Skin Morphological and Resident Stem Cells' Changes in Morbid Obese and Massive Weight Loss Female Patients Subjected to Sleeve-Gastrectomy: Histological, Biochemical and Clinical Study

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Original Article

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ABSTRACT

Background and Objectives: Overweight patients meet obvious anatomical changes in the appearance of their skin and consequently morphological changes are expected. Weight loss in a massive manner results in redundancy of the skin following sleeve gastrectomy. The current work targeted to determine impact of morphological and morphoquantitative changes of epidermis, dermal fibers and endogenous stem cells (SCs) on surgical outcome in female patients with class III overweight and those with weight loss in a massive manner subjected to sleeve gastrectomy.

Patients and Methods: Skin biopsies were obtained from excised skin during surgical intervention in thirty female patients, classified into three groups (10 patients for each group). Normal weight group presented with history of no weight loss who have done abdominoplasty. Morbid obesity (MO) group who have done sleeve gastrectomy. Massive weight loss (MWL) group presented with history of massive weight loss after sleeve gastrectomy and were submitted to abdominoplasty. Skin specimens were taken during abdominoplasty after surgical excision of excess abdominal skin in 1st and 3rd groups and during sleeve operation from wound edge in the 2nd group. Sections were subjected to histologic, biochemical, Phenotypic, morphoquantitative studies and quantitative polymerase chain reaction (qPCR).

Results: Wound complications that occurred in MWL patients, were less in MO patients, and some of them required reoperation due to recurrence of redundancy. Associated phenotypic and morphometric changes in epidermal barrier, dermal fibers, degeneration marker and endogenous SCs were less evident in MO patients.

Conclusions: Patients presenting with MWL following sleeve gastrectomy should accept higher complication rates and revisional procedures for recurrent redundancy due to altered skin behaviour. The beneficial therapeutic outcome could be related to the plasticity of skin SCs to transdifferentiate into adult skin cells of epidermal barrier and dermal fibers responsible for skin integrity.

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Key Words: CD105, dermal fibers, massive weight loss, morbid obesity, resident SCs.

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INTRODUCTION

Skin strength and elasticity, particularly the epidermal barrier and dermal content of fibres, may be factors driving both immediate and long-term complications. Even though skin collapse is primarily a mechanical event brought on by adipose tissue resorption and remodelling. However, relatively few research has addressed this issue and contradictory information that is available[1].

It is generally known that overweight patients of high grade undergo obvious anatomical changes and consequently expected morphoquantitative changes in their skin. These include inflammatory process followed by degenerative structural alterations in the epidermis and dermis of the skin including epidermal barrier, dermal fibers and appendages. Class III obesity results in skin redundancy after sleeve gastrectomy[2].

Patients with weight loss in a massive manner experience various medical complications, as seroma formation, skin infection and necrosis, while being examined during postoperative follow up. In addition, weight loss in a

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massive manner precipitates skin redundancy following abdominoplasty due to affected skin integrity and physical appearance[3].

Stem cells proliferation represent a backbone in various stages developing during wound healing. Limited stem cell plasticity is often met with wounds associated with complications[4]. Adult skin resident stem cells were proved to have high ability of transdifferentiation. They were localised among the basal epidermal cells. They were recorded to induce various epidermal and dermal structural elements helping renewal and repair[5].

Since skin redundancy develops with a high percentage following weight loss in a massive manner, single or multiple plastic surgeries may be demanded. As body image has the potential to improve or worsen quality of life and psychological condition, indication of plastic surgery is a professional problem. For the patient as well as the surgeon, unpleasant cosmetic outcome, especially persistent redundant skin, can be unsatisfactory[6].

Loss of extreme amount of weight leads to skin redundancy, which affects strength, integrity and consequently tightness of the skin . Patients are searching for plastic surgery to resume the cosmetic normal appearance of the skin before gaining excess weight[7].

The current work targeted to determine impact of morphological and morphoquantitative changes of epidermis, dermal fibers and endogenous stem cells (SCs) on surgical outcome in female patients with class III overweight and those with weight loss in a massive manner subjected to sleeve gastrectomy.

PATIENTS AND METHODS

Skin biopsies were obtained from excised abdominal skin during abdominoplasty and from wound during sleeve gastrectomy in thirty female patients, classified into three groups (10 patients for each group). The operations were done in Kasr El Aini Hospital in the period from 2017 to 2019.

The first group (Normal weight group)[8]: included 10 patients with no history of weight loss who were submitted to abdominoplasty. These patients were presenting with redundant abdominal skin (Figure 1a).

The second group [Morbid obesity (MO) group]: included 10 patients presenting with morbid obesity[2], (also called Class III obesity, a complicated chronic illness where the patients have a high index of body mass of forty or more)[8], which were subjected to sleeve gastrectomy (Figure 1b) .

The third group [Massive weight loss (MWL) group]: included 10 patients with history of massive weight loss[3], (50% or greater loss of the excess weight)[8], after sleeve gastrectomy submitted to abdominoplasty 6 months later. They were presenting with redundant abdominal skin (Figure 1c).

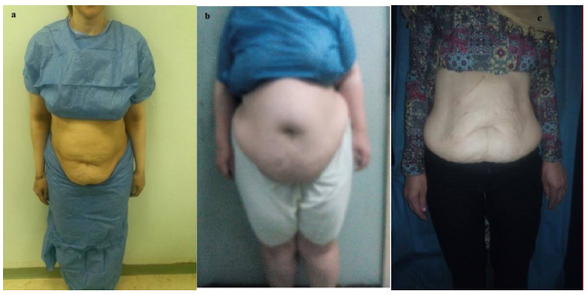


Fig. 1: a. Patient belonging to normal weight group. b. Patient belonging to morbid obese group c. Patient belonging to massive weight loss group

INCLUSION CRITERIA

30 to 40 years of age, females underwent sleeve gastrectomy or abdominoplasty.

Body mass index (BMI) of patients in the three groups is shown in (Table 1).

Table 1: BMI (Kg/m^2) in female patients of the three groups

BMI	Group I (Normal Weight Group)	Group II (Morbid Obese Group)	Group III (Massive Weight Loss Group)
1	24.9	40	26
2	24	42.5	27
3	23.5	40.5	27.5
4	22.5	41.6	28
5	24.5	40	30
6	24.9	43.9	29.5
7	23	31.5	28.5
8	24	42.4	27.5
9	24.9	41.9	30
10	24	42.9	29

Exclusion criteria

- History of previous operation
- Cushing syndrome or inflammatory bowel disease
- Corticosteroid users
- Clinical assessment of defective absorption or malnutrition
- Smoking
- Hospital admission in the previous 8 weeks
- No patient's consent.

Preoperative assessment

1. Preoperative laboratory investigations included complete blood count, renal and hepatic functions, fasting and postprandial blood sugar, coagulation profile, and chest X ray.
2. Preoperative marking while the patient in the standing position

3. Informed consents were obtained from the patients and have been signed by all study participants.

Operative technique

1. Induction with inhalation general anesthesia via endotracheal tube.
2. Patient in supine position.
3. After sterilization, surgery will be done
4. Skin biopsies from wound during sleeve gastrectomy and abdominal skin excised during abdominoplasty.

Post-operative assessment and follow up

1. Follow up for patients as regards skin redundancy and wound complications within one month and after 6 months.
2. Histological assessment for skin biopsies.
3. Image Analyzer System for quantitative assessment of thickness and area of epidermal changes, in addition to area% of dermal fibers, degeneration and endogenous SCs.
4. Quantitative polymerase chain reaction (qPCR) for tumor necrosis factor (TNF) α.
5. Biochemical study for oxidative stress.
6. Statistical Analysis for the results.

In control patients and in patients with history of losing extensive weight skin specimens were obtained from the excised skin of abdomen 4cm x 4cm, during abdominoplasty after the surgical excision of excess abdominal skin. In morbid obese group, skin specimens were 2cm x 2cm obtained from the edge of the wound during laparoscopic sleeve gastrectomy. Fixation was performed in 10% formol saline for 2 days. 5µm thick paraffin sections were cut and the underlying studies were done:

Morphological Study

1. Hematoxylin and eosin[9].
2. Masson's trichrome stain to demonstrate collagen fibers[10].
3. Orcein stain to demonstrate elastic fibers[10].

Immunohistochemical Study

Caspase 3 Ab[11]: is rabbit polyclonal IgG Ab (Abcam Organization, Cambridge, UK, catalogue number: ab4051) to detect apoptosis. It was supplied as 500 µl of antibody (1 mg/ml), used concentration is 0.002 mg/ml. The reaction is cytoplasmic and nuclear.

CD105 Ab[12]. (Catalogue number MA5-11854, SN6h). It was supplied as 0.1 ml prediluted primary Ab goat polyclonal (Thermo Fisher Scientific, USA) to detect resident stem cells. Reaction is membranous. The +ve control was human tonsil for caspase3 and CD105.

Morphoquantitative Study

Leica Qwin 500 LTD (Cambridge UK) image analysier, measurement of epidermal thickness, area (µ2) of apoptotic nuclei, cytoplasmic vacuolations and area of separation between connective tissue (CT) fibers were performed in 10 non overlapping fields using interactive menu. The area% of collagen fibers , that of elastic fibers, that of caspase3 +ve immunoexpression (IE) and that of CD105 +ve IE were performed by binary menu. All measurements done in fields were selected in the prepared slides from the specimens belonging to the three groups of patients.

Quantitative polymerase chain reaction (qPCR) for tumor necrosis factor (TNF)α

Reverse Transcription was carried out using paraffin sections[13]. The TNF α primer was, 5′-oligonucleotide primer -5′ ACCACGCTCTTCTGTCTACTG -3′ and 3′-oligonucleotide primer 5′-CTTGGTGGTTTGCTACGAC -3′. The complementary DNA strand was created from RNA. The TNF DNA was set up on PCR program. β-actin was used as reference for values.

Biochemical study

1 ml normal saline was used for homogenate formation of half of skin specimens in all groups by using (Ortoalresa homogenizer, Spain). Centrifugation for 15 minutes was followed. The supernatant was kept in epindorff tubes at - 20°C. DNA fragmentation[14] was estimated. In addition, reduced glutathione (GSH), catalase and malondialdehyde (MDA)[15] were assessed by using colorimetric method kits (Biodiagnostic, Egypt)

Statistical Analysis[16]

Means and standard deviations were analysed using ANOVA. Any significant difference was determined by *P-values* <0.05. Calculations were made on Statistical Package for Social Sciences software version 16 (Chicago, USA).

RESULTS

Postoperative results

In the first group, the normal weight 10 patients underwent abdominoplasty, 9 patients passed with no complications postoperatively. The healing of the wound was optimum with no delayed wound healing, there was no seroma collection and no skin necrosis, wound infection was minimal in only one case and was treated. With follow up after 6 months, we found that there was no recurrence of redundancy in the skin of this group of patients, so there was no need for reoperation.

In the second group, the 10 MO patients underwent sleeve gastrectomy, surgery resulted in greater improvement in weight loss outcomes. Postoperative, 7 patients showed infection to their wounds, recurrent infection, back and abdominal pain, and were treated. With follow up after 6 months, we found that patients showed massive weight

loss with abdominal skin redundancy. Patients also showed striae and stretch marks in their skin.

In the third group, the 10 MWL patients underwent abdominoplasty. Post operative complications were frequent and had a higher reoperation rate, and longer hospital stay. Patients showed skin laxity, pain and wound seromas, infection, cellulitis, hematoma, skin and umbilical necrosis and were treated. With follow up after 6 months, we found that recurrence of skin redundancy occurred in 8 patients and required reoperation.

Histological Results

The postoperative results were associated with phenotypic changes:

(H&E) Stained Sections

Group I (Normal Weight Group): Examined sections of skin of abdomen showed epidermal keratinocytes, papillary and reticular dermis with normal density of the fibers (Figure 2a). Close observation showed keratinocytes arranged in the form of single basal, thick multilayered spiny, thin granular and horny layers formed of keratin filaments (Figure 2b). Keratinocytes of basal and spiny layers exhibited pale nuclei, while granular layers showed dark basophilic cytoplasm. Dark nuclei and vacuolated cytoplasm were noticed in accidental cells in spiny layers by closer observation (Figure 2c). Apparently normal sweat glands and sweat ducts were surrounded by apparently normal dense fibers (Figure 2d). Close observation recruited basal clear cells and apical dark cells lining the sweat glands (Figure 2e).

Group II (MO Group): In this group, some areas of thickened epidermis with hypertrophied spiny layer and atypical horny layer were observed. Congestion blood vessels was found in some vessels in the papillary dermis (Figure 3a). Close observation demonstrated multiple dark nuclei and vacuolated cytoplasm among the epidermal cells of these areas (Figure 3b). Cellular infiltration was observed around vessels showing congestion in the superficial dermis and areas separating fibers were evident in the reticular dermis (Figure 3c). Fat cells were seen among sweat glands (Figure 3d). Close observation revealed dark nuclei of some lining cells of sweat glands and vacuolated apical cytoplasm of others. Unilocular fat cells with peripheral flat nucleus and thin rim of cytoplasm were clarified (Figure 3e).

Group III (MWL Group): Sections belonging to this group recruited undulant skin folds (Figure 4a). Relatively decreased thickness of epidermis and obvious separation of the fibers in the reticular dermis were found in other fields (Figure 4b). Some other fields revealed atypical epidermal cells with dark nuclei and cytoplasmic vacuolations (Figure 4c). Remarkable separation of dermal fibers was noticed around sweat glands (Figure 4d). In addition, loss of architecture of some sweat glands and some others showed apoptotic nuclei and cytoplasmic vacuolations in some cells (Figure 4e).

Masson's Trichrome Stained Sections

Control group, showed dense collagen dermal fibers (Figure 5a), group II, some disrupted collagen fibers appeared in the dermis (Figure 5b) and in group III, obviously disrupted collagen fibers and fibers with atypical staining character were seen in the dermis (Figure 5c).

Orcein Stained Sections

Group I revealed normal dermal wavy thin elastic fibers (Figure 5d). In group II, some of the dermal elastic fibers were seen fragmented (Figure 5e). In group III, multiple elastic fibers were found to be fragmented (Figure 5f).

Immunohistochemical Results

Caspase3 Immunostained sections

Few Caspase3 +ve immunostained cells were seen in superficial epidermal layers in control group (Figure 6a). In morbid obesity group, multiple positive epidermal cells existed (Figure 6b). In massive weight loss group, most of the epidermal cells were +ve (Figure 6c).

CD105 Immunostained sections

In control group +ve CD105 immunostaining was detected in basal cell layer (Figure 6d). In morbid obesity group, +ve cells were evident in basal and suprabasal layers (Figure 6e). In massive weight loss group, a few +ve cells appeared (Figure 6f).

Morphometric Results

A significant (P<0.05) the mean thickness of epidermis was increased in morbid obesity group, versus the other two groups, a significant decrease in massive weight loss group compared to control group. An increase was also found area (μ2) of dark nuclei in morbid obesity group and in massive weight loss group versus control group. In addition, an increase was found in area of cytoplasmic vacuolations in morbid obesity group and in massive weight loss group versus control group. Finally, a increase in area of separation of fibers was detected in massive weight loss group versus morbid obesity group (Table 2, Histogram 1). A decrease was found in area% of collagen and elastic fibers in massive weight loss patients versus the other 2 groups. In addition, a decrease was found in morbid obesity group versus control group as regards area% of elastic fibers (Table 3, Histogram 2). An increase was found in area% of caspase3 IE in massive weight loss patients, versus the other 2 groups and in morbid obese patients compared to control group. On contrary, a decrease was evident in area% of CD105 IE in massive weight loss group and an increase in morbid obesity group (Table 4, Histogram3).

PCR results

Estimation of TNFα in pg/mg was (0.15±0.03) in control group, (0.98±0.07) in morbid obesity group and (2.86±0.05) in massive weight loss group, indicating an increase in weight loss group and in morbid obesity group versus control group (Histogram 4).

Biochemical results

DNA fragmentation and MDA values recorded an increase in MWL patients versus MO and normal weight patients, the MO group was significantly increased versus normal weight group While, GSH and catalase values determined a decrease in MWL patients versus MO and normal weight patients, the MO group was significantly decreased versus normal weight group (Table 5, Histogram 5).

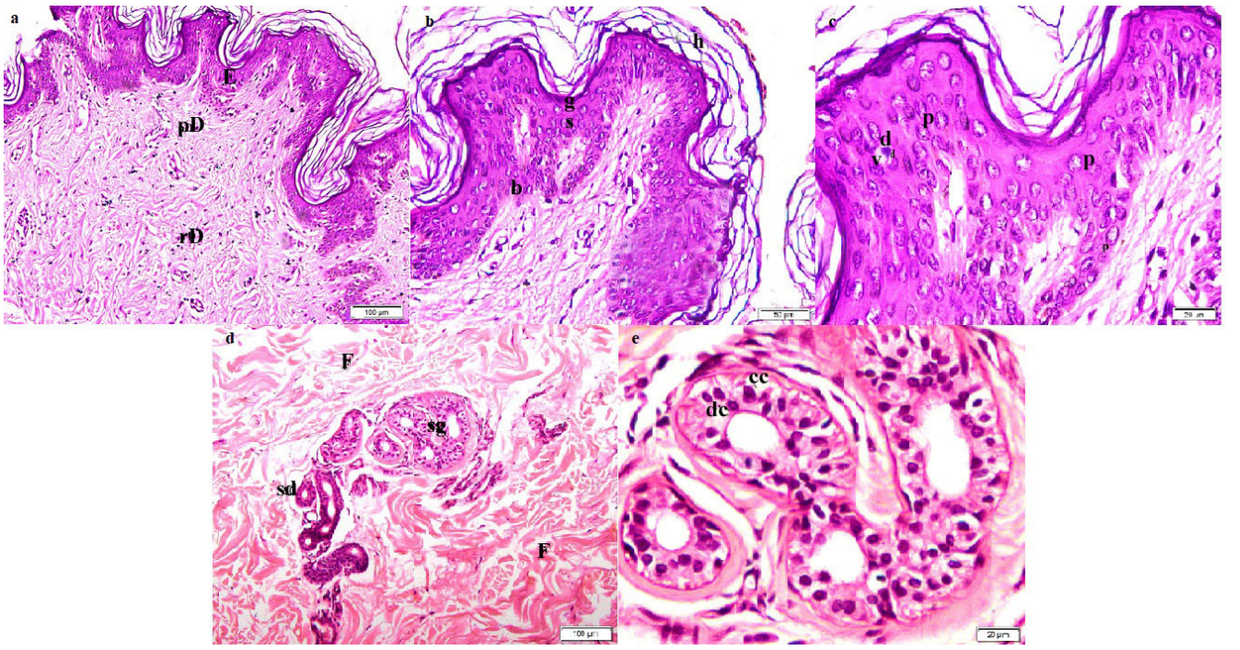


Fig. 2: Sections from the skin of abdomen of female patients in group I (Normal weight group) showing: a. layers of epidermal Keratinocytes (E), papillary (pD) and reticular (rD) dermis (H&Ex100). b. single basal (b), thick spiny (s), thin granular (g) and horny (h) layers, the latter formed of keratin filaments (H&Ex200). c. pale nuclei (p) in most of the epidermal cells. Dark nucleus (d) and vacuolated cytoplasm (v) of a cell in spiny layers are noticed (H&Ex400). d. apparently normal sweat glands (sg) and sweat ducts (sd) surrounded by apparently normal dense fibers (F) (H&Ex100). e. lining clear cells (cc) and dark cells (dc) of sweat gland (H&Ex400).

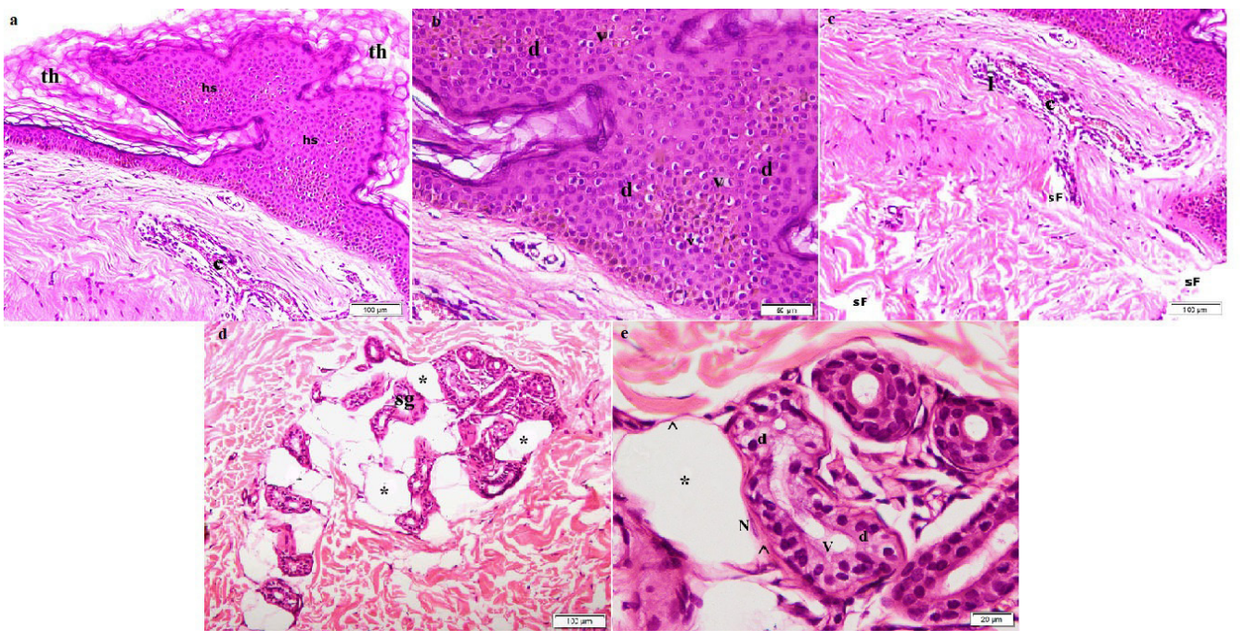


Fig. 3: Sections in the skin of patients in group II (Morbid obese group) showing: a. an area of thickened epidermis with hypertrophied spiny layer (hs) and thick horny (th) layer . Note a congested (c) blood vessel with minimal surrounding cellular infiltration in the papillary dermis (H&Ex100). b. multiple dark nuclei (d) and vacuolated cytoplasm (v) among the epidermal cells (H&Ex200). c. cellular infiltration (I) around a congested (c) blood vessel and areas separating fibers (sF) in the reticular dermis (H&Ex100). d. fat cells (*) among sweat glands (sg) (H&Ex 100). e. dark nuclei (d) of some lining cells and vacuolated (v) apical cytoplasm of others of sweat gland. Note unilocular fat cell (*) with peripheral flat nucleus (N) and thin rim of cytoplasm (^) (H&Ex400).

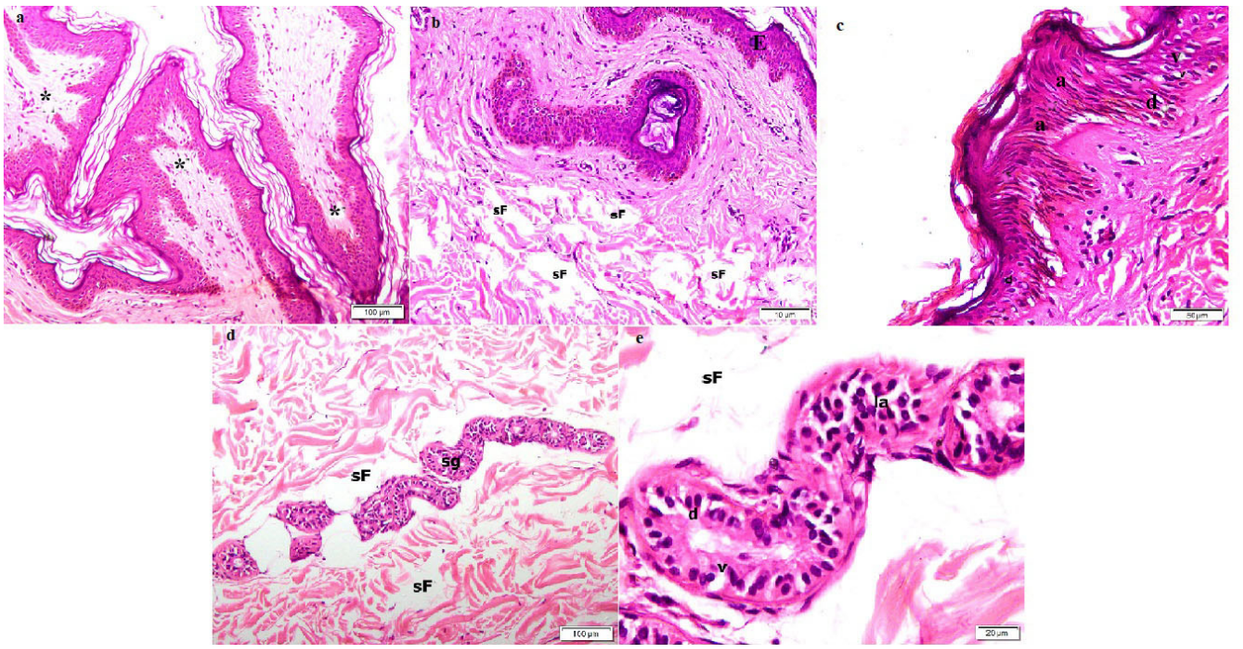


Fig. 4: Sections in the skin of patients in group III (Massive weight loss group) showing: a. undulant skin folds (*) (H&Ex100). b. relatively apparent decrease in thickness of the epidermis (E) and obvious separation of the fibers (sF) in the reticular dermis (H&Ex100). c. atypical epidermal cells (a). Note dark nuclei (d) and cytoplasmic vacuolations (v) (H&Ex200). d. obvious separation of dermal fibers (sF) around the sweat glands (sg) (H&Ex100). e. loss of architecture (la) of a sweat gland and another showing multiple darkly stained nuclei (d) and vacuolated cytoplasm (v) in some cells. Note surrounding separated fibers (sF) (H&Ex400).

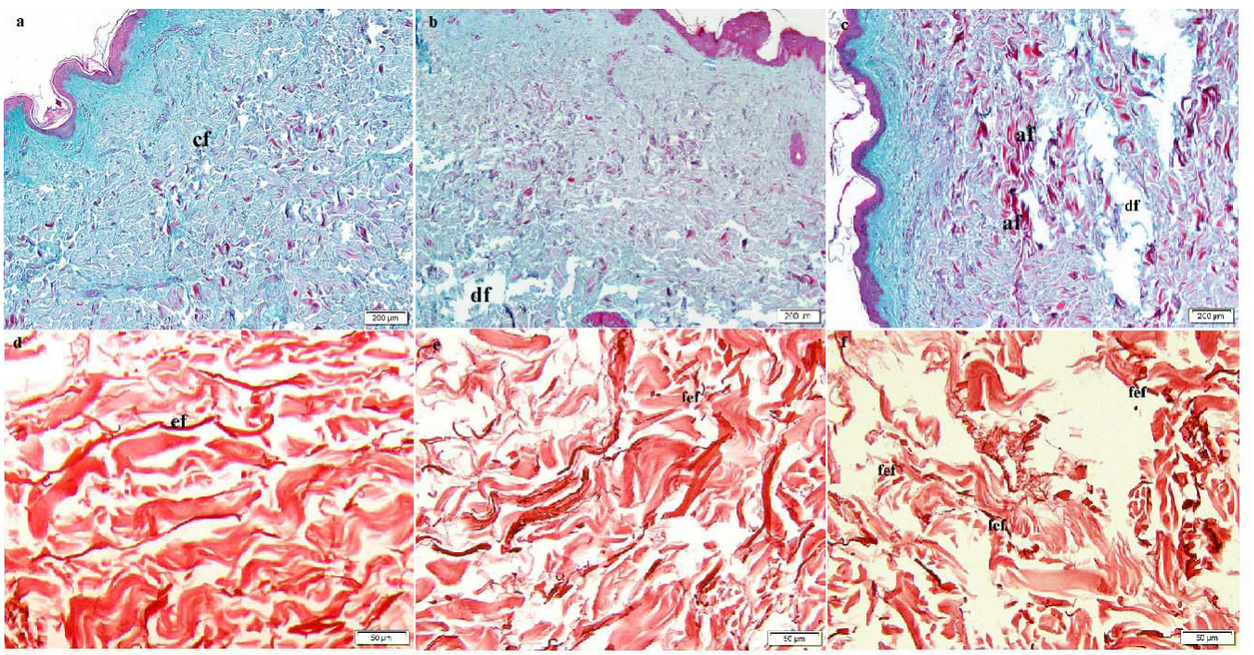


Fig. 5: Sections in the skin of patients showing: a. dense collagen fibers (cf) in the dermis of group I. b. disrupted collagen fibers (df) in the dermis of group II. c. obviously disrupted collagen fibers (df) and atypical red fibers (af) in the dermis of group III (Masson's trichrome, x 40). d. normal wavy thin elastic fibers (ef) in the dermis of group I. e. some fragmented elastic fibers (fef) in the dermis of group II. f. multiple fragmented elastic fibers (fef) in the dermis of group III. (Orcein stain, x 200)

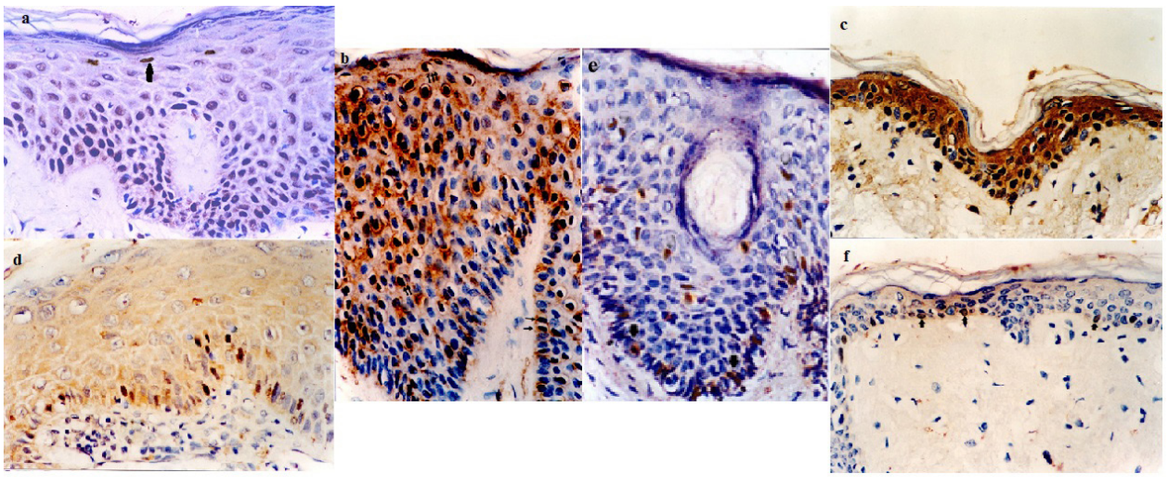


Fig. 6: Sections in the skin of patients showing: a. few +ve cells (arrows) in superficial layers of epidermis in group I. b. multiple +ve epidermal cells (arrows) in group II. c. most epidermal cells are +ve (arrows) in group III (Caspase 3 immunostaining, x400). d. +ve cells (arrows) in basal layer of epidermis in group I. e. multiple +ve cells (arrows) in basal and supra basal layers in group II. f. few cells are +ve (arrows) in group III (CD105 immunostaining, x400).

Table 2: Mean ± standard deviation (SD) of the thickness (th) of epidermis (ep), area of dark nuclei (dN), area of cytoplasmic vacuolations (cV) and area of separation (sp) between CT fibers in the different patients groups

Groups	th of ep	Area of dN	Area of Cv	Area of sp between CT fibers
Group I (Normal Weight Group)	49.00±13.25	0.84±0.13	1.07±0.15	0
Group II (Morbid Obese Group)	75.92±9.18*	6.03±1.75*	7.43±1.23*	35.34±5.89
Group III (Massive Weight Loss Group)	30.28±3.38•	3.02±0.18•	4.22±0.60•	69.94±7.19#

* significant compared to the other groups • significant compared to group I # significant compared to group II

Table 3: Mean ± SD of area% of collagen and elastic fibers in the different groups

Groups	Area% of collagen fibers	Area% of elastic fibers
Group I (Normal Weight Group)	71.07±6.15	7.39±1.15
Group II (Morbid Obese Group)	63.43±11.23	5.93±1.23•
Group III (Massive Weight Loss Group)	49.22±5.60*	3.52±0.60*

* significant compared to the other 2 groups • significant compared to group I

Table 4: Mean ± SD of area% of caspase3 +ve IE and that of CD105 +ve cells

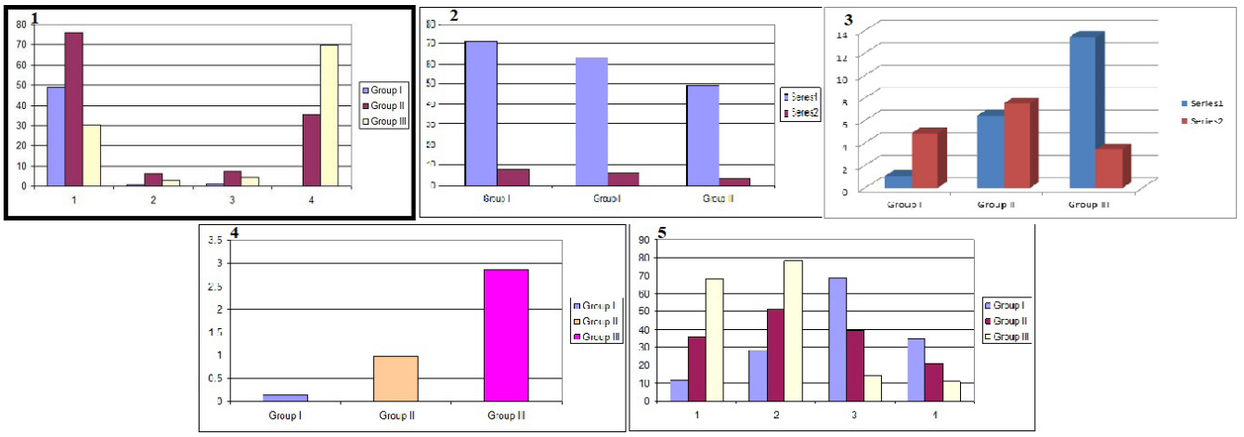
Groups	Area% of Caspase3 +ve IE	Area% of CD105 +ve cells
Group I (Normal Weight Group)	1.05±0.05	4.85±0.87
Group II (Morbid Obese Group)	6.43±1.03•	7.51±0.95∞
Group III (Massive Weight Loss Group)	13.42±2.50*	3.42±0.23^

• significant increase compared to group I
* significant increase versus other 2 groups
∞ significant increase versus other 2 groups
^ significant decrease versus other 2 groups

Table 5: Mean± standard deviation (SD) DNA fragmentation (fr)%, MDA, GSH and catalase values in control and experimental groups.

Group	DNA fr%	MDA (nM/gm)	GSH (mg/gm)	Catalase (U/gm)
Group I	11.04±0.93	28.24±3.92	68.76±8.04	34.42±3.93
Group II	35.88±5.02^	51.32±4.60^	39.41±3.22^	20.97±1.57^
Group III	68.02±3.91*	78.56±6.03*	13.93±5.58*	10.57±2.03*

* significant increase/decrease versus other 2 groups ^ significant increase/decrease versus group I



Histogram: 1: (1) Mean th of ep, (2) area of dN, (3) area of cV and (4)area of sp between CT fibers. 2: Mean area% of collagen and elastic fibers. 3: Mean ± SD of area% of caspase3 +ve IE and that of CD105 +ve cells. Series1 caspase3, series2 CD105. 4: Mean PCR values. 5: Mean DNA fragmentation (fr) %, MDA, GSH and catalase values.

DISCUSSION

A common misconception among plastic surgeons is that patients who have undergone significant weight loss will have significantly worse skin quality. This can be seen in a variety of ways, starting with a straightforward clinical examination of the skin, which reveals thin, inelastic, wrinkled, and highly folded dry skin[17]. It is also noted that after cosmetic procedures, as in abdominoplasty, postoperative complications are frequently more common in MWL patients and include delayed wound healing, wound disruption, and postoperative skin redundancy, particularly in comparison to normal weight people who underwent the same procedures[18].

Numerous dermatological abnormalities are induced by obesity, including changes in the epidermal barrier, appendages, vascularisation, and wound healing[19]. Morbid obesity is considered to increase the incidence of wound complications and post-operative infection[20].

Obesity-related diseases have remarkebly improved in response to sleeve gastrectomy, and more patients are achieving significant weight loss in response to media promotion of diet and exercise. Following weight loss, most of these patients visit a plastic surgeon for abdominoplasty[21].

A considerable decline in obesity-related medical disorders has resulted from such extreme weight loss, however postoperative wound problems are still prevalent[22]. Studies aiming to explain this increased complication risk usually include assessment of nutritional factors, weight loss mechanisms, and cutaneous changes. Extreme weight loss affects skin integrity, surgical wound problems, and dermal fibers[23].

A rising number of patients are seeking substantial body contouring procedures because of the rise in class III obese individuals and the need for sleeve gastrectomy. In the present time, lower body lift procedures started to be performed in addition to traditional abdominoplasties[24].

It was discovered that elevating the flap during an abdominoplasty in a plane superficial to the traditional suprafascial technique may reduce seroma formation[25]. In order to ensure that the superior flap had an adequate blood supply during undermining, the costomarginal branch of the deep superior epigastric artery was preserved. To further lessen the danger of flap necrosis, limited lateral undermining, not extend over the anterior axillary line, as well as minimal excision or suction, were performed[26].

In the present work, histological sections from MO group showed some areas of thickened epidermis with hypertrophied spiny layers and atypical horny layers and congested blood vessels. Other fields demonstrated cellular infiltration and separated dermal fibers. This was proved morphometrically. These findings indicated inflammatory changes that are consistent with other researchers[27] who found a significant increase in thickness and alterations in structure of abdominal skin in obese patients. A close association was reported between skin inflammatory diseases and obesity, due to inflammatory chemical mediators released from fatty tissue and activation of immune cells[28].

Other fields demonstrated multiple dark nuclei and cytoplasmic vacuolations among the epidermal cells, confirmed morhometrically when compared to the other groups. These findings indicated degenerative changes. In support, a mechanism in which obesity contributes in affection of skin integrity and health is the increase in saturated fatty acids from high fat diet that cause oxidative skin injury[29]. Cell death was proved to be influenced and triggered by obesity[30].

In this group, some disrupted collagen fibers and fragmented elastic fibers appeared in the dermis. This was proved morphometrically. These results were supported by the work done by[31], where the authors found decreased dermal collagen and elastic fibers density in obese patients. In agreement, Makihara *et al*[32] reported that the higher the BMI the less becomes the skin elasticity. They mentioned

that relative decrease of dermal fibers formation is unable to face the increase in skin area followed by mechanical weakness of skin.

Fat cells were seen among sweat glands, as evidenced by Sami *et al*,[33]. Wen *et al*,[34] related fat infiltration of non-adipose tissues to increased fat intake and consequently deposition. In addition, dark nuclei of some lining cell of sweat glands and vacuolated apical cytoplasm of others were observed. Ibuki *et al*,[35], reported aging-like skin changes as decreased skin hydration and dermal collagen contents in obese subjects. Zhang *et al*,[36] postulated that hypertriglyceridemia appears to mediate cell apoptosis, via significant up-regulation of catabolism genes.

Sections belonging to MWL group recruited undulant skin, relatively less epidermal thickness, obvious separation of dermal fibers and infiltration of the epidermis by fibroblast-like cells with dark nuclei and cytoplasmic vacuolations. This was proved morphometrically. The previous findings suggested degenerative changes. In support, marked thinning, deformities and degenerated keratinocytes were observed in the epidermis[33]. It was added that body weight loss over a shorter period activates autophagy and ubiquitin-proteasome systems in cells and tissues when compared to slower weight reduction[37]. Klassen *et al*,[38] correlated excessive weight loss with remarkable unhealthy skin in patients following bariatric surgery.

Obviously disrupted collagen fibers and fibers with atypical staining character as well as multiple fragmented elastic fibers were seen in MWL patients. These findings were proved morphometrically. The previous results were consistent with Manzoni and Weber,[39]. and Rocha *et al*,[40] whom revealed structural dermal fibers alterations in group III after sleeve gastrectomy.

Dermal adipocytes secrete adipocytokines, such as adiponectin resulting in increased production of hyaluronic acid, collagen, elastin and matrix metalloproteinases (MMPs) of dermis that promotes healing in the skin[41]. The marked adipose tissue loss following sleeve gastrectomy leads to decrease in adipokine production. Consequently the dermal fibroblasts decrease production of hyaluronic acid, collagen and elastic fibers[42]. Prist *et al*.[43] demonstrated that massive weight loss after sleeve gastrectomy decreased expression of MMP, contributing for defective healing. This is further supported by Simonacci *et al*,[44], who found collagen and elastic fibers depletion, when weight loss is massive confirmed by histomorphometric assessment.

In addition, loss of architecture of some sweat glands and some others showed degeneration of lining cells in group III. Concomitantly, it was stated that calories restriction and weight loss when massive over a short period may exert impact on cell and tissue integrity[45].

Caspases are key mediators of apoptosis, caspase-3 is necessary for efficient programmed cell death and prevents the formation of ROS[46]. In group II, multiple +ve epidermal cells were detected, while in group III, most of the epidermal cells were +ve. It was proved that pathogenesis of apoptosis is related to are ROS and inflammation. ROS overproduction destroys antioxidant cellular activity and metabolic processes[47].

In the present study, in MO group, CD105 +ve stem cells were evident in basal and suprabasal layers while in MWL group, few +ve stem cells appeared. In support, resident skin SCs support cellular turnover during skin homeostasis, healing and regeneration after injury. Skin SCs reside in interfollicular epidermis[48]. Mesenchymal stromal cells (MSC) have high plasticity referred to exosome release and paracrine effect that exert immunomodulatory impact. Proliferative, migratory and differentiation abilities, in addition to angiogenesis, are involved in skin regeneration[49]. In damaged skin, MSCs have healing capacities by stimulating production of collagen and elastic fibers[50]. SCs are also sensitive to systemic changes in skin histophysiology, injuries, and surgeries[51] Co-expression of SC marker CD105 supports identity of MSCs[52].

CONCLUSIONS

It can be concluded that patients presenting with massive weight loss following sleeve gastrectomy should accept higher complication rates and revisional procedures for recurrent redundancy due to altered skin behaviour. The beneficial therapeutic outcome could be related to the plasticity of skin SCs to transdifferentiate into adult skin cells of epidermal barrier and dermal fibers responsible for skin integrity.

CONFLICT OF INTERESTS

There are no conflicts of interest.

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الملخص العربى

التغيرات المورفولوجية للجلد والخلايا الجذعية المقيمة في المرضى الإناث الذين يعانون من السمنة المفرطة وفقدان الوزن الهائل الخاضعين لعملية تكميم المعدة دراسة هيستولوجية كيميائية اكلينكية

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الخلفية والاهداف: يواجه المرضى الذين يعانون من زيادة الوزن تغيرات تشريحية واضحة في مظهر جلدهم وبالتالي من المتوقع حدوث تغييرات شكلية. يؤدي فقدان الوزن بشكل كبير بعد عملية تكميم المعدة إلى ترهل الجلد. يستهدف العمل الحالي تحديد تأثير التغيرات المورفولوجية والكمية للبشرة والألياف الجلدية والخلايا الجذعية الذاتية على نتائج الجراحة لدى النساء اللواتي يعانين من زيادة الوزن من الدرجة الثالثة والذين يعانون من فقدان الوزن بشكل كبير بعد تكميم المعدة.

المرضى وطرق العلاج: تم الحصول على عينات الجلد من الجلد المستأصل اثناء العمليات الجراحية من ثلاثين مريضة ، تم تصنيفها إلى ثلاث مجموعات (١٠ مرضى لكل مجموعة). مجموعة ذات وزن طبيعي مع تاريخ من عدم فقدان الوزن والذين خضعوا لعملية شد البطن. مجموعة السمنة المرضية والتي خضعت لعملية تكميم المعدة. مجموعة خسارة الوزن الهائلة والتي لديها تاريخًا من فقدان الوزن الهائل بعد تكميم المعدة والتي خضعت لعملية شد البطن. تم أخذ عينات الجلد أثناء عملية شد البطن بعد الاستئصال الجراحي لجلد البطن الزائد في المجموعتين الأولى والثالثة وأثناء عملية التكميم من حافة الجرح في المجموعة الثانية. تم إخضاع المقاطع للدراسات الهيستولوجية والكيميائية الحيوية والنمطية والكمية إلى جانب التفاعل الكمي للبوليميراز المتسلسل (qPCR)

النتائج: كانت مضاعفات الجروح التي حدثت في مرضى فقدان الوزن الهائل أقل في مرضى السمنة المفرطة ، وبعضهم يحتاج إلى إعادة الجراحة بسبب تكرار الترهلات مرة اخري . كانت التغيرات المظهرية والمورفومترية المصاحبة لحاجز البشرة والألياف الجلدية وعلامات التنكس و الخلايا الجذعية الذاتية أقل وضوحًا في مرضى السمنة المفرطة.

الاستنتاجات: يجب على المرضى الذين يعانون من فقدان الوزن بشكل كبير بعد عملية تكميم المعدة قبول معدلات مضاعفات أعلى وإجراءات مراجعة للترهل المتكرر مرة اخري بسبب سلوك الجلد المتغير. يمكن أن تكون النتيجة العلاجية المفيدة مرتبطة بمرونة الخلايا الجزعية للجلد لتتحول إلى خلايا الجلد البالغة لحاجز البشرة والألياف الجلدية المسؤولة عن سلامة الجلد.