Comparative study between enriching fat grafting with autologous adipose-derived stem cells and traditional fat transfer for facial tissue augmentation

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Background
It is still debatable if adding stem and regeneration cells [adipose-derived stem cell (ADSCs)] to conventional fat grafting is the best way to enhance results and achieve more predictable outcomes. This study aimed to compare the effectiveness and safety of standard fat grafting to ADSC-enhanced fat transfer for face tissue augmentation.
Patient and methods
This was a prospective comparative study of 40 patients who were eligible for facial tissue augmentation using split-face technique using fat transfer alone in one half of the face and fat enriched with ADSCs in the other half.
Results
The half of the face injected with fat enriched with ADSCs showed improved longevity of results after comparing with MRI scanning and assessor assessment.
Conclusion
Enriching transferred fat with ADSCs is a safe and reproducible technique with no added risk of complications to the lipofilling technique.

Keywords:
adipose-derived stem cell, fat transfer, MRI

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Introduction

The fat grafting procedure's minimally invasive nature and current broad range of therapeutic applications in the fields of cosmetic and reconstructive surgery are the result of a protracted evolution of concepts that began at the end of the 19th century and have continued to advance to the present.

The systematization of the technique by Coleman [1], which was essential to the success of the results, and secondly to the discovery of the adipose-derived stem cells (ADSCs) made by the Pittsburgh team of plastic surgeons and researchers at the beginning of the new millennium, are related to the recent surge in popularity, which made the procedure part of the current plastic surgeons' toolkit [2,3].

Regarding appropriate techniques for collecting, processing, placement, and recipient site preparation, in particular, there are several unresolved issues in fat grafting. Over time, techniques have seen significant improvement. Nonetheless, they have not been standardized yet [4].

Some researchers have successfully treated challenging wounds that did not heal with conventional treatment using autologous fat. Although the exact cause of this phenomenon is still unknown [5,6], most researchers believe that the ADSC's capacity for regeneration plays a role in fat grafting's ability to treat complicated abnormalities.

It is still debatable if adding stem and regeneration cells (ADSCs) to conventional fat grafting is the best way to enhance results and achieve more predictable outcomes [7].

Despite being in its infancy, stem cell treatment is promising, especially given the ability of the ADSCs in fatty tissue to promote regeneration.

Patients and methods

This study was intended to be a prospective and comparative one. A total of 40 patients were hospitalized to the plastic and reconstructive surgery unit between December 2020 and October 2021 for the study.

All patients were female patients seeking for facial tissue augmentation.

Ethical approval

This research was performed at the Department of General Surgery, Alexandria University Hospitals. Ethical Committee approval and written, informed consent were obtained from all participants.

Inclusion criteria

Participation of both sides of the face was the inclusion criteria.

Exclusion criteria

The following were the exclusion criteria:

(1) Skin infection at the site of injection.
(2) Active smoking.
(3) History of skin diseases.
(4) Blood dyscrasias.
(5) Diabetic patients.
(6) Patients on regular diet.
(7) Bariatric surgery patients.

Procedure

(1) History taking was done, focusing on duration of the condition, previous medications, presence of active skin diseases.
(2) General medical examination was performed.

Liposuction

After the (donor site) had been sterilized with povidone iodine 10%, a tiny incision was made under local or general anesthesia through which an infiltration cannula was inserted.

Infiltration anesthesia was then carried out using the modified Klein's solution. After 15 min, a 60-ml syringe with a blunt-tipped cannula attached was inserted into the incision; the volume that was typically collected was 50 ml. The lipo-aspirate was then emptied into a sterile tube and instantly sent to the stem cells laboratory as the cannula was pushed back and forth while gradually pulling fat into the syringe.

Collection and preparation of the stem cells were done as follows.

Washing the lipo-aspirate

Once the adipose layer's volume had been determined, an equal volume of the washing solution [sterile phosphate-buffered saline (PBS)+antibiotic/ antimycotic, to a final concentration of: 100 IU/ml penicillin, 100 lg/ml streptomycin, and 2.50 lg/ml amphotericin] had been prepared. The lipo-aspirate layers had then been added, and they were left to infra-natant from the bottom was aspirated and decanted. The adipose tissue fraction was washed an average of six times, or until the layer had a golden-yellow tint.

Enzymatic digestion of the lipo-aspirate

Type IA collagenase (0.1%) was dissolved in PBS (equal volume to the adipose tissue layer).

After the final wash, collagenase was applied to the adipose layer and put in a shaking 37°C water bath for about an hour. Then, every 5–10 min, the collagenase/ adipose mixture was gently stirred. As the digestion went on, the adipose tissue layer seemed 'smoother.'

Following digestion, the infra-natant containing the stromal vascular fraction (SVF) was aspirated into sterile 15-ml sterile falcon centrifuge tubes, and an equal volume of complete media (10% fetal bovine serum, DMEM with 4.5 g/l glucose with L-glutamine, 10 000 IU penicillin, 10 000 lg/ml streptomycin 1%) was added. Centrifugation was done for 10 min at 300 g to pelletize the SVF after adding to each tube to inactivate the collagenase. One centrifuge tube containing all of the SVF pellets was filled with 10 ml PBS, through a 100-Um cell strainer, and centrifuged at 300g for 5 min. The SVF pellet was re-suspended in 5 ml of an red blood cell lysis solution and incubated at ambient temperature for 5 min to lyse red blood cells if the pellet was bloody (red) after centrifugation. This procedure produced a clear SVF pellet, which was then centrifuged for an additional 5 min. This step before was optional (skipped if the pellet appeared clear after the first centrifugation). The pellet was then put back into 5 ml of PBS, and a sample of 100 l was obtained for cell counting using the Neubauer hemocytometer and the trypan blue counting technique. The pellet was then supplied in a sterile Eppendorf for injection after being twice rinsed in PBS and redissolved in 1 ml of PBS.

Technique of injection

(1) One half of the face was injected with fat enriched with ADSCs.
(2) Traditional fat transfer for the other half was done.
(3) Injection was done by a small blunt cannula within the subcutaneous level under local anesthesia or general anesthesia.

Evaluation of the work

Objective

MRI volume evaluation for both sides of the face after 6 months of injection was the objective fo the study.

Subjective

The study had the following subjective evaluations:

(1) Patient satisfaction: using binary scale of satisfied or not satisfied.
(2) Assessment by three senior plastic surgeons not involved in the study.

Statistical analysis of the data

With the aid of the IBM SPSS software package, version 20.0, data were input into the computer for analysis (IBM Corp., Armonk, New York, USA). Number and percentage were used to describe qualitative data. The normality of the distribution was confirmed using the Kolmogorov–Smirnov test. Mean and SD were used to describe quantitative data. *F* test (analysis of variance) was used for numerical variables with normally distributed distribution. At the 5% level, significance of the results was determined.

Results

Overall, 90% of the studied group was females, and the age of the included group ranged from 23 up to 61 years.

MRI assessment of the site of injection was determined at the most prominent area of the check bone in each side of the face before, after 1 month, and after 6 months of fat transfer to assess the thickness of the fat bad and to determine the percentage of fat resorption. This is represented in Table 1 and Fig. 1.

Subjective evaluation of the study

This was done using two methods:

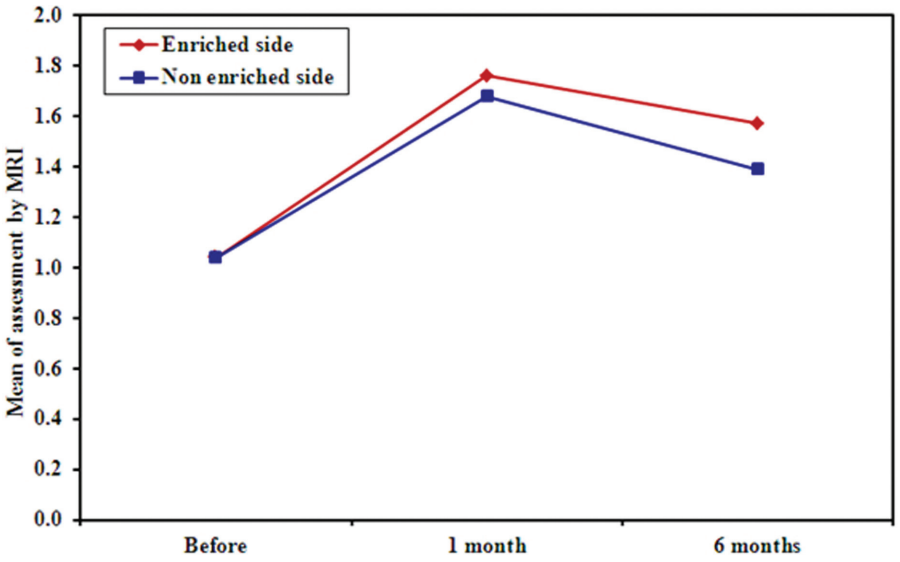
(1) Assessment by three senior plastic surgeons not involved in the study.
(2) Patient satisfaction: using binary scale of satisfied or not satisfied.

Table 1 Comparison between the different periods according to assessment by MRI (*N*=40)

Assessment by MRI	Before	1 month	6 months	*F*	*P*
Enriched side					
Minimum–maximum	0.80–1.30	1.50–2.10	1.30–1.80		
Mean±SD	1.04±0.16	1.76±0.14	1.57±0.13	781.053*	<0.001*
Median (IQR)	1.0 (0.90–1.2)	1.8 (1.7–1.8)	1.6 (1.5–1.65)		
Sig. bet. periods		P_1<0.001*, P_2<0.001*, P_3<0.001*			
Nonenriched side					
Minimum–maximum	0.80–1.30	1.40–1.90	1.10–1.70		
Mean±SD	1.04±0.14	1.68±0.14	1.39±0.17	383.377*	<0.001*
Median (IQR)	1.10 (0.9–1.2)	1.70 (1.6–1.8)	1.40 (1.3–1.5)		
Sig. bet. periods		P_1<0.001*, P_2<0.001*, P_3<0.001*			

*Significant at $P \leq 0.05$.

Figure 1



Comparison between the different periods according to assessment by MRI (*n*=40).

This is represented in

Tables 2 and 3 and Figs 2 and 3.

Some of our cases are represented in Figs 4–7.

Discussion

The filler that comes closest to being optimal is autologous fat. It is affordable, easy to get, and widely accessible. Host rejection or other immunologic reactions are not a possibility. The treatment is simple to carry out, causes little morbidity, and requires little recovery time.

The unexpected resorption pattern of the fat grafting process is a significant drawback.

Different authors have noted varying levels of fat resorption.

It is unknown how long transplanted fat will last. Additional therapies could be necessary to get more positive results.

With classical fat grafting, several strategies have been suggested to provide results that are more reliable over time. These methods seek to increase adipocyte cell survival, which will lead to more accurate clinical outcomes. Most procedures that have been reported have emphasized various processing processes before fat injection. The utilization of full fat rather than suction-assisted lipectomy, processing fat with nutrients, anabolic hormones, or the use of bioenhancers are some of these techniques. Centrifugation to remove the nonliving components (oil, blood, water, and lidocaine) is another (e.g. insulin, insulin growth factor, and type 1 collagen). Despite a great deal of interest in graft survival, a review of the literature finds that there is not enough evidence to support any one strategy. There is no widely accepted graft preparation technique, and there is no agreement on how to get consistent outcomes.

The technique for stabilizing outcomes and generating more predictable results may involve enriching conventional fat grafts with ADSCs. Angiogenic and neovasculogenic abnormalities exist in ADSCs. A total of 40 examples of facial tissue atrophy and ageing that required augmentation were included in the current study; the necessity for tissue augmentation was not for reconstructive purposes but rather for esthetic ones such as face ageing and atrophy.

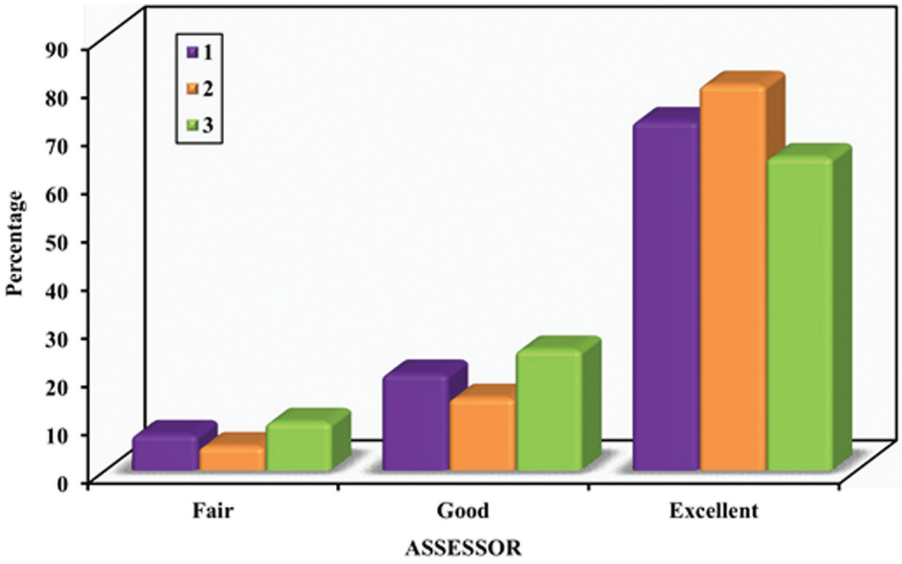
Table 2 Distribution of the different periods according to senior evaluation (*N*=40)

Assessor	1 *n* (%)	2 *n* (%)	3 *n* (%)
Fair	3 (7.5)	2 (5.0)	4 (10.0)
Good	8 (20.0)	6 (15.0)	10 (25.0)
Excellent	29 (72.5)	32 (80.0)	26 (65.0)

Table 3 Distribution of the studied cases according to patient satisfaction

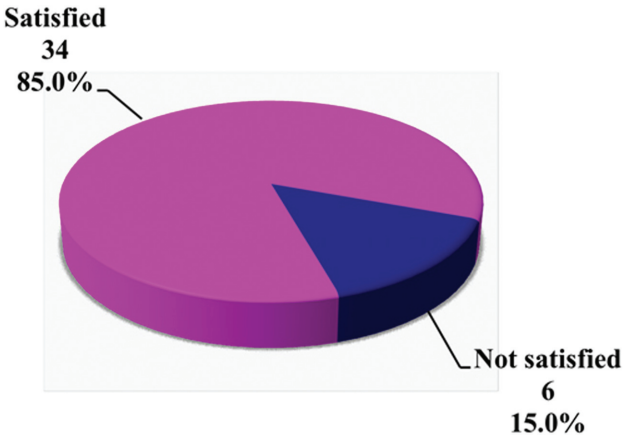
Patient satisfaction	*n* (%)
Not satisfied	6 (15.0)
Satisfied	34 (85.0)

Figure 2



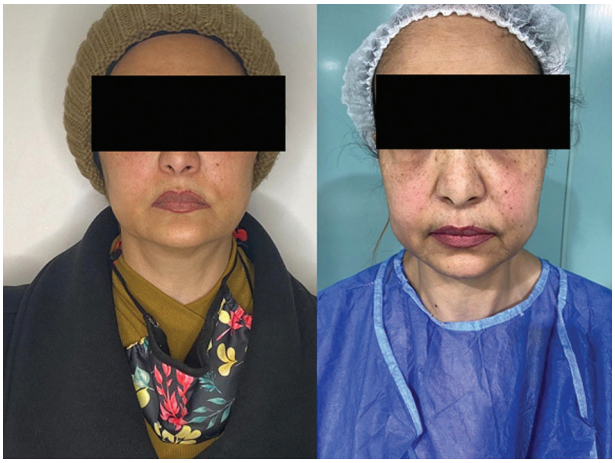
Distribution of the different periods according to senior evaluation (*n*=40).

Figure 3



Distribution of the studied cases according to patient satisfaction.

Figure 4

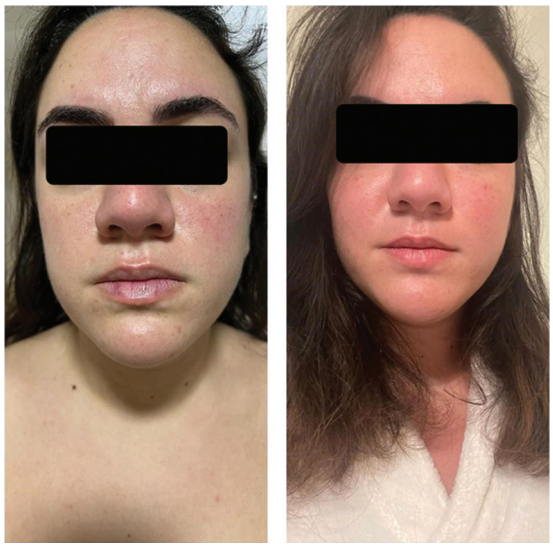


Case 1.

Figure 5

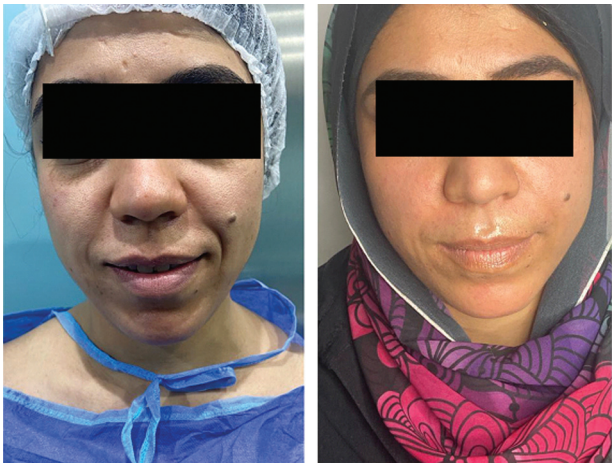
Case 2.

Figure 6



Case 3.

Figure 7



Case 4.

In our study, 90% of cases were female patients and 10% were male patients, with age ranging from 23 to 61 years old and BMI ranging from 22 to 41, with no statistically significant differences in the outcomes.

By contrasting the two sides of the faces of each patient who participated in the trial, our study compares the effects of supplementing the autologous transplanted fat with ADSCS.

Autologous fat was injected into one side of the face and autologous fat enhanced with ADSCs into the other.

The proportion of the mean volume gain out of the mean volume injected is the volume retention rate of face fat grafting.

In our study, the mean volume loss after 6 months compared with 1 month in the enriched side is 10.60 ±4.30, whereas it is 16.93±7.07 in the nonenriched side. According to our findings, which are in line with those of other research studies, ADSC enrichment improves the efficiency of autologous fat grafting and results in grafts that have a greater survival rate and better volume maintenance than unenriched adipose tissue.

The determination of fat graft volume retention is now moving toward more objective imaging techniques, such as 3D surface imaging or computed tomography and MRI scanning.

MRI provides a measuring technique to evaluate the volume and density of soft tissues.

As long as the patients are not claustrophobic and do not have any other contraindications, these scans may be used without difficulty because there is no radiation danger to the patient.

For an objective assessment of fat grafting, some writers have employed MRI. It enables accurate and error-free examination of data when paired with clinical evaluation.

In our study, three MRI scans were taken for each patient: before surgery, after 1 month and after 6 months of the surgery. Both sides of the face were compared in each time to precisely determine the percentage of change in thickness between the enriched and the nonenriched sides during the period of follow-up.

The difference between the readings at 1-month and 6-month follow-ups showed that there was significant preservation of fat in the enriched side than the nonenriched side.

In the current study, another two methods for subjective evaluation were included: patient satisfaction and senior plastic surgeons assessments.

The patient satisfaction rate was 85% and the evaluation results of three assessors ranged from 65% up to 80%, which are very encouraging results.

Conclusion

(1) Enriching transferred fat with ADSCs is a safe and reproducible technique with no added risk of complications to the lipofilling technique.
(2) Follow-up results showed that enriched fat with ADSCs has a better longevity result than fat only.
(3) MRI scan is an accurate and safe technique for assessing and monitoring the results of fat transfer to the face.

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Nil.

Conflicts of interest
There are no conflicts of interest.

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