Research

Liposuction Preserves the Morphological Integrity of the Microvascular Network: Flow Cytometry and Confocal Microscopy Evidence in a Controlled Study

Aesthetic Surgery Journal
2016, Vol 36(5) 609–618

DOI: 10.1093/asj/sjv209
www.aestheticsurgeryjournal.com
OXFORD UNIVERSITY PRESS

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Abstract

Background: Liposuction is a very popular technique in plastic surgery that allows for the taking adipose tissue (AT) on large surfaces with little risk of morbidity. Although liposuction was previously shown to preserve large perforator vessels, little is known about the effects of liposuction on the microvasculature network.

Objectives: The aim of this study was to analyze the effect of liposuction on the preservation of microvessels at tissue and cellular levels by flow cytometry and confocal microscopy following abdominoplasty procedure.

Methods: Percentage of endothelial cells in AT from liposuction and en bloc AT was determined by multicolor flow cytometry. Moreover, vessel density and adipocyte content were analyzed in situ in 3 different types of AT (en bloc, from liposuction, and residual AT after liposuction) by confocal microscopy.

Results: Flow cytometric analysis showed that en bloc AT contained 30.6% ± 12.9% and AT from liposuction 21.6% ± 9.9% of endothelial cells ($CD31^{pos}/CD45^{neg}/CD235a^{neg}/CD11b^{neg}$) ($P = .009$). Moreover, analysis of paired AT from the same patients (n = 5) confirmed a lower percentage of endothelial cells in AT from liposuction compared to en bloc AT (17.7% ± 4.5% vs 21.9% ± 3.3%, $P = .031$). Likewise, confocal microscopy showed that en bloc AT contained 8.2% ± 6.3%, AT from liposuction only 1.6% ± 1.0% ($P < .0001$), and AT after liposuction 8.9% ± 4.1% ($P = .111$) of $CD31^{pos}$ vessels. Conversely, adipocyte content was 39.5% ± 14.5% in the en bloc AT, 45% ± 18.4% in AT from liposuction ($P = .390$), and 18.8 ± 14.8% in AT after liposuction ($P = .011$).

Conclusions: For the first time, we demonstrate that liposuction preserves the microvascular network. Indeed, a low percentage of endothelial cells was found in AT from liposuction and we confirm the persistence of microvessels in the tissue after liposuction.

Accepted for publication September 14, 2015; online publish-ahead-of-print November 3, 2015.

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Liposuction has become widely popular since it was first described in 1983.[1,2] The procedure was originally used to treat localized fat excess.[3] Liposuction is now used in combination with other surgical techniques, such as abdominoplasty, medial thighplasty, and brachioplasty. For instance, the use of liposuction reduces flap thickness, thus easing tissue mobilization, while preserving lymphatic and perforating blood vessels, as well as connective tissues. Liposuction does not damage perforator vessels larger than 1 mm, as confirmed by Doppler ultrasound analysis carried out postoperatively.[4,5] The fact that liposuction preserves large perforating vessels raises the possibility of undermining by liposuction with maintenance of blood supply to the flap.

To our knowledge, no study has assessed the effect of liposuction on the microvascular network. Microcirculation is very important as it allows fluid, molecular, gas, and cellular exchanges.[6-9] Gas exchange was observed in vessels of up to 100 µm in diameter, which explains why microvessels are defined by an internal diameter measuring less than 100 µm.[10]

The aim of this study was to examine the effect of liposuction on the preservation of the microvascular network at tissue and cellular levels, and in particular, whether liposuction preserves the microvasculature network. The study was not designed to demonstrate if liposuction preserves microcirculation. This study is the first step towards improving our knowledge at the microvascular network level in plastic surgery. To address this issue, we analyzed adipose tissue (AT) harvested during abdominoplasty. AT consists of a large number of distinct cell types including adipocytes, endothelial cells, blood cells, adipose-derived stromal cells, and mural cells. Endothelial cells are the main components of microvessels. At the cellular level, we determined the proportion of endothelial cells in AT without liposuction (en bloc AT), compared to that in AT obtained from liposuction.

To analyze the percentage of endothelial cells in AT, the tissue was first dissociated by proteolytic enzymes to obtain the stromal vascular fraction (SVF), which contains all cell types except adipocytes. The percentage of endothelial cells ($CD45^{neg}/CD235a^{neg}/CD11b^{neg}/CD31^{pos}$) was determined by flow cytometry. If liposuction preserved the microvasculature, the number of endothelial cells found after enzymatic digestion of the aspirated abdominal fat (AT from liposuction) should be reduced, compared to the digested en bloc fat (AT without liposuction) from abdominoplasty. In addition, since the maintenance of microvascular functionality is only possible if the integrity of the vascular network is preserved, we used a complementary approach of confocal microscopy to determine the morphology of the microvasculature network in en bloc AT, AT from liposuction, and residual AT after liposuction, to correlate tissue morphology with results obtained at the cellular level by flow cytometry. Morphological examination demonstrated that the anatomical integrity of the vascular network was maintained in situ in the remaining tissues following liposuction.

METHODS

Adipose Tissue Collection

This study was approved by the Institutional Review Board, Centre Hospitalier Universitaire of Toulouse, and the study protocol was performed in accordance with the principles of the Declaration of Helsinki (1964) and in agreement with French bioethics laws of July 7 2011. Written informed consent was obtained from all study patients. AT was harvested during plastic surgery from 18 healthy adult patients at Rangueil Hospital (part of CHU of Toulouse). Inclusion criteria included AT harvested during abdominoplasty, adult patients aged over 18 years, no past medical history, and sample processing within 4 hours after AT harvest. Exclusion criteria included AT from other reconstructive procedures (eg, brachioplasty, medial thigh lift), previous abdominal surgery, and the presence of medications. Smokers were invited to stop smoking to be eligible for surgery. All eligible patients for the study were enrolled between March and October 2014. In the first 13 patients, AT from liposuction was analyzed; en bloc AT was analyzed when liposuction was not performed. In seven patients, lipoaspirates were harvested at the beginning of abdominoplasty using the traditional suction-assisted lipectomy (SAL) technique.[11] Briefly, a wetting solution (approximatively 500 mL of lactated Ringer's solution containing adrenaline) was infiltrated manually into subcutaneous fat using a 60-cc syringe to reduce bleeding before SAL. Liposuction was performed through a small incision with the use of 3 and 4 mm diameter cannulae and a vacuum pressure of 600 mmHg. In contrast, in six other patients, abdominoplasty was performed en bloc without the use of liposuction. Finally, to eliminate inter-individual differences, in the remaining five patients, liposuction was done on half of the abdominal wall and en bloc resection without liposuction on the other half of the abdominal wall (Figure 1). Samples from these five patients were analyzed by flow cytometry. In addition, of these five patients, AT samples in two patients were analyzed by confocal microscopy to evaluate the residual AT after liposuction; flow cytometry could not be used as the connective tissue was poorly digested.

Isolation of Stromal Vascular Fraction

Adipose tissue was digested for 45 minutes in α-Minimum Essential Medium (MEM; Life Technologies, Carlsbad, CA) containing collagenase NB 4 (final concentration: 0.4 IU/mL, Serva Electrophoresis, Heidelberg, Germany), Dispase II

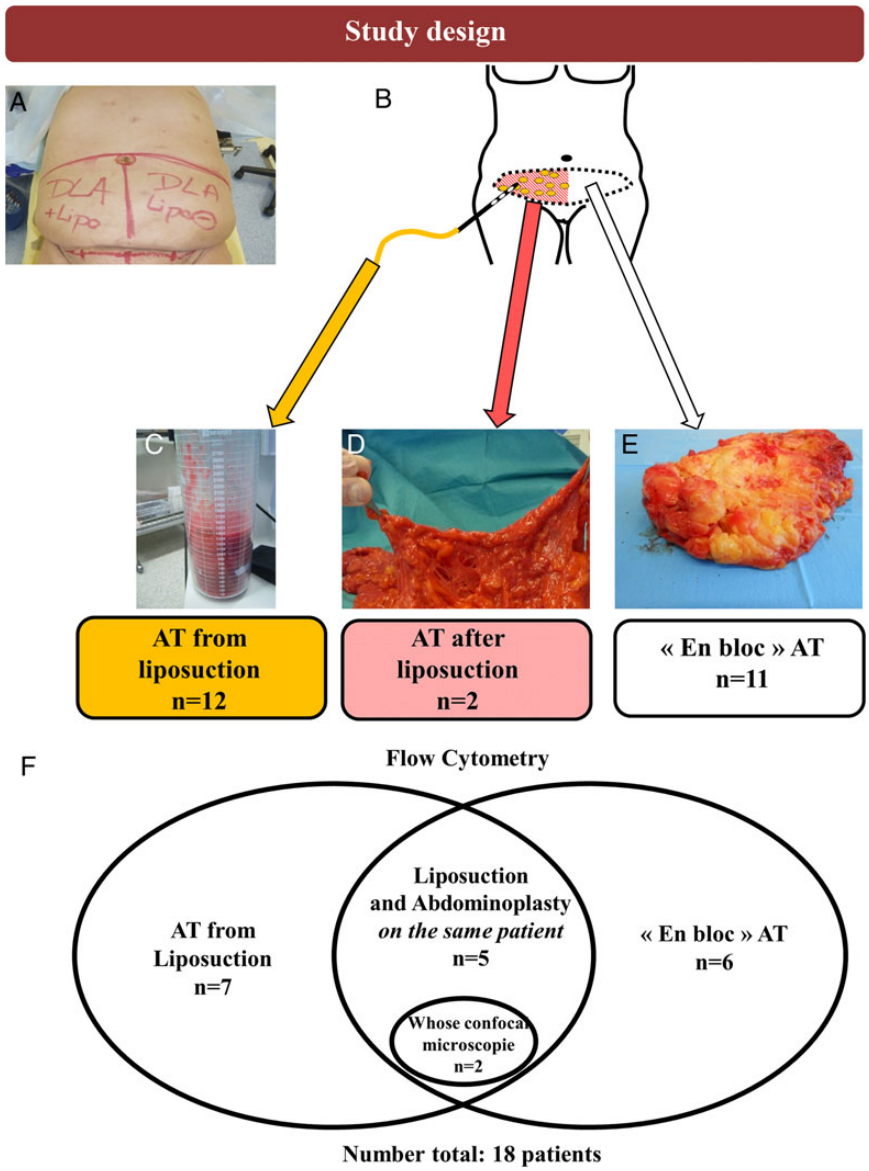

Figure 1. Design of the study. (A) Preoperative view of patient (54-year old man) with drawings. (B) General outline of the study. (C) For seven patients, we obtained only abdominal lipoaspirates (AT from liposuction). (D) The remaining AT after liposuction was analyzed by confocal microscopy only. (E) For six patients, we recovered abdominal tissue without liposuction (en bloc AT). (F) Five patients underwent liposuction on half of the abdominal wall and en bloc resection without liposuction on the other half of the abdominal wall. To eliminate inter-individual differences, AT from these patients were comparatively analyzed by flow cytometry (C and E; n = 5) and by confocal microscopy (C, D, and E; n = 2).

(final concentration: 1.6 UI/mL, Roche, Basel, Switzerland), Pulmozyme (final concentration: 10 UI/mL, Roche), and penicillin-streptomycin (Life Technologies) in a shaking water bath at 37°C. After digestion, an equal volume of α-MEM containing 10% fetal bovine serum was added to stop enzymatic digestion. The cells were passed through a 100 µm filter (Steriflip, Millipore, Billerica, MA) and then centrifuged. The pellet was resuspended in α-MEM containing 10% fetal bovine serum and 1% penicillin-streptomycin, and the total number of cells in the SVF was counted.

Flow Cytometry

Freshly isolated cells from the SVF were incubated for 20 minutes at 4°C in running buffer (Miltenyi, Bergisch Gladbach, Germany) containing an FcR blocking reagent (Miltenyi) to minimize nonspecific antibody binding. Then cells were stained simultaneously for 30 minutes at 4°C with the following monoclonal mouse antihuman fluorochrome-conjugated antibodies: CD45-fluorescein isothiocyanate (FITC; Miltenyi), CD11b-FITC (Miltenyi), CD235a-FITC

(Beckman Coulter, Fullerton, CA), CD31-PerCPeFluor710, and CD34-APCeFluor780 (both from eBiosciences, Santa Clara, CA). Cells were washed for 10 minutes in lysis buffer containing ammonium chloride (Stemcell Technologies, Vancouver, BC, Canada) to remove red blood cell contaminants and then filtered through a 40 µm filter. Finally, cells were centrifuged, the supernatant removed, and cells resuspended in phosphate-buffered saline (PBS) and 4′,6-diamidino-2-phenylindole (DAPI). Acquisitions were performed using a BD Influx cytometer (Becton Dickinson, Franklin Lakes, NJ). For compensation purposes, mouse immunoglobulin (Ig) and negative control compensation particles (BD CompBeads; Becton Dickinson) were stained with single fluorochrome-conjugated antibodies (FITC, PerCPeFluor710, APCeFluor780). Gating was achieved using isotype control staining.

Data were analyzed using the Kaluza software (Beckman Coulter). First, we plotted a diagram showing the size (forward scatter) and granularity (side scatter) for cell selection by excluding cell debris and platelets. Live cells (DAPI-negative cells) were then selected before exclusion of doublets. Hematopoietic cells were excluded based on the expression of CD45 (panhematopoietic marker), CD235a (marker for mature erythrocytes and erythroid precursor cells), and CD11b (myeloid cells). Endothelial cells were selected based on CD31 marker expression. The gating strategy is shown in Figure 2.

Confocal Microscopy

Adipose tissue was harvested by one of the authors (BC). Preparation and staining of adipose samples for confocal microscopy were performed by another author (NB). Nine or 10 samples were prepared for each tissue group (en bloc AT, AT from liposuction, AT after liposuction). All samples were taken randomly at different sites of the AT (between the superficial fascia system and the rectus fascia). Image acquisition of anonymized samples was performed by a third author (SBM) by selecting a representative point from entire samples. Data analyses used Imaris software (Bitplane, South Windsor, CT). Having two different individuals independently performing image acquisition and data analysis helped to limit study bias.

Briefly, AT samples (4-6 mm^3) were fixed in 4% paraformaldehyde for 12 hours at 4°C, blocked in PBS/3% horse serum/5% bovine serum albumin (BSA)/0.2% Triton for 3 hours at room temperature, and incubated in PBS/3% horse serum/5% BSA/0.2% Triton containing the primary antibody (monoclonal mouse antihuman CD31; Dako, Glostrup, Denmark) overnight at 4°C. After washing in PBS, AT samples were incubated overnight with the secondary antibody (donkey antimouse Alexa Fluor-488; Invitrogen, Carlsbad, CA). Adipocytes were stained with BODIPY 558/568 (dilution 1:2000; Invitrogen) for 1 hour at room temperature. Nuclei were stained with DRAQ5 (dilution 1:2000; Biostatus, Leicestershire, UK) for 15 minutes at room temperature. Image acquisition was performed blindly from the sample preparation using a LSM510 confocal laser scanning microscope and an Axiovert 200 M inverted microscope (Carl Zeiss, Jena, Germany). AT samples were visualized with a 10 × magnification objective lens and excited fluorescence using three laser lines (488, 543, and 633 nm). Emitted fluorescence was collected using appropriate filters (BP 475-525, BP 560-615, and LP 650 filters, respectively). Five image stacks with a z-step of 1 µm were collected from samples of AT from liposuction, residual AT after liposuction, and AT without liposuction (en bloc AT) from the same two individual patients. Image stack thickness varied from 20 to 50 µm.

In all experiments, the final image was produced by overlapping 10 optical slices using Fiji software. Three-dimensional reconstruction and quantification of $CD31^{pos}$ vessels and $BODIPY^{pos}$ adipocytes were performed using the Imaris software (Bitplane, Zurich, Switzerland). Surface objects on the desired channel were created using the same fluorescence threshold for each image. The vascular or adipocyte density was defined as the ratio between the total volume of surface objects (representing blood vessels or adipocytes, respectively) and the volume of the image.

Statistical Analyses

Statistical analyses were performed using GraphPad Prism 5 software (GraphPad Software, La Jolla, CA). Results were expressed as mean ± standard deviation. The Mann-Whitney nonparametric test was used to compare the two groups assessed by flow cytometry and the three groups assessed by confocal microscopy. The Wilcoxon matched-pairs signed rank test was used to analyze the flow cytometry results of en bloc AT and AT from liposuction from the same patients. Differences between groups were considered significant at a P-value < .05.

RESULTS

Eighteen patients who underwent abdominoplasty after weight loss were enrolled in the study. Two patients were male and 16 were female. The average age was 42.4 ± 11.7 years (range, 27-62 years). The mean weight was 72.7 ± 13 kg (range, 56-106 kg), the mean height was 170 ± 6 cm (range, 155-180 cm), and the mean body mass index was 26.5 ± 4.7 kg/m^2 (range, 21.5-39.9 kg/m^2). Of those 18 patients, five had liposuction carried out on half of the abdominal wall and en bloc resection without liposuction on the other half of the abdominal wall to minimize inter-individual differences (Figure 1).

Flow cytometry analysis was performed on freshly isolated SVF cells from the two types of AT. The proportion of cells

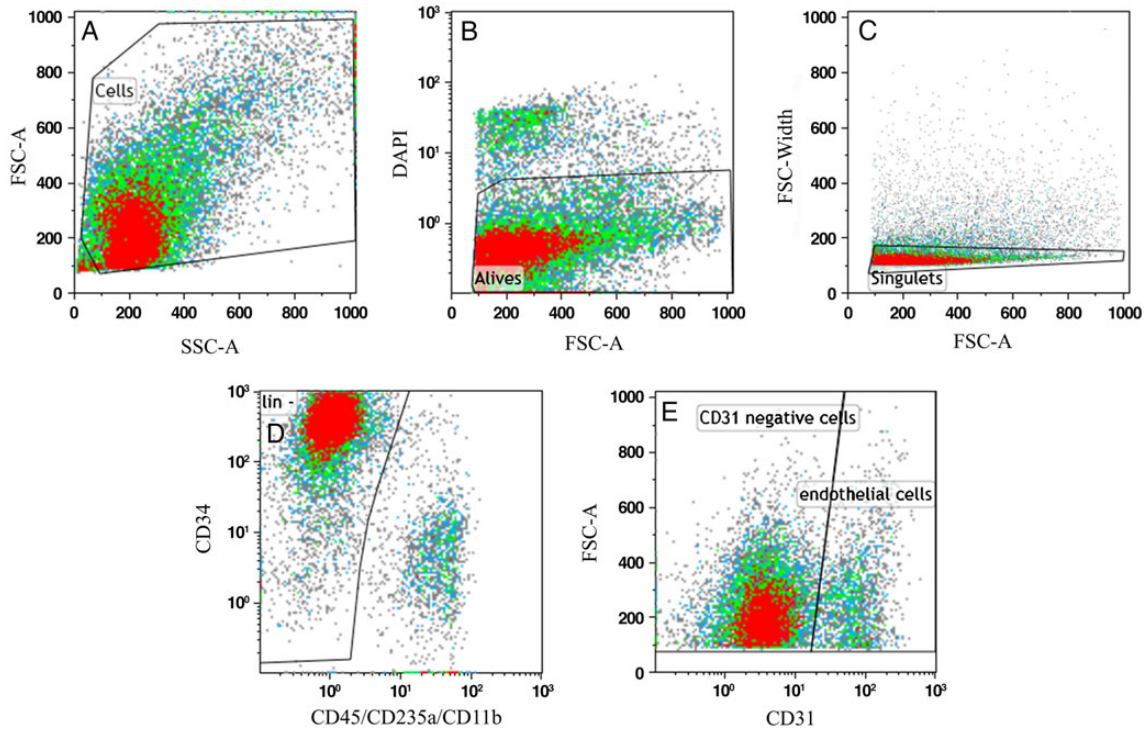

Figure 2. Gating strategy for the quantification of endothelial cells. (A) The cells were initially selected based on their physical parameters concerning the forward and side scatter area (excluding cell debris and platelets). (B) The dead cells identified using 4′,6-diamidino-2-phenylindole were excluded. (C) The single cells were selected based on forward area and forward width. (D) Hematopoietic cells were excluded upon the expression of CD45 (pan hematopoietic marker), CD235a (mature erythrocytes and erythroid precursor cells marker), CD11b (expressed on the surface of leukocytes including monocytes, macrophages, neutrophils and granulocytes). The CD34 marker (hematopoietic stem cells and endothelial marker) was used for the gating strategy. (E) Endothelial cells were selected on the expression of CD31 marker.

expressing CD45 (panhematopoietic marker), CD11b (myeloid cells), and CD235a (erythrocytes) was 25.6% ± 8.2% in en bloc AT and 29.9% ± 16.2% in AT from liposuction (P = .49). After exclusion of hematopoietic cells, $CD31^{pos}/CD45^{neg}/CD235a^{neg}/CD11b^{neg}$ endothelial cell content was analyzed in the SVF from samples of en bloc AT (n = 11) and AT from liposuction (n = 12). There was a statistically significant difference in the proportion of endothelial cells between the two AT groups: 30.6% ± 12.9% in en bloc AT and 21.6% ± 9.9% in AT from liposuction (P = .009; Figure 3A). To minimize inter-individual differences, we analyzed the percentage of endothelial cells in paired en bloc AT and AT from liposuction from the same five individual patients. A higher percentage of endothelial cells was evident in en bloc AT compared to AT from liposuction (21.9 ± 3.3 vs 17.7% ± 4.5%, respectively, P = .031; Figure 3B). However, tissue digestion was incomplete in the en bloc AT group, since connective tissue and macroscopic vessels were still present after 45 minutes of enzymatic digestion (Figure 4). Therefore, we could infer an underestimated number of endothelial cells in en bloc AT.

The decrease in the endothelial cell content in liposuction suggested that liposuction partially preserves the microvasculature. To confirm this hypothesis, confocal microscopy analysis was performed on two patients to investigate the vascular network in en bloc AT, AT from liposuction, and residual AT after liposuction (Figure 1). We performed multicolor staining using CD31 for labeling endothelial cells, BODIPY for adipocytes, and DRAQ5 for nuclei. Nine to 10 samples were prepared from each tissue and one acquisition was performed from each sample. In en bloc AT, we observed a well-organized vascular network (Figures 5D and 6A–C). In residual AT after liposuction, many large microvessels exceeding 80 µm were preserved as well as many intermediate microvessels (between 30 and 80 µm). Moreover, we noted areas where all adipocytes were aspirated, but with a microvascular network comprising vessels with a diameter of less than 30 µm still visible (Figures 5H and 6D–F). In AT from liposuction, we observed that adipocytes were less organized compared to en bloc AT. Interestingly, the vascular network was disrupted and anarchic. However, we did not visualize any vessel with a diameter greater than 30 µm (Figures 5L and 6G–I). Liposuction can remove elements of the microcirculation localized around the adipocytes. Presently, however, liposuction retained many microvessels in the remaining

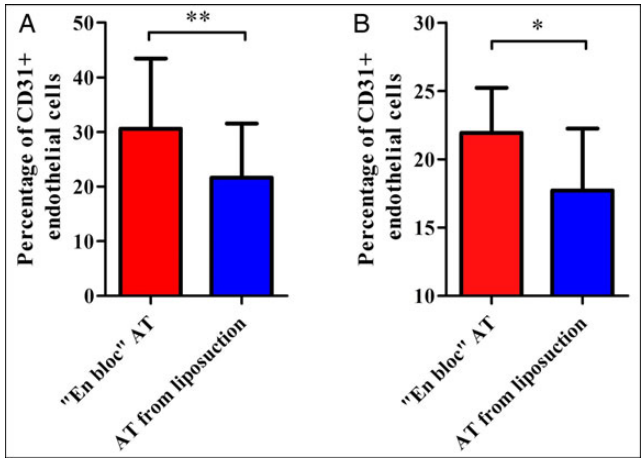

Figure 3. Flow cytometry results. (A) Percentage of endothelial cells ($CD31^{pos}/CD45^{neg}/CD235a^{neg}/CD11b^{neg}$) on the total population (non-paired samples). (B) Percentage of endothelial cells on the five patients who underwent both surgical techniques (samples from the same patient): en bloc abdominoplasty and liposuction. $^{*}P < .05$, $^{**}P < .01$.

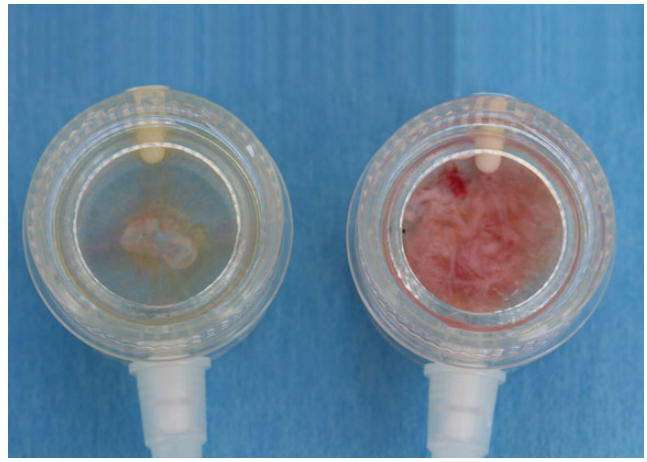

Figure 4. Results of undigested tissue in a 100 µm Steriflip filter (Millipore). AT from liposuction (left) and en bloc AT (right).

connective tissue. Figure 6 shows three examples for each group. We then quantified the vascular density using the Imaris software by calculating the proportion of microvascular network in the tissues. En bloc AT contained 8.2% ± 6.3% of vessels, AT from liposuction 1.6% ± 1.0% ($P < .0001$), and residual AT after liposuction 8.9% ± 4.1% ($P = .111$) (Figure 7A). Similarly, we observed that the adipocyte density in en bloc AT was 39.5% ± 14.5%, compared to 18.8% ± 14.8% in residual AT after liposuction ($P = .011$). In contrast, no difference was found between en bloc AT and AT from liposuction (45% ± 18.4%, $P = .39$) (Figure 7B). These results show that, after liposuction, part of the microvasculature was preserved, while as expected the adipocyte content was decreased.

DISCUSSION

First described in 1983,[1] liposuction has become a highly effective procedure in plastic surgery and is used increasingly for body contouring surgery.[2] The technique has undergone many refinements and has evolved over the last 25 years.[11] In this study, our aim was to gain further insight into the preservation of the microvascular network after traditional SAL.[11] Liposuction preserves perforator vessels detectable on ultrasound, but little is known about this effect at the microvasculature level where fluid, molecular, and cellular exchanges occur. To our knowledge, only one study has described the structure and cellular composition of excised AT and lipoaspirate.[12] No tissue analysis after liposuction was performed. The present data demonstrate the partial preservation of microvessels after liposuction.

This study provides an experimental and novel way of demonstrating the effect of liposuction on the microvasculature. Flow cytometry allows the characterization of the SVF, which contains all cell types except adipocytes. After enzymatic digestion, flow cytometry showed a higher percentage of $CD31^{pos}$ endothelial cells in en bloc AT, compared to AT from liposuction. Of note, tissue digestion was incomplete in en bloc AT, since connective tissue and macroscopic vessels (perforators) still remained after 45 minutes of enzymatic digestion. Consequently, it is possible that our results underestimated the number of endothelial cells in en bloc AT, and the difference observed in the percentage of endothelial cells probably relied on the microvasculature density, rather than on the presence of large blood vessels. Our results differ from those of Eto et al[12] who found a higher percentage of endothelial cells in aspirated AT compared to intact tissue. These authors explained that the cellular composition of the SVF might be biased by incomplete tissue digestion and emphasized that their results obtained by flow cytometry did not match the images of whole-mount tissue, unlike the present findings. One difference between their study and ours was the composition of the digestion buffer (collagenase plus Dispase vs collagenase) and the digestion time (45 vs 30 minutes). The addition of Dispase to the digestion buffer induces improved tissue digestion with the disruption of fibronectin and collagen types I and IV. Thus, it is likely cellular digestion was more effective in our study, which would explain the differences in our findings compared to previously reported results. Importantly, despite some variability between donors, we consistently obtained fewer endothelial cells in AT after liposuction than in en bloc AT from those five

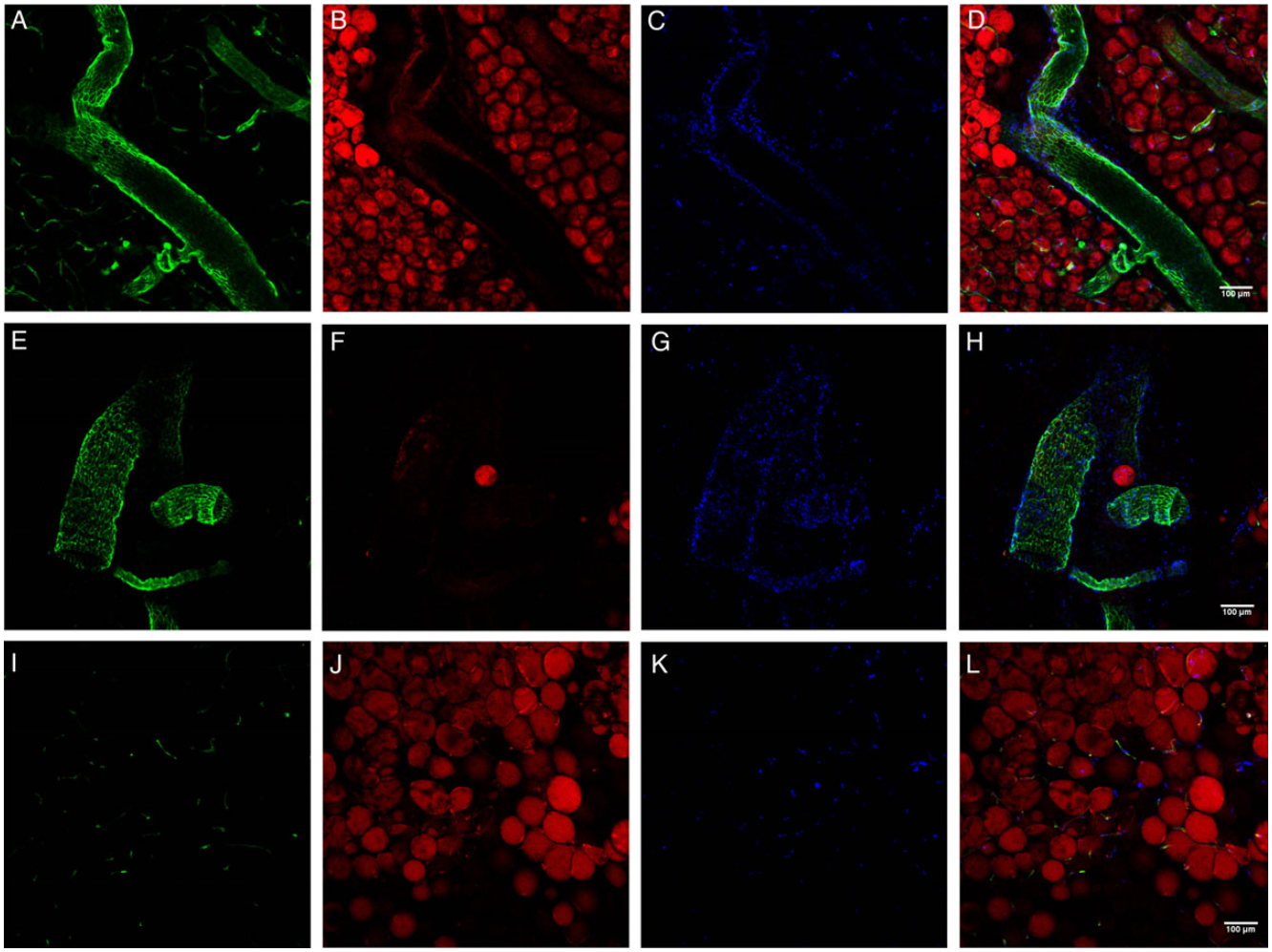

Figure 5. Confocal microscopy analysis. (A-D) En bloc AT (without liposuction): (A) CD31 A488 staining (endothelial cells, green); (B) Bodipy 558/568 staining (adipocytes, red); (C) DRACQ5 staining (nuclei, blue); (D) Merged image. (E-H) Residual AT after liposuction: (E) CD31 A488 staining; (F) Bodipy 558/568 staining; (G) DRACQ5 staining; (H) Merged image. (I-L) AT from liposuction: (I) CD31 A488 staining; (J) Bodipy 558/568 staining; (K) DRACQ5 staining; (L) Merged image.

patients studied for comparison between these two AT groups. Furthermore, our flow cytometry gating strategy was representative of the endothelial cell rate. Indeed, the CD31 adhesion molecule, also known as PECAM-1, was expressed in large amounts on endothelial cells, as well as on hematopoietic cells (excluded using the CD45 marker) with $CD45^{neg}CD31^{pos}$ platelets. Previous data from our group demonstrated that enzymatic digestion did not remove platelets and hematopoietic cells. Therefore, in this study, following digestion and staining, we performed washing in ammonium chloride lysis buffer to remove red blood cell contaminants, as well as cell centrifugation at 600 g to pellet down all cells, including small ones, such as platelets. Finally, since a previous study demonstrated that platelets were not lysed by ammonium chloride,[13] platelets were removed from analysis, based on their size (2-4 µm vs endothelial cells > 20 µm), via adjustment of the cytometer threshold on the FSC/SSC plot.

Confocal microscopy analysis confirmed the flow cytometry data and added important morphological information. To our knowledge, this is the first report that, after liposuction, a large number of large microvessels (diameter > 80 µm) and intermediate microvessels remained present in the tissue, while displaying a normal morphology. This is important information; if the remaining vessels were damaged by the liposuction procedure, their function in blood-tissue exchange would be impaired. Therefore, our data suggest that liposuction partly preserves microvessels and probably part of the microvasculature also. Further research is needed to confirm these findings.

Liposuction does not damage perforator vessels larger than 1 mm. Salgarello et al[5] used Doppler sonography to

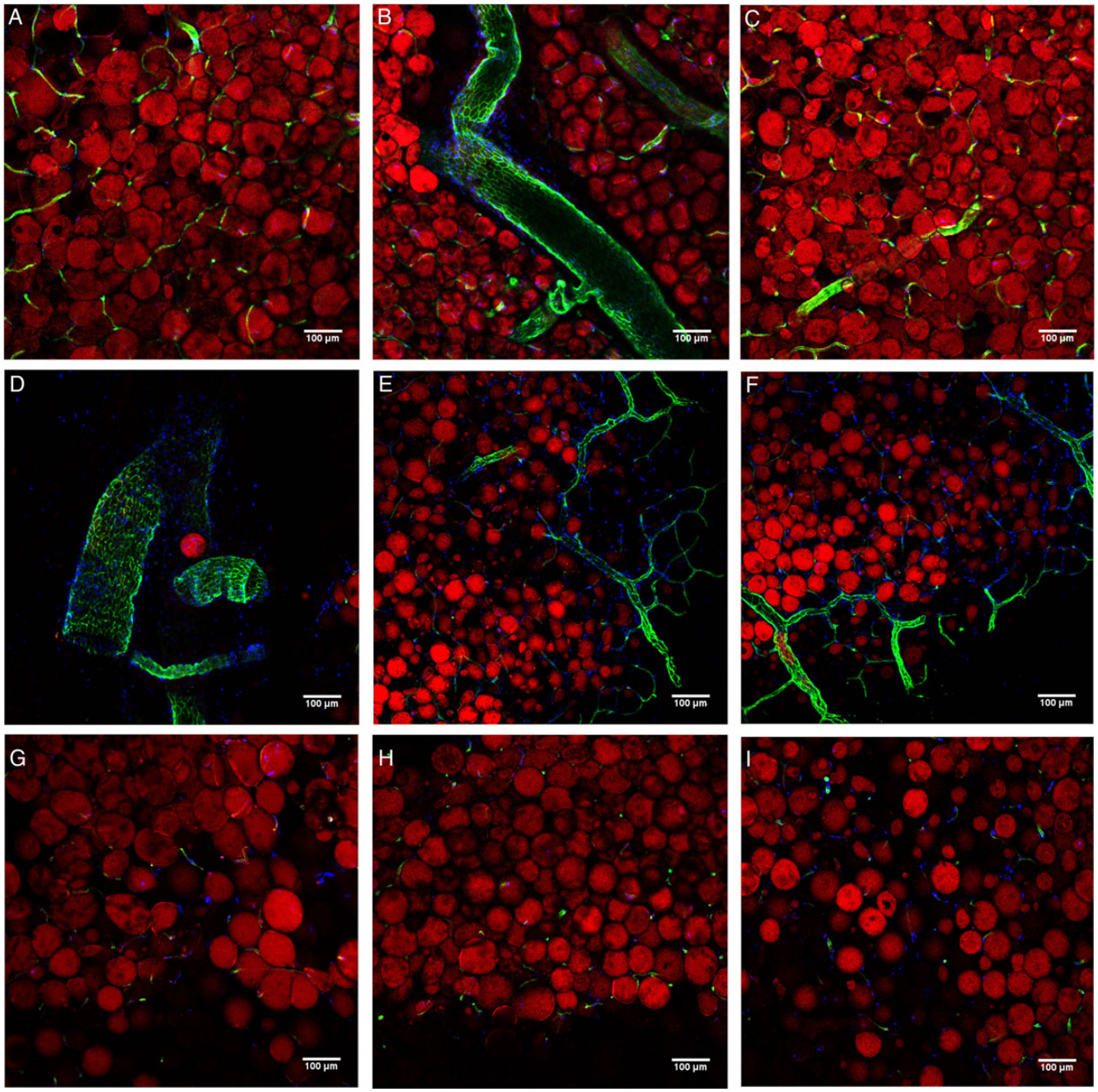

Figure 6. Confocal microscopy analysis. (A-C) En bloc AT (without liposuction). (D-F) Residual AT after liposuction. (G-I) AT from liposuction. Adipose tissues were stained with CD31 (endothelial cells, green), bodipy (adipocytes, red), and DRAQ5 (nuclei, blue). This figure shows three examples for each group with the three colors merged.

evaluate the feasibility of breast reconstruction with a Deep Inferior Epigastric Perforator flap in the aftermath of a liposuction procedure; by studying the inferior epigastric perforator vessels after liposuction on six patients, the authors found the same number of vessels before and 6 months after the operation, with equivalent average diameters and blood flow. Similar results were found in a study that evaluated abdominal perforators by Doppler flowmetry analysis after lipoabdominoplasty on 20 patients before surgery and 15 days postsurgery.[4] These studies showed the preservation of perforator vessels that function mainly to enable blood transport. Our study brings important new insights showing that, after such surgery, liposuction not only preserves large vessels, but also microvessels. These

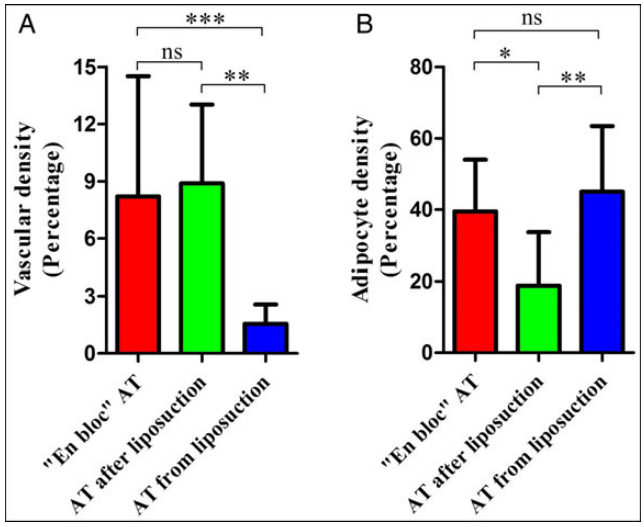

Figure 7. Confocal microscopy results. (A) Vascular density on en bloc AT, AT after liposuction, and AT from liposuction. (B) Adipocyte density. $^{*}P < .05$, $^{**}P < .01$, $^{***}P < .001$.

results are particularly interesting because exchanges between blood and tissue occur in whole organs at the level of the microvasculature.

Our study demonstrating the partial preservation of microvessels reinforces the clinical interest of liposuction with possible better preservation of the physiology of remaining tissues that can retain fluid, gaseous, nutrient, and waste exchanges.[8,9] This could explain why, after undermining with liposuction, some surgeons make the closure without drains in the absence of further noticeable complications.[14] One of the major problems of body contouring surgery after weight loss is the seroma, which is defined as a liquid collection with exudative characteristics formed in a dead space (eg, under the abdominal flap in abdominoplasty). Seroma formation is a common serious problem for patients and surgeons. The reasons for seroma formation are still not clear. Assumptions proposed in the literature include cutting and disrupting a large number of lymphatic and blood vessels,[15] release of inflammatory mediators from traumatized tissues,[16] occurrence of dead space after a large undermining,[17] and shearing forces between the flap and aponeurosis.[17] To resolve this problem after an abdominoplasty procedure, surgeons have developed different techniques, such as the use of quilting sutures to reduce the dead space[18-21] or the use of a mini-abdominoplasty technique to minimize the undermining.[22,23] Only patients with moderate excess below the umbilicus can benefit from this technique (Type 2, according to the classification of Matarasso).[24] One of the most important advances is the use of liposuction combined with abdominoplasty, termed lipoabdominoplasty. This procedure includes the use of superficial and deep liposuction under the resection area associated with resection of the skin only (epidermis and dermis).[25] The advantage of this technique is the preservation of lymphatic[26] and blood vessels, as well as connective tissues. Saldanha et al[25] reported a rate of 0.4% of seroma in a series of 445 patients operated by lipoabdominoplasty. In view of our findings, it is likely that this extremely low rate of seroma was possibly due to, at least in part, the preservation of the microvascular network. Overall, these results certainly help to further our knowledge on liposuction and could be particularly useful in the field of abdominoplasty where such an approach remains debated, since most plastic surgeons are currently using liposuction combined with medial thighplasty and brachioplasty to obtain satisfactory outcomes (with very low rates of seroma).[27,28]

This study has several limitations. First, enzymatic digestion remains a limiting factor which can bias the results. In this study, the areas analyzed were very small, compared to the surgical site. Vessel density was higher in AT after liposuction compared to en bloc AT, with no statistically significant difference ($P = .111$). However, we believe that, if whole tissue after abdominoplasty could be analyzed in situ, it is likely this vessel density would be lower. Indeed, liposuction inevitably removes elements of the microvascular network, and only a study of the entire abdominal wall would provide a reliable quantification. Moreover, in our study, we did not assess the functionality of the remaining vessels. Finally, no power analysis was performed to determine the minimum number of patients required, and therefore a study with a greater number of patients will be useful to confirm our findings. Therefore, further work is needed to determine and understand any associations between the integrity of the microvascular network and its function. For example, it would be interesting to compare the preservation and functionality of the remaining tissues with the occurrence of postoperative complications such as seroma, skin necrosis, and wound dehiscence.

CONCLUSIONS

Demonstrating that the microvascular network is partially preserved after liposuction would prove useful to plastic surgeons, with a view to encouraging the use of liposuction in body contouring procedures. With the increasing prevalence of obesity worldwide, the number of patients undergoing body contouring surgery after weight loss is increasing steadily. The goal of surgical management is to optimize the functional results obtained from bariatric surgery or diet with the removal of redundant skin folds. Within this field, abdominoplasty, medial thighplasty, and brachioplasty procedures are often combined with liposuction to reduce flap thickness and increase tissue mobilization. Liposuction has the advantage of preserving connective tissues which contain nerves, and lymphatic and blood vessels. Our study shows that an

important part of the connective tissue microvascular network is preserved by liposuction.

Acknowledgement

The authors thank Dr. Alexia Zakaroff-Girard for his support in flow cytometry analysis at the Cytometry and Cell Sorting Platform (INSERM/UPS-UMR1048) of Rangueil Hospital in Toulouse, and Dr Sonia Severin for her knowledge in platelets biology.

Disclosures

The authors declared no potential conflicts of interest with respect to the research, authorship, and publication of this article.

Funding

This work was supported by grants from APR 2012-2014, EFS, Région Midi-Pyrénées grant n°11011520, the European Center for Transplantation Sciences and Immunotherapy (IHU CESTI, ANR-10-IBHU-0005), and the Infrastructure Program ECELLFRANCE (ANR-11-INSB-005). The salary of the first author (NB) was provided by a Médaille d'or from Rennes University Hospital for one year. The funding was used for all consumables that are necessary to the achievement of experiences (reagent, antibodies), for technical platform facilities (cytometry, microscopy), and medical writing and editing assistance.

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