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**ORIGINAL ARTICLE**

**Facial skin rejuvenation and quality improvement using nanofat graft**

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**ABSTRACT**

**Objective:** The purpose of this study is to evaluate the effect of nanofat injection in face

**Methods:** This intervention study included twenty patients who presented with obvious facial scar, dark hollow under eyes and wrinkles admitted to Zagazig University Hospital in the period from December 2017 to November 2018. Patients were fully informed about the procedure. Written consents were obtained.

**Results:** This study was carried out on patients who had the following inclusion criteria: Patients with facial scars. Dark-colored lower eyelid and superficial rhytides. This thesis included different age groups and both sexes had been included.

**Conclusion:** In this study we illustrated the nanofat grafting effect on treatment of facial scars, wrinkles, and discoloration of the skin (dark hollows under eyes). Our results show improvement of skin quality and Treated skin seems to regain clinical characteristics similar to normal skin prior to scarring.

**Key words;** Clinical Outcome, Nanofat, Injection, Face. Plastic Surgery.

**INTRODUCTION**

The first reports on autologous fat grafting were published in the early twentieth century. It became more widely implemented after the introduction of liposuction by Illouz in the 1980s. Since the standardization of the fat grafting technique by Coleman one decade later, lipofilling has become a very important and valuable tool in plastic surgery. Adipose-derived stem cells were first discovered in 2001. Since then, the adipose-derived stem cell (ADSCs) therapy extend far beyond the field of plastic surgery (1)

With respect to delicate areas of the face such as eyelids and lips, a novel technique of nanofat grafting can be used. The clinical application of the nanofat grafts and sought to determine the cellular content of nanofat grafts. An amount of the microfat was sheared into finer particles using a Leur-to-Leur

connector with+ two 10-cc syringes. The nano particles were then filtered and collected (2)

The nanofat grafts are devoid of adipocytes and the native architecture is disrupted. However, the nanografts retain a rich supply of adipose stem cells. The initial goal of local injections with nanofat using 27-gauge needles was to use it as filler for superficial rhytides. Because of the reduced number of viable adipocytes in the emulsified fat, the filling capacity of nanofat is obviously very limited (3)

Usually, nanofat grafting is combined with other modalities of microfat grafting such as sharp needle intradermal fat grafting to obtain a soft-tissue filling effect, where the nanofat is layered more fanwise in an intradermal level to enhance the skin quality. (4)

Several clinical cases using nanofat grafts resulted in improved skin quality within 2 to 4

months post-operatively, Therefore nanografts do not contain viable adipocytes, the high content of stems cells in nanografts may be clinically useful in skin rejuvenations (5)

## **PATIENTS AND METHODS**

### **Technical design:**

This intervention study included twenty patients who presented with obvious facial scar, dark hollow under eyes and wrinkles admitted to Zagazig University Hospital in the period from December 2017 to November 2018. Patients were fully informed about the procedure. Written consents were obtained.

**Study design:** intervention study.

**Population or subjects:** This study was carried out on patients who had the following inclusion criteria:

Patients with facial scars. Dark-colored lower eyelid superficial rhytides. This thesis included different age groups and both sexes .The patient must be older than 10 years old and had a facial scar more than one year at least .

**While the exclusion criteria were:**  
**Hypopigmented scars.Children below 10 years old**

Pregnant women ,Recent (within the past month) treatment of targeted study sites by other method of skin rejuvenation and improving skin quality

### **Operational design:**

#### **Study design:**

All Patients were subjected to a detailed thorough history taking, clinical examination, investigations, operative procedure and postoperative assessment and follow up. Pre and postoperative assessment was done. Post-operative patient questionnaire was used to assess the degree of patient satisfaction with the procedure.

**- History taking: This included: Personal history, Present history ,Past history & chronic illness,Family history.**

**Clinical examination: This included: Vital signs.Local examination of facial scars, discolouration under eyes and wrinkles**

**Investigation: CBC ,RBS,LIVER AND KIDNEY FUNCTION TEST .**

**Treatment:All patients were operated by the same surgeon.Marking of the donor and recipient sites was done.Pre operative**

**photos were taken**

**Anaesthesia :General anaesthesia or local anaesthesia with intra venous sedation.**

**Preparation of the recipient site: Under complete aseptic conditions, the recipient and the donor site were prepared using Betadine® surgical scrub (containing 7.5% povidoneiodine).**

**Fat graft harvesting from the donor site: Tumescence technique: we added to each 1000 ml of 0.9% saline solution, one ampoule of adrenaline 1 mg/ml and 15 ml of lidocaine hydrochloride 2% without adrenaline. Then infiltration of tumescent fluid into donor site (mostly into abdomen ) using infiltration cannula .**

**Fat processing: The aspirated fat was transmitted to 10 ml syringes connected to a 3 mm liposuction cannula then centrifuged at (3000 rpm) for 3minutes to concentrate the fat particles and separate them from fluids and debris which are then discarded.**

**Nanofat preparation:** After centrifugation, the lipoaspirate is mechanically emulsified by shifting the fat between two 10 cc syringes connected to each other by a female-to-female single use 3 way Luer-Lock connector. After at least 30 passes, the fat changed into an emulsion. At the end of the fragmentation process, the fat became liquid and took on a whitish appearance being "nanofat".

**nanofat Injection: In cases with facial scars a 3 ml Luer-Lock syringe with nokor needle attached to its nozzle was used. The nokor needle was used to release the adherences of the scar in all its planes and create subcutaneous tunnels in a fan shaped manner and multiple levels into which the nanofat graft was deposited when the needle withdrawn.**

The amount of nano fat grafted depends on the extension of scar that needs to be treated. Over correction of about 25 % - 30 % was done. Contour irregularities are minimized using digital pressure immediately after placement.

In cases with dark hollow under eyes and wrinkles superficial injection of nanofat and subdermal microfat graft injection was performed using sharp needles (up to 27-

gauge) mounted on a Luer-Lock syringe. Injection was performed until a yellowish discoloration of the skin showed up.

In cases of facial scars and wrinkles we used a subdermal microfat graft to have a lipofilling effect. In some cases we used to add PRP (platelet rich plasma) to nanofat graft to enhance fat graft survival.

The treated area was covered with dressing for 1 week and the patients were instructed not to apply pressure or friction to the recipient sites to limit the displacement of fat infiltration. External pressure with elastic compressive dressing was applied to the donor sites to minimize postoperative oedema and to prevent hematoma formation.

**Postoperative care: Patients were managed in the Plastic and Reconstructive Surgery Department for several hours and further evaluation of postoperative outcome was performed in the outpatient clinic. All patients received antibiotics (augmentin 1gm) twice daily for 7 days, anti-edematous drugs like ambezim.g tablets twice daily and analgesics like cataflam 50mg twice daily . The first look for the recipient site was on the 7th post operative day. Regarding to the donor site, the first dressing was performed on the 7th postoperative day. Sutures were removed 7-10 days postoperative.**

**Follow-up & assessment: Results were assessed by clinical examination and post-operative photographs one week and three weeks postoperatively then at 3 and 6 months postoperatively. The postoperative outcome was evaluated according to aesthetic appearance. Patient satisfaction as regarding overall improvement of scars and improvement of color of skin under eyes and disappearance of wrinkles. The therapeutic response was determined according improvement criteria.**

The study was approved by the research ethical committee of Faculty of Medicine, Zagazig University. The work has been

**carried out in accordance with The Code of Ethics of the World Medical Association (Declaration of Helsinki) for studies involving humans**

#### **Statistical analysis**

Data collected throughout history, basic clinical examination, laboratory investigations and outcome measures coded, entered and analyzed using Microsoft Excel software. Data were then imported into Statistical Package for the Social Sciences (SPSS version 20.0) (Statistical Package for the Social Sciences) software for analysis. According to the type of data qualitative represent as number and percentage , quantitative continues group represent by mean  $\pm$  SD , the following tests were used to test differences for significance;. difference and association of qualitative variable by Chi square test ( $X^2$ ) . Differences between quantitative independent groups by t test. P value was set at  $<0.05$  for significant results &  $<0.001$  for high significant result.

#### **RESULTS**

In this study Age was distributed as  $32.15 \pm 10.87$  with minimum 12 and maximum 46

Female represent 80% and male 20%. (**table 1**).

As in table (2), patient with facial scar represent 50% of cases, while the wrinkles cases was 35% and the dark colored was 15%. (**table 2**). The distribution of site of wrinkles was as follow :

Naso-labial peri-orbit 40%, Forehead peri-orbit 35% And Naso-labial forehead 25%. (**table 3**).

About 30% of patient needed another session of nanofat grafting specially facial scars.

PRP(platelet rich plasma) was used in about 85% of patient added to nanofat graft before injection .

General anesthesia used in about 85% of all operations. (**table 4**).

Regarding to satisfaction of operation results , most of patients satisfied about results specially patients with scars and wrinkles.

**Table (1): Age and sex distribution among studied group (N=20)**

		Age	
<b>Mean± SD</b>		<b>32.15±10.87</b>	
<b>Median (Range)</b>		<b>33.5 (12-46)</b>	
		<b>N</b>	<b>%</b>
<b>Sex</b>	<b>Male</b>	<b>4</b>	<b>20.0</b>
	<b>Female</b>	<b>16</b>	<b>80.0</b>
	<b>Total</b>	<b>20</b>	<b>100.0</b>

**Table (2): Type distribution among studied group**

		<b>N</b>	<b>%</b>
<b>Type</b>	<b>Dark hollow under eyes</b>	<b>3</b>	<b>15.0</b>
	<b>Scar</b>	<b>10</b>	<b>50.0</b>
	<b>Wrinkle</b>	<b>7</b>	<b>35.0</b>
	<b>Total</b>	<b>20</b>	<b>100.0</b>

**Table (3): Site distribution among studied group**

		<b>N</b>	<b>%</b>
<b>Site</b>	<b>Forehead peri-orbit</b>	<b>7</b>	<b>35.0</b>
	<b>Naso-labial forehead</b>	<b>5</b>	<b>25.0</b>
	<b>Naso-labial peri-orbit</b>	<b>8</b>	<b>40.0</b>
	<b>Total</b>	<b>20</b>	<b>100.0</b>

**Table (4): Procedure characters**

		<b>N</b>	<b>%</b>
<b>Session</b>	<b>One</b>	<b>14</b>	<b>70.0</b>
	<b>Need more</b>	<b>6</b>	<b>30.0</b>
<b>PRP</b>	<b>Not</b>	<b>3</b>	<b>15.0</b>
	<b>Used</b>	<b>17</b>	<b>85.0</b>
<b>Anesthesia</b>	<b>Local</b>	<b>3</b>	<b>15.0</b>
	<b>General</b>	<b>17</b>	<b>85.0</b>
	<b>Total</b>	<b>20</b>	<b>100.0</b>

## DISCUSSION

Facial rejuvenation is the art and science of improving the aesthetics of the face areas to give a more youthful appearance.

Nowadays, scar revision is main procedure to treat the scars, which always leads to unpleasant complications like increasing the scar length and redness and may be recurrent hypertrophic scar. Fat transfer has been shown to have beneficial effects on contracted scars, which were initially treated for volume loss. In 1893, it is the first time to use autologous fat graft for treatment the subcutaneous defect.

Recently, fat harvesting technique and injection have been improved and considered the best way to transplant material for the patient. Fat tissue has been observed to have a regenerative effect to skin and also a rejuvenative effect on skin (6.)

Fat tissue contains a regenerative stem cells (ADSCs) that are responsible for ability of adipose tissue to regenerate a new blood vessels and skin tissue (7).

Through mesenchymal cells and numerous growth factors already contained in the lipoaspirate,

It was shown that fat grafting restores collagen organization. Fat grafting most likely

promotes improvement, which contributes to skin and scar remodeling. In several studies, elasticity and plasticity of the scar tissue was increased, which was also revealed microscopically in posttreatment scar samples (8)

In 2013, Tonnard et al described a new technique of fat grafting called nanofat graft in order to predominantly make use of its regenerative properties. They made a comparison between the new nanofat graft and the classic type of fat graft (macrofat graft and microfat graft), they found that there is no viable adipocytes in nanofat graft in comparable to macrofat and microfat graft. However, nanofat graft contains numerous adipose derived stem cells (ADSCs) (6)

Adipocytes only comprise of 25% of the total cell count in fat tissue. Based on this, since the SVF and the ADSCs are not removed from the solution before injection, the prepared tissue can still be termed as a form of fat. Tissue regeneration and remodeling is considered to be. The main clinical application of nanofat is not filling effect.

Adipose tissue is emulsified between two syringes connected together with a 3 way cannula to create nanofat. A 30 times emulsification of the lipoaspirate simplifies its injection. The intensity and approach to the emulsification procedure might have an impact on the yielded nanofat. Osinga et al, however demonstrated that the mechanical procedure of shuffling lipoaspirated fat does not alter the tissue viability or its microscopic structure, and that there is no impact on the SVF.

It was demonstrated that nanofat contains no viable adipocytes, however, an amount of ADSCs that is comparable to microfat. In an attempt to characterize the potential regenerative benefits, it was experimentally shown that nanofat processing via mechanical lipoaspirate emulsification does not disrupt adipose cells to create an injectable liquid (9)

The viscosity of nanofat was analyzed and SVF was isolated and quantified. Mechanical processing was shown to lead to a shear-stress-induced upregulation of certain progenitor cell phenotypes that are associated

with multipotency and pluripotency. Specifically, adipose-derived-stem cells and endothelial progenitor cell phenotypes were detected in larger amounts in the mechanically processed lipoaspirate, which might be the reason why nanofat grafting seems to have potent regenerative effects with clinically remarkable results.

Intradermal injection of nanofat using a sharp needle. So a needling effect cannot completely be ruled out in nanofat grafting, and may actually contribute to the overall beneficial effect of nanofat. However, we should have to avoid microwounds by needling to prevent an unnecessary outflow of the fluid.

Tonnard et al published remarkable clinical results of skin quality improvement after nanofat treatment in 67 patients that are consistent with the aesthetically satisfactory results that were obtained in our 20 own patient series. Increasing flexibility and softness of skin have been noticed, and discolorations blended into the surrounding healthy skin. The advantage of nanofat for scar treatment is that it introduces regenerative properties and does not create new scars. Furthermore, the risk of abnormal healing is low as no new surgical intervention is applied to the skin. There is no special treatment needed after operation. In most cases, it seems that a single session of nanofat graft is sufficient. Another advantage is that it can be easily combined with traditional fat grafting (microfat grafting). So, skin quality improvement can for example prevent or improve cases where severe retractile scarring is prevalent. We however suggest that as nanofat causes tissue rejuvenation and improves skin quality it can also increase dermal elasticity, and therefore the elasticity of deep wrinkles.

In this study, a skin quality improvement or appearance was noticeable up to 6 months posttreatment on average. However, from our clinical experience, after the evaluation period we often noticed that the effects are long-lasting and may even further improve after 3 months. Further long-term studies are needed in order to clearly document the longevity of this action in greater detail. The

average age of the treated scars in this study ranged from 15 months to 12 years and the majority of them were traumatic. Therefore, at time of nanofat grafting all treated scars were already mature, therefore ruling out the physiological regenerative process as a possible cause for scar improvement as the natural scar formation process is usually terminated after 18 to 24 months. Further research is of course necessary in order to clarify whether scar age and etiology and causes of dark hollow of eye lid and causes of wrinkles have any sort of influence on nanofat results. Additionally, the amount of nanofat needed to be effective still remains unclear and has to be defined in the future.

The evaluation of patient satisfaction conducted by live interviews with a physician may represent a potential limitation of the study.

With that, we believe that this study provides essential information in terms of the results after nanofat graft treatment. As of now there only limited data on the clinical results are available, especially as wrinkles and discolorations under eyes are scarcely investigated in this field.

### CONCLUSION

In this study we illustrated the nanofat grafting effect for the treatment of, wrinkles, scars and dark hollow under eyes. Our results showed that nanofat conveys beneficial effects on skin quality and aesthetic appearance. Treated skin seems to regain clinical characteristics similar to normal skin prior to scarring. Nanofat grafting is an effective alternative procedure to the surgical excision of scars without the risk of potential worsening or expensive short-term dermatologic treatments of wrinkles and discoloration. This safe and feasible application, together with the low tissue

morbidity, makes it a valuable new tool in the repertoire of every plastic surgeon.

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