd and 7th postoperative day visits, while late surgical complications were assessed at the 1st and 3rd month visits.

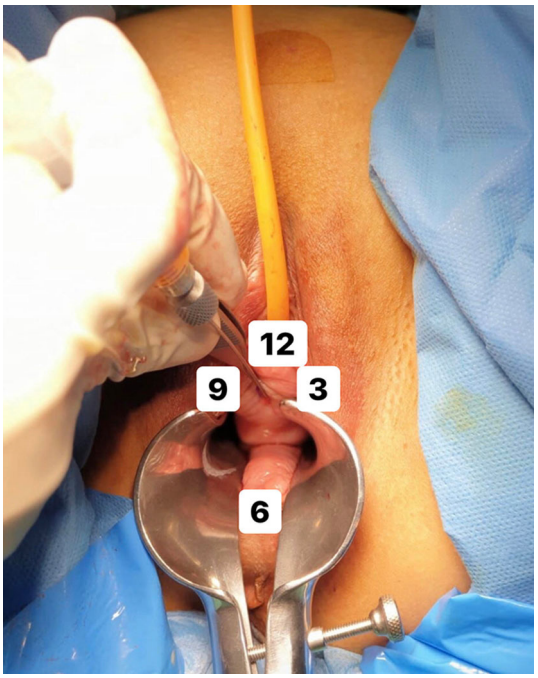

Fig 5. Vaginal injection

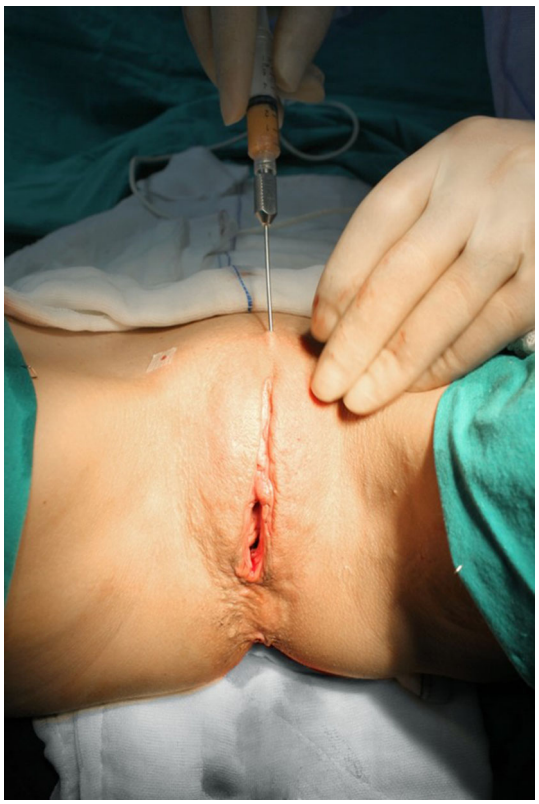

Fig 6. Labium majus injection

The Female Genital Self-Image Scale (FGSIS) [3] was applied before surgery and also during postoperative 6-12 months.

Statistical Analysis

The coding and statistical analysis of the data was performed by using IBM SPSS Statistics for Windows, version 22.0 (IBM Corp., Armonk, NY). Friedman test was used in the analysis of discrete data. Data were expressed as mean ± standard deviation (SD). . $p < 0.05$ was considered statistically significant.

Results

The mean age of the patients was 34.4 ± 4.2 years and the mean body mass index (BMI) was 25.7 ± 4.3 kg/m^2. In the first postoperative visit (Day 3), ecchymosis of labia majus was noted in 3 patients, ecchymosis of clitoral hood in 1 patient, tenderness in the pubic area in 1 patient and pain at the vaginal entrance points was noted in 2 patients. In the second (Day 7) visit, all these complaints regressed. Infection, edema, lipoma or granuloma formation was not observed in any patient (Figs 7, 8, 9).

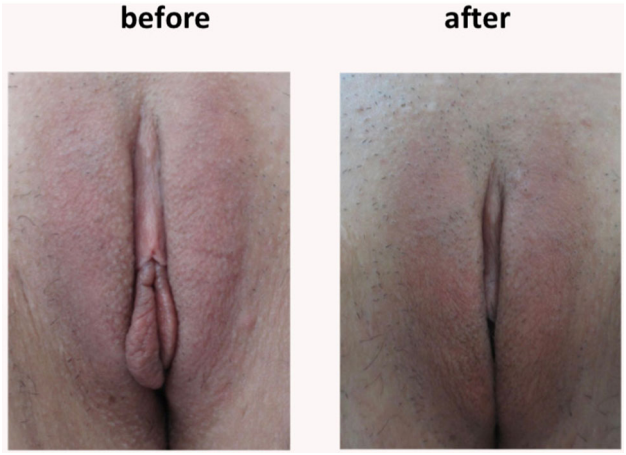

Fig 7. Preoperative and postoperative appearance

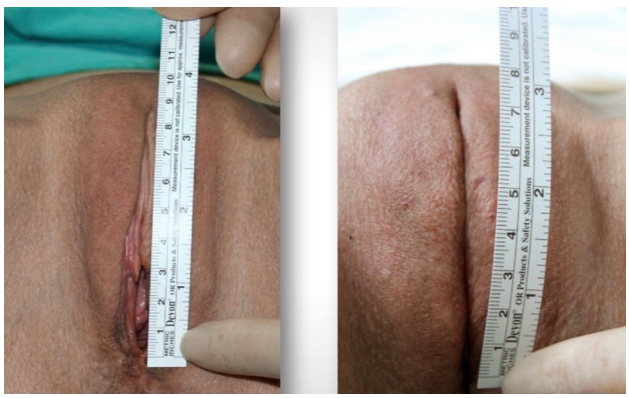

Fig 8. Preoperative and postoperative appearance

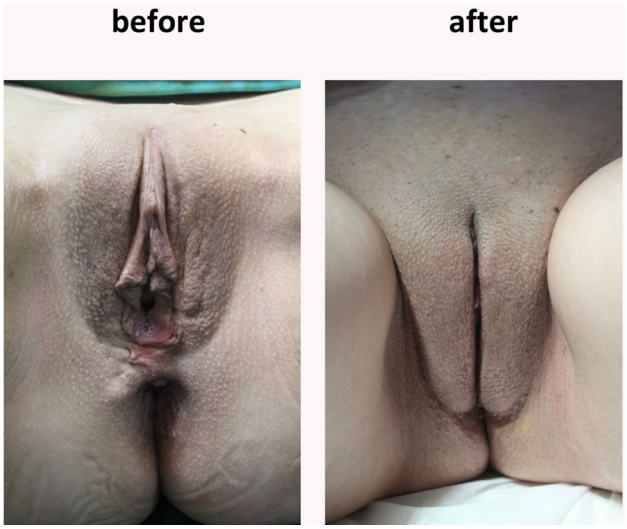

Fig 9. Preoperative and postoperative appearance

A total of 97 patients were prospectively evaluated to compare the FGSIS scores before and 6 and 12 months after the operation. The mean age of the patients was 36.4 ± 5.2 years and the mean BMI was 25.8 ± 4.4 kg/m^2. The FGSIS parameters 1, 2, 3, 6 and 7 increased significantly in the postoperative 6th month as compared to pre-operative values. (respectively, $p < 0.001$, $p = 0.024$, $p < 0.001$, $p = 0.015$, $p = 0.035$). In the postoperative 12th month, no significant change was noted in the parameter 1, 2, 4, 5, 6 and 7 scores, whereas parameter 3 scores continued to increase significantly in the 12th month ($p < 0.001$). Total mean FGSIS score was 17.7 ± 1.6 in the pre-operative period, and increased significantly to 20.9 ± 1.4 and 22.2 ± 1.8 in the postoperative 6th month ($p < 0.001$) and 12th month ($p < 0.013$), respectively. (Table 1).

Discussion

Aging and rapid weight-loss lead to decrease in the amount of dermal collagen and hyaluronic acid as well as in adipose tissue of the labia majora. As a result, volume loss, wrinkles, sagging and skin loosening of the labia majora are observed [4]. There are various approaches for the rejuvenation of the atrophy-related problems in the labia majora. The most popular applications are autologous fat injection and hyaluronic acid filling applications [5].

The first autologous fat graft lipofilling application for correction of volume loss and atrophy in labia majora was described by Felicio in 2007. In this study, 60 ml autologous fat lipofilling was applied to each labium majus and 30 ml to each labium minus [6]. Since then, several labium majus autologous fat graft techniques have been described in the literature. Different techniques were reviewed in the study by Jabbour et al. [7]. The lipofilling applications with different amounts ranging between 18-120 ml were performed using these techniques.

Cihantimur and Herold described a genital beautification surgery technique including autologous fat injections of a total of 18 ml as applied 9 ml to each labium majus [8].

In labia majora autologous fat transfer study by Hersant et al., authors centrifuged the obtained adipose tissue for 1 minute at 1500 Rpm. Afterward, they performed fat injection, more intensely into the middle part of the labium majus, by a single incision over the suprapubic region. Using a multiple-layer injection involving superficial and deep layers of the labium majus, they increased the likelihood of injected fat to remain in the tissue [9]. Similar to the study by Hersant et al., we aimed to increase the duration and ratio of adipose tissue to remain in the tissue by applying autologous fat injection to multiple superficial and deep layers of the labia majora.

In autologous fat transfer, the injected fat tissue was mixed with PRP in a ratio of 4:1 in order to increase the survival and amount of injected fat in the tissue [10]. In our

Table 1. Comparison of preoperative and postoperative (6th and 12th month) Female Genital Self-Image Scale scores

FGSIS parameters	Preoperative Mean ± SD	Postoperative 6th month Mean ± SD	*P**	Postoperative 12th month Mean ± SD	*P***
1	3.1 ± 0.5	3.7±0.6	**<0.001**	3.6±0.6	0.442
2	2.9 ± 0.4	3.5 ± 0.5	**0.024**	3.6 ± 0.7	0.398
3	1.9 ± 0.5	2.7 ± 0.6	**<0.001**	3.9 ± 0.7	**<0.001**
4	2.2 ± 0.7	2.1 ± 0.5	0.384	2.2 ± 0.8	0.152
5	2.5 ± 0.7	2.4 ± 0.6	0.23	2.5 ± 0.4	0.054
6	2.6 ± 0.6	3.3 ± 0.4	**0.015**	3 ± 0.7	0.11
7	2.7 ± 0.5	3.1 ± 0.6	**0.035**	3.2 ± 0.7	0.06
Total score	17.7 ± 1.6	20.9 ± 1.4	**<0.001**	22.2 ± 1.7	**0.013**

Bold values indicate statistical significance ($p < 0.05$)

SD Standard Deviation, *FGSIS* Female Genital Self-Image Scale

*P**: Significance between Preoperative and Postoperatif 6th

*P***: Significance between Postoperatif 6th and Postoperatif 12th

study, in order to increase the survival time and the amount of the transferred fat, we mixed the transferred fat with the final product consisting of adipose-derived mesenchymal stem cell and stromal vascular fraction, which we call fat juice.

Vogt et al. performed autologous fat transfer for the labia majora in cases of unsuccessful surgical reconstructions after oncologic surgery. They reported that this procedure preserves the sensation after the first surgery in the external genital area and also improves the symptoms of vaginal dryness by affecting the mucosal exposure [11]. In our study, we also aimed to provide rejuvenation in the skin of the outer genitalia and the mucosa of the inner genitalia.

In the treatment of labia majora atrophy, hyaluronic acid filling applications are performed in addition to autologous fat transfer. Zerbinati et al. treated 37 patients suffering from labium majus atrophy with 28 mg/ml Polyethylene glycol cross-linked HA. 2 ml HA was described for each labium majus [12].

In other HA filling studies, 19 and 20 mg/ml HA products were used. A total of 2-6 ml of HA was applied to deep tissues either subcutaneously per se or to the deep tissue below Dartos fascia together with subcutaneous injection. Minor complications such as ecchymosis, edema, redness and nodule formation have been reported after these applications [13, 14]. Palpable nodule formation is a complication encountered due to the formation of capsule form around the injected material. In this case, the use of intra-nodular corticosteroid or hyaluronidase is required [14]. In our study, we did not encounter post-surgical nodule, granuloma or lipoma formation since we used fat products that were micronized with Adinizer® and reduced to 400 microns.

Autologous adipose tissue and HA fillings are used in vaginal tightening. Dobbeleir et al. reported that repeated doses are required as HA fillings dissolve over time. The autologous fat tissue has been reported to have positive effects on the vaginal mucosa in postmenopausal women [15]. The authors also indicated the partial melting of fat tissue and granuloma formation. In our study, there was partial melting in the transferred fat tissue, but no granuloma or lipoma formation was observed.

Meadows et al. performed autologous adipose tissue injection into the anterior vaginal wall. The authors reported the effects of autologous adipose tissue transfer to be longer and more permanent compared to other filling materials [16]. In the current study, we preferred autologous adipose tissue transfer as it has more permanent effect compared to other filling materials and does not require repeat of the procedure in a short time.

Brambillla et al. also applied autologous fat transfer to treat vaginal laxity in their study [17].

Kim et al. applied autologous adipose tissue together with PRP during lipofilling and reported successful results in vaginal atrophy and lichen sclerosis with this technique along with the positive effects on the vaginal mucosa [18]. In order to benefit from such a synergistic effect, we have also applied autologous fat application to the external and internal genitalia by using Fat Juice, which is a mixture of the adipose tissue-derived mesenchymal stem cell and the stromal vascular fraction.

A case of pulmonary thromboembolism developed after the use of polyacrylamide hydrogel for vaginal tightening was reported in a study [19]. A 34-year-old woman, who kept private that she had undergone genital aesthetic procedure, admitted to the hospital with complaints of abdominal pain and diarrhea, and died 1.5 hours after development of respiratory distress. Autopsy revealed that

pulmonary embolism was developed due to hydrogel injection.

In a study of Park et al., the autopsy result of a patient with use of HA for G-spot filling was noted, which indicated development of non-thrombotic pulmonary embolism secondary to hyaluronic acid-based filling procedure [20].

In our study, we used autologous adipose tissue obtained by liposuction. In the literature, no fatal complication or embolism has been reported after application of internal or external genital fat transfer with autologous adipose tissue.

There is no control group in our study. This may appear as a limitation of our study. We will need the results of other rejuvenation methods in future studies so that our results can be evaluated and compared.

Conclusion

The use of autologous fat, called 360 vaginal beautification, in the labia majora augmentation and vaginal tightening appears to be a safe technique due to use of autologous tissue transfer and to be associated with high satisfaction rate and an advantage of being more minimally invasive than surgical labia majora augmentation and vaginal tightening. Future randomized controlled studies are needed to compare different techniques and to evaluate their effects on functional outcomes.

Funding None.

Declarations

Conflict of interest The authors declare that they have no conflict of interest.

Ethical Approval This study was conducted according to the standards of Good Clinical Practice (ICH-E6) and the principles of the Declaration of Helsinki.

Informed Consent Informed consent was obtained from every patient participating in this study.

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