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SPECIAL TOPIC
Breast

Reproducible Volume Restoration and Efficient Long-term Volume Retention after Point-of-care Standardized Cell-enhanced Fat Grafting in Breast Surgery

Severiano Dos Anjos, PhD*
Aina Matas-Palau, BS*
Josep Mercader, PhD‡
Adam J. Katz, MD, FACS†
Ramon Llull, MD, PhD*†

Background: Lipoaspirated fat grafts are used to reconstruct volume defects in breast surgery. Although intraoperative treatment decisions are influenced by volume changes observed immediately after grafting, clinical effect and patient satisfaction are dependent on volume retention over time. The study objectives were to determine how immediate breast volume changes correlate to implanted graft volumes, to understand long-term adipose graft volume changes, and to study the "dose" effect of adding autologous stromal vascular fraction (SVF) cells to fat grafts on long-term volume retention.
Methods: A total of 74 patients underwent 77 cell-enhanced fat grafting procedures to restore breast volume deficits associated with cosmetic and reconstructive indications. Although all procedures used standardized fat grafts, 21 of the fat grafts were enriched with a low dose of SVF cells and 56 were enriched with a high SVF cell dose. Three-dimensional imaging was used to quantify volume retention over time
Results: For each milliliter of injected fat graft, immediate changes in breast volume were shown to be lower than the actual volume implanted for all methods and clinical indications treated. Long-term breast volume changes stabilize by 90–120 days after grafting. Final volume retention in the long-term was higher with high cell-enhanced fat grafts.
Conclusions: Intraoperative immediate breast volume changes do not correspond with implanted fat graft volumes. In the early postoperative period (7–21 days), breast volume increases more than the implanted volume and then rapidly decreases in the subsequent 30–60 days. High-dose cell-enhanced fat grafts decrease early postsurgical breast edema and significantly improve long-term volume retention. *(Plast Reconstr Surg Glob Open 2015;3:e547; doi: 10.1097/GOX.0000000000000511; Published online 23 October 2015.)*

*From the *Stem Center, Palma de Mallorca, Spain; †University of Florida, Gainesville, Fla.; and ‡University of the Balearic Isles, Alimentómica, Palma de Mallorca, Spain.*
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Minimally invasive fat grafting represents an ideal therapeutic strategy for treating congenital, cosmetic, traumatic, or postoncological volume deficits. This approach is of particular interest for both reconstructive and cosmetic breast deformities, and has recently garnered significant

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attention from researchers, patients, clinicians, and professional societies alike. Published reports and specific reviews on lipoaspirated grafting for volume restoration present inconsistent results and disclose conflicting data, reflecting continued challenges of a procedure that is seemingly simple yet remains largely unpredictable and inefficient.[1]

The technique's unpredictability is apparent in the literature as the surgical technique itself is subjected to significant variability in graft harvesting, processing, and delivery methods. Modifications of these various steps can alter the relative percentages and quality of tissue, as well as aqueous and oil fractions within the lipoaspirated fat graft—with both known and unknown consequences. Even those reports that purport to rigorously standardize fat graft processing often lack descriptions of basic parameters such as the implanted graft's physical composition (such as relative aqueous, oil, and tissue fractions; eg, the "dry" tissue/graft volume), and the implanted graft's physiological characteristics after processing and before implantation, such as osmolarity and pH.

Certainly tissue, whether dead or alive, will restore volume, but only live and surviving tissue will optimally retain volume in the long term. A nonviable or apoptotic fat graft will transiently restore a volume deficit, yet will ultimately lose most of its volume and consequently yield a clinically disappointing result. Volume restoration obtained at the expense of necrotic and inflammatory tissues will not safely reproduce the biological properties of the recipient subcutaneous tissues and may require multiple grafting (or other) procedures to mend complications and satisfy both patient and surgeon.

Therefore, the physical and physiological fat graft conditions are fundamental to achieve consistency (reproducibility) and high volume retention (efficacy) in fat grafting procedures. Although graft composition and viability are responsible for volume restoration and retention, volume maintenance does not necessarily imply graft survival. However, because volume retention is the closest to the clinical goal and can be precisely quantified using high-definition 3-dimensional (3D) imaging, one can consider volume retention as an indirect indicator of fat graft viability.

In an effort to potentiate the engraftment process and its associated volume retention, Moseley et al[2] and Tholpady et al[3] have long pursued a cell-based strategy: it is our main hypothesis that addition to a particle fat graft of an autologous mesenchymal cell population, which is constitutively responsible for the repair (fibroblasts),[4] vascular supply (endothelial and perivascular elements),[5] and immunomodulatory control (mononuclear immune-competent cells)[6,7] of its supporting parenchyma, would contribute to fat graft survival and hence increase long-term volume retention clinically.

Although intraoperative treatment decisions are influenced in real time by volume changes observed immediately during and after grafting, clinical effect and patient satisfaction are ultimately dependent on volume retention over time. The objectives of this study were to determine how intraoperative and immediate postoperative volume changes in the breast correlate to implanted graft volumes, to understand how long-term graft volume retention relates to early postgraft volume changes, and to study the "dose" effect of adding autologous adipose-derived stromal vascular cells to fat grafts on both early volume changes and long-term volume retention.

MATERIALS AND METHODS

The Ethics Committee of the Balearic Isles authorized the research aspects of the present study. The clinical use of autologous adipose-derived cells was authorized by the regional Health authorities. The fat grafting preparation device (GID-700) is both CE mark and USA 510(K) certified. The cell dissociation device (GID SVF-1) is a CE marked medical device available in Europe. Patients were specifically counseled on the potential limitations and risks of breast augmentation with cell-enhanced fat grafts during the informed consent process (**See Supplemental Digital Content 1**, which details experimental procedures, ***http://links.lww.com/PRSGO/A135***).

Female patients with congenital, cosmetic, or postoncological defects were included in the study. This is a retrospective, nonrandomized observational clinical study.

Surgical Procedure

All cases were performed under general anesthesia. All surgeries and patients were performed by the same plastic surgeon, the same surgical team, and at the same hospital setting. All patients underwent cell-enhanced fat grafting implanted within the subcutaneous, prepectoral, and/or intramuscular[1] planes. A modified Klein's tumescent solution Klein (modifications include Ringer's Lactate, no lidocaine) was infiltrated in a volume equal to the estimated procurement volume. Patients underwent conventional power-assisted (Microaire) liposuction using a cannula (PAL-404LS) connected to a conventional vacuum pump at an average pressure of 53.3 kPa (0.52 atm).

The initial harvest of raw lipoaspirate was consistently aspirated from the infraumbilical area and flanks, and collected in the GID SVF-1 device for stromal vascular fraction (SVF) isolation. Adipose tissue intended as the graft material was collected and processed in the GID-700 device following manufacturer instructions. Before the addition of SVF cells, harvested adipose tissue intended to serve as the graft "foundation" was analyzed for physical and physiological parameters using methods previously reported (See Table 1; **See Supplemental Digital Content 2**, which displays fat graft physical and physiological parameters, ***http://links.lww.com/PRSGO/A136*** and **See Supplemental Digital Content 3**, which displays methodology to quantify fat graft standard parameters, ***http://links.lww.com/PRSGO/A137***).

Cell-enriched Fat Graft Preparation

The GID SVF-1 device containing adipose tissue was moved to a side table in the same operating room for subsequent tissue processing. SVF isolation was carried out using the GID SVF-1 as described previously.[8]

Freshly isolated and resuspended adipose SVF cells were combined with washed fat graft (in catheter tip 50-mL syringes) using a catheter tip to Luer-Lock adapter. For every 50 mL of processed fat graft, 1–2 mL of resuspended SVF cells were mixed in using gentle back and forth transfer between the 2 syringes.

Implantation

Cell-enhanced fat grafts were transferred to 20-mL Luer-Lock syringes. Two 2-mm skin incisions located at medial and lateral ends of the inframammary fold were used for introduction of injection cannulas and "cross-hatched" grafting passes on different planes. For fat graft implantation, 20-mL Luer-Lock syringes connected to spoon-tip, 20-mm-long SuperLuerLock injection cannulas of 2.1-mm diameter (Tulip Company) were used. Long passes were executed laying no more than 2 mL of cell-enhanced fat graft per pass. Grafting was terminated early in only 7 occasions (3 implant coverage, 3 implant replacement, and 1 aplasia), when tissue pressure exceeded the implantation pressure generated by normal manipulation of the fat graft syringes, manifest as graft "reflow" (ie, immediate graft extrusion from access sites).

Breast Volume Measurement (3D Imaging)

To evaluate clinical effect, a 3D imaging scan was utilized to quantify breast volume changes. The 3D digital breast surface images (or meshes) were obtained using an ARTEC MHT 3D Scanner and then superposed to measure the volume difference, using the manufacturer's provided software (Fig. 1). In all cases, a 3D scan was obtained in decubitus position pre- and postgrafting. In 3 hypoplasia cases and 3 post-LD volume deficit cases, sequential scans were taken immediately after delivery of incremental aliquots of graft. 3D contour profile precision and accuracy was validated with magnetic resonance imaging (MRI).

Graphs and Statistical Analysis

All graphs, plots, and statistical analyses were carried out using GraphPad Prism 5.0 Software (GraphPad, San Diego, Calif.). Error bars represent standard deviation (SD) and not standard error of the mean.

RESULTS

Patient Demographics and Graft Preparation

Patient demographics and graft properties are summarized in Table 1. Patients were all female and ranged from 18–61 years of age. Patients with congenital and/or cosmetic indications were between 18 and 45 years old, whereas postoncological reconstructive patients were between 38 and 61 years of age. Patient age and BMI were similar between low- and high cell-enhanced patients. All patients maintained a stable BMI throughout the follow-up period (data not shown). High cell-enhanced grafts had approximately a 10-fold higher number of SVF cells per cm^3 of fat graft (on average, 435,918 cells/cm^3 of graft) than low cell-enhanced grafts/patients. The total surgical time for these cases ranged from 3

Table 1. Patient Population Data (*n* = 77)

	n	BMI (kg/m²)	Age (y)	Total Nucleated Cells per cm^3 of Fat Graft	Fat Graft Volume Injected per Breast (mL)	Digested Dry Lipoaspirate Volume for SVF Isolation (mL)
Low cell enhancement (Mean ± SD)	21	21.59 2.09	37.80 18.89	42,528 12,370	229.09 63.42	21.73 8.24
High cell enhancement (Mean ± SD)	56	21.57 1.75	39.36 12.83	435,918 284,921	270.74 55.60	253.09 75.29

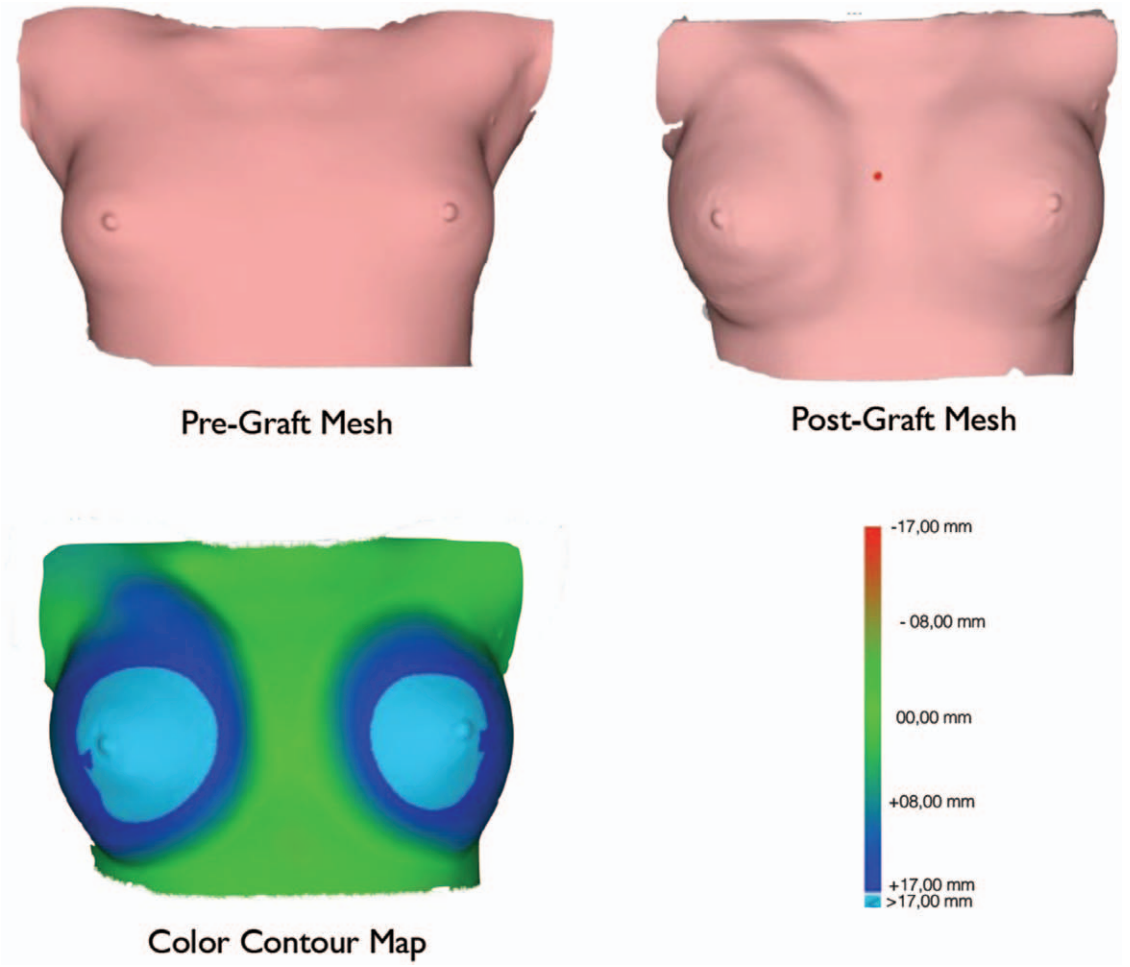

Fig. 1. Methodology to obtain intraoperative (A) pregraft and (B) postgraft mesh, superpose both to generate a color contour map (C) and quantification of volume changes using gated point differences (ie, light blue color isometric contour on postgraft mesh indicates distance differences with respect to equivalent pregraft mesh greater than 17 mm); (D) Map color scale (mm).

to 4.5 hours and proceeded as described in Figure 2 (Mean cell processing time was 60–70 minutes). Fat grafting proceeded intraoperatively without complications. However, a reflow point was reached in 7 of the 77 procedures. All of these cases involved "coverage" of an existing silicone implant.

Fat Graft Standardization

The process for fat graft preparation using the GID-700 device (now marketed as Revolve, LifeCell) has been previously described (**See Supplemental Digital Content 2**, which displays fat graft physical and physiological parameters, ***http://links.lww.com/PRSGO/A136***).[9] The current work further validates these previous findings that the method renders consistent fat graft tissue with low lipocrit and aqueous fractions (**See Supplemental Digital Content 2**, which displays fat graft physical and physiological parameters, ***http://links.lww.com/PRSGO/A136***, $n = 5$). Our samples also confirmed restoration of adipose graft osmolarity, virtual elimination of free triglycerides (oil) and lactate dehydrogenase, and hematocrit within normal physiological levels.

SVF Cell Suspension

The mean viable cell yield values obtained using the GID technology was also within previously published range[8] at around 600,000 nucleated SVF cells per cm^3 of dry adipose tissue (60 million nucleated cells per 100 cm^3 of adipose tissue) (Table 2). Table 1 summarizes the average amount of digested tissue and the total nucleated SVF cells utilized to enhance fat grafts. The amount of cell enhancement was utilized to categorize procedures as low (<50,000 SVF cells/cm^3 graft) versus high cell enhancement (>200,000 SVF cells/cm^3 graft). The mean cell viability was 83% of the total nucleated cell population isolated.

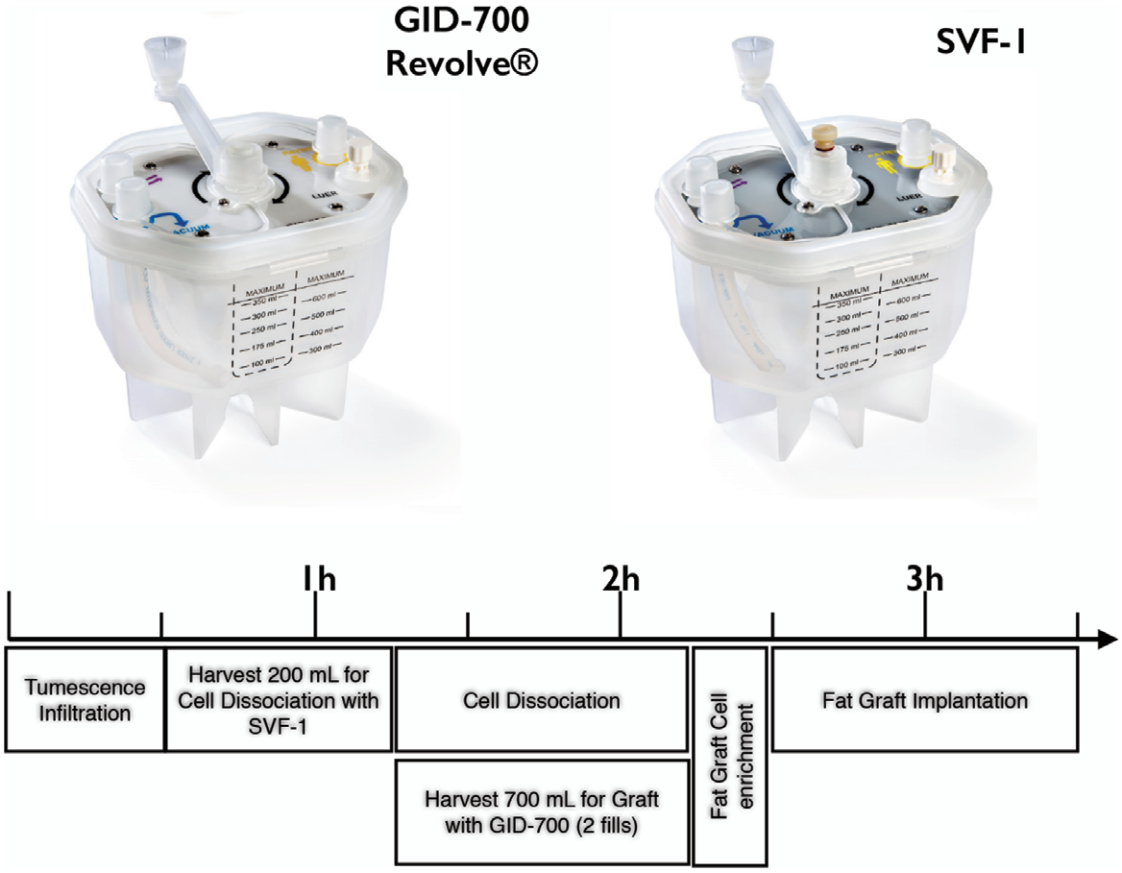

Fig. 2. Fat graft processing device (GID700) and SVF cell isolation device (GID SVF-1). Both devices are connected in line between the surgeon's cannula and the waste canister. The GID SVF-1 is filled first during liposuction and handed to the technician; then the GID-700 is filled with lipoaspirated particle fat graft. General overview of entire procedure is depicted at the bottom.

Mammometrics Methodology

3D imaging measurements of implanted silicone implants were compared to MRI measurements as a means to validate the use of 3D imaging for quantifying changes in breast volume (**See Supplemental Digital Content 4**, which displays comparison between 3D scans and MRI volume calculations, ***http://links.lww.com/PRSGO/A138*** and **See Supplemental Digital Content 5**, which demonstrates trueness and validation of mammometrics methodology, ***http://links.lww.com/PRSGO/A139***). Silicone implants were chosen as relevant controls because they do not change volume once implanted in the breast tissue. As shown in **Supplemental Digital Content 4** (which displays comparison between 3D scans and MRI volume calculations, ***http://links.lww.com/PRSGO/A138***) and **Supplemental Digital Content 5** (which demonstrates trueness and validation of mammometrics methodology, ***http://links.lww.com/PRSGO/A139***), 3D-scan color contour maps rendered precise values (±2%) with respect to known ex vivo silicone implant volumes as compared to blinded measurements of similar implants using MRI.

Table 2. Cell Quality and Safety Analysis of SVF Cell Suspension (*n* = 52)

Cell Quality and Safety Analysis	Average	Units	SD
Nucleated viable cells per cm^3 of strained, washed, dried adipose tissue (not raw lipoaspirate)	5.83×10^5	cells/mL	$\pm2.88\times10^5$
Cell viability	82.79	(%)	±8.14
Proliferation index (resting cells)	94.92	(%)	±1.69
Apoptotic index	26.95	(% DAPI positive cells)	±9.54
Endotoxin levels*	1.43	(EU/mL)	±1.22
Culture in agar chocolate CFU after 72 hours	0 (none)	colonies	NA

Table 3. Intraoperative Volume Restoration Index (IVRI) Data

		No. of Procedures		Total, *n*	Graft Volume				IVRI					
		Cell Enhancement			Left Breast		Right Breast		Left Breast		Right Breast		Both	
Field	Indication	Low (*n* = 21)	High (*n* = 56)	(*n* = 77)	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Congenital	Aplasia (unilateral)	1	2	3	190.00	28.28			0.91	0.03			0.91	0.03
	Breast tuberous deformity	1	2	3	195.00	7.07	195.00	7.07	0.92	0.03	0.93	0.02	0.92	0.02
Cosmetic	Hypoplasia	5	28	33	288.00	59.58	262.81	61.83	0.82	0.26	0.91	0.22	0.89	0.24
	Silicone implant coverage	3	5	8	203.33	105.04	216.67	86.22	0.84	0.02	0.87	0.01	0.85	0.02
	Silicone implant conversion	3	7	10	283.33	28.43	296.67	20.21	0.80	0.12	0.71	0.27	0.76	0.20
Reconstructive	Post-TRAM upper pole deficiency	5	6	11	181.67	31.75	195.00	18.03	0.84	0.06	0.99	0.20	0.92	0.15
	Post-LD muscle flap volume deficiency	3	6	9	200.00	0.00	232.50	24.75	0.63	0.00	0.64	0.07	0.64	0.05
Reference	Silicone implant alone (no fat graft)	NA	NA	5			270.00	75.08			0.84	0.03		

Complications

Complications included a self-resolved Mondor's disease and 3 revisions due to asymmetry or under-augmentation (all of which were low cell-enhanced procedures). During our follow-up, 9 of 74 cases complained of transient palpable subcutaneous lumps, mostly evident 6 months or later postoperatively. After sonographic diagnosis of oil cysts and radiographic confirmation, all palpable cysts were subjected to external manual pressure during examination: 6 of them ruptured during such manipulation and left a discrete discomfort for 4–6 days, but then resolved. The 3 remaining cysts were referred to radiology, and fine-needle aspiration disclosed fat necrosis. When surveyed at long-term follow-up (>1 year), all patients were subjected to sonographic survey using a Sonosite M turbo ultrasound system. The presence of oil cysts as defined as round, subcutaneous, anechoic, homogeneous cysts increased to 14 of 74 cases. No complication type was statistically different between low- and high cell-enhancement groups.

Intraoperative and Postoperative Volume Changes in Grafted Breasts: Graft versus Recipient Site Relationship

Intraoperative Volume Restoration Index (IVRI) of cell-enhanced fat grafts (Table 3)

Changes in breast volume were measured immediately postoperatively using 3D scanning methods in the context of a variety of clinical cases. As shown in Figure 3, the measured change in volume never equaled the actual volume of graft placed, regardless of the clinical condition being treated (as reflected in an IVRI of <1). Although the majority of clinical indications treated in our study correlated to an IVRI of 0.85–0.92, at least 2 conditions were associated with lower values (silicone implant conversion,

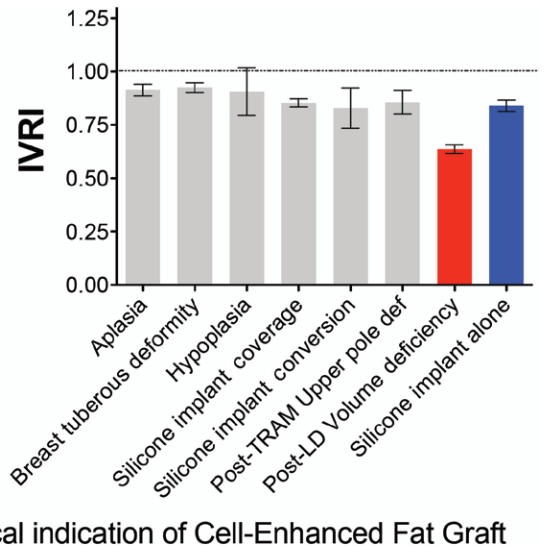

Fig. 3. IVRI across different cell-enhanced fat graft procedures. When a continuous linear elastic body such as silicone implant (blue column) is introduced into a nonlinear, viscoelastic breast tissue content, the breast volume thus generated does not reach the anticipated total volume resulting from the arithmetic addition of preoperative volume plus the implant volume (theoretical IVRI of 1). The response is different when the breast tissue continent is significantly loose, such as in the empty envelope generated following immediate skin-sparing mastectomy and LD-vascular bed preconditioning (* *P* value < 0.001).

$n = 10$; and post-LD muscle flap volume deficiency, $n = 9$). To explore this phenomenon further, traditional silicone implant cases (without fat grafting, $n = 5$) were studied as a control group. One might expect to find an IVRI of 1 for a silicone implant, meaning 1 cm^3 of implanted volume yields 1 cm^3 of breast volume expansion. However, IVRI for silicone implants (control group, blue bar, Fig. 4) reached only 0.84. IVRI values for different cell-enhanced fat grafting procedures are shown in Figure 3.

The nonlinear and yet variable IVRI measurements—relative to both clinical application and escalating graft volume—is readily apparent in Figure 5A. It shows intraoperative changes in IVRI associated with incremental graft volumes in a highly compliant post-LD volume deficit breast content. Figure 5B shows the different compliance behavior between post-LD volume deficit and hypoplasia patients as a function of grafted volume.

Postoperative Volume Retention Index of Standardized Cell-enhanced Fat Grafts

Breast volume changes were measured from 7 days postoperatively out to 540 days. The results demonstrate that there was no volume loss during the first 2–3 weeks after grafting. In fact, there was an increase in breast volume during the first few weeks [as reflected in an increased postoperative volume retention index (POVRI)]. Interestingly, this initial increase in volume is higher for low cell-enhanced fat grafts compared to high cell-enhanced grafts (Fig. 4). After 2–3 weeks, breast volume gradually decreases over time (relative to immediate postoperative volume), as reflected in the POVRI. As shown in Figure 4A, the POVRI stabilizes at around 90–100 days after the surgery (3 months) and throughout 1.5 years of follow-up for both low- and high cell-enhanced procedures.

There are significant differences between those adipose tissue grafts prepared with low- (≤50,000) and high (≥200,000) SVF cell enrichment. As mentioned, the volume increase noted in postoperative weeks 1–3 is notably lower in those fat grafts enriched with a high dose of SVF cells compared to those enriched with low dose of SVF cells (Fig. 4). Over time, however, the long-term plateau of the POVRI is much higher when using high cell-enriched fat grafts versus low cell-enriched fat grafts.

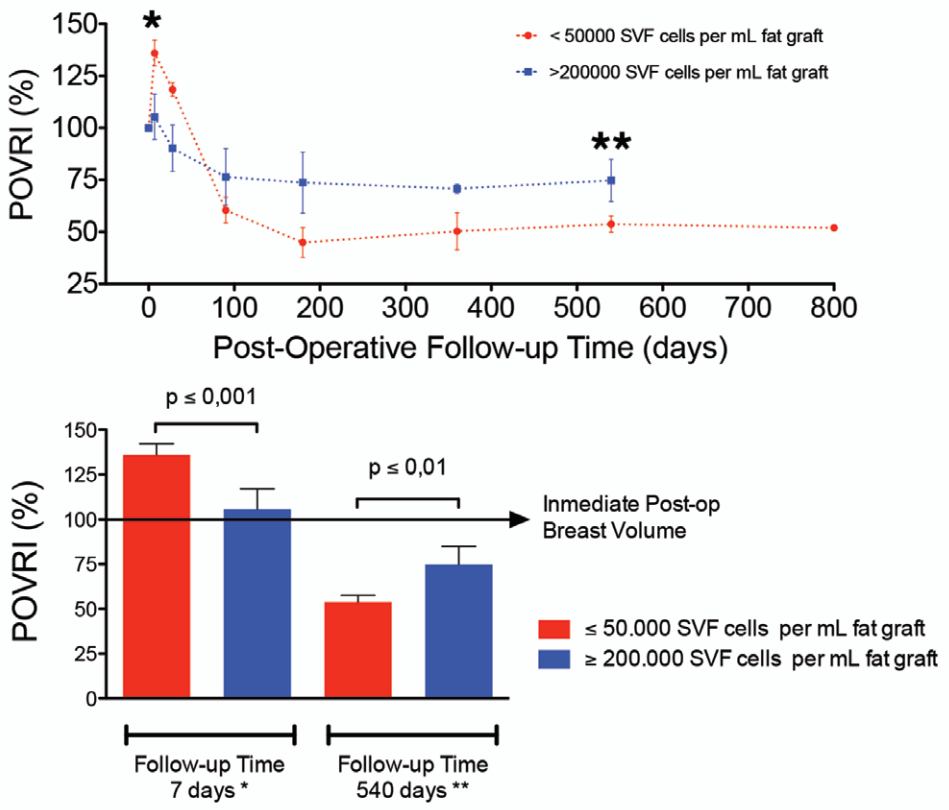

Fig. 4. Postoperative volume retention index (POVRI). A, POVRI changes over time for high and low cell-enhanced fat grafts. B, POVRI statistical analysis of low- versus high cell-enhancement fat grafting procedures at 7 days and 540 days follow-up ($n = 11$ for low cell-enhanced fat grafts and $n = 38$ for high cell-enhanced fat grafts).

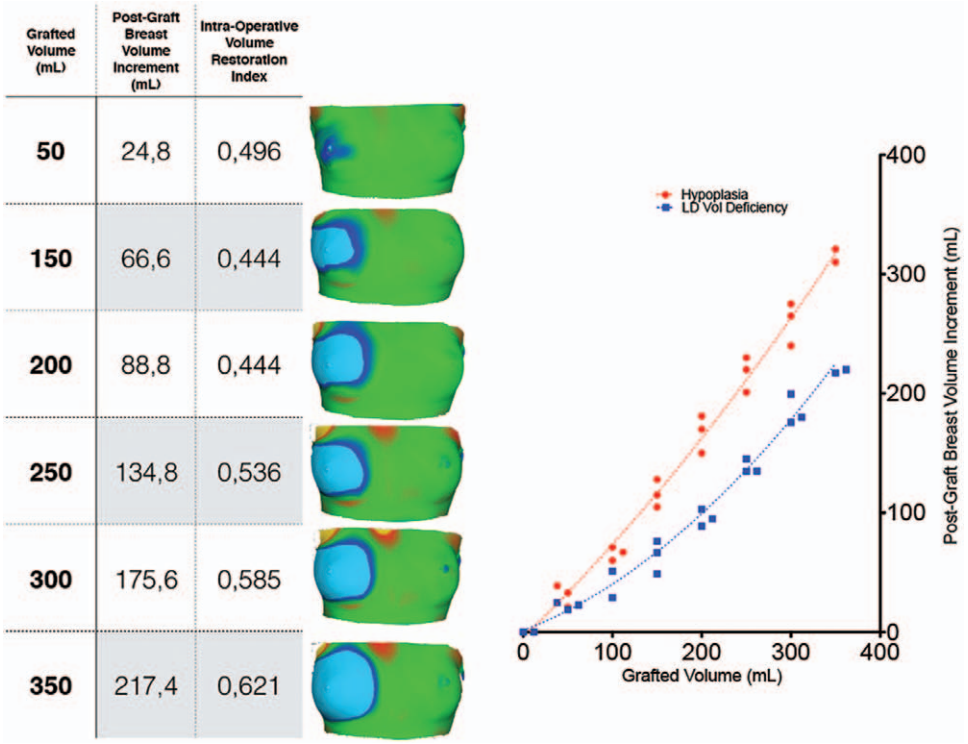

Fig. 5. Postgraft breast volume increments resulting from progressive graft lipoaugmentation. A, 3D scans sequentially taken during a fat grafting procedure to restore a post-LD volume deficit. The fat graft IVRI is determined after implantation of 50-mL fat graft (notice the resulting interim color contour maps). Initial small graft volumes (50–200 mL) result in a low restoration capacity (IVRI of 0.49–0.44). As contents grow within the continent, the IVRI rises to a moderate index (0.61). B, The IVRI is represented here as the slope (IVRI = postgraft breast volume increment/graft volume) or tangent of the regression lines depicting the differences in sequential IVRI measurements during 3 cases of fat graft augmentation for hypoplasia versus 3 cases of fat graft reconstruction of post-LD volume deficit (Fig. 7).

The mean volume retention index for high cell-enriched fat grafts observed after 1.5 years is 75%, whereas breasts implanted with adipose tissue grafts containing low cell enhancement maintained only 50% of the initial postoperative volume 1.5 years after the surgery. This difference in long-term volume retention (25%) was statistically significant between these 2 groups 1.5 years after the surgery as shown in Figure 4B. Figures 6 and 7 are illustrative examples of high cell-enriched fat grafting cases with clinical photos, corresponding 3D maps and quantitative POVRI changes over an 18-month follow-up.

DISCUSSION

Volume loss after autologous fat grafting is a well-documented event described by Delay et al.[1] Different techniques for fat graft preparation have been previously reported. However, the success of volume retention in fat grafting is still limited due to several factors, such us harvesting and reinjection technique or recipient site vascularity.[10]

Our data support the use of 3D scanning as a sensitive and practical method for quantitating breast volume over time. As MRI is considered by some to be a gold standard technique for quantitating breast volume,[11] we have validated our methods relative to MRI evaluation of known volumes (ie, silicone implants both ex vivo and in vivo). Compared to MRI, however, 3D scanning may be cheaper and perhaps more available and practical to the average clinician.

The fact that both fat grafts and silicone implants do not restore volume defects on a one-to-one basis was a surprising and new finding. This is reflected in our data as IVRI < 1.One could hypothesize that this phenomena is likely explained by the viscoelastic behavior or compliance of the recipient breast tissue components, its response to the implantation of an elastic body (implant), and the fat graft composition itself. Although it is logical that graft volume will be predictably lost by absorption/dissipation of its water (and oil) phase, it is not clear how quickly this occurs. This speaks, however, to the importance of having a standardized, reproducible fat graft with known physical composition.

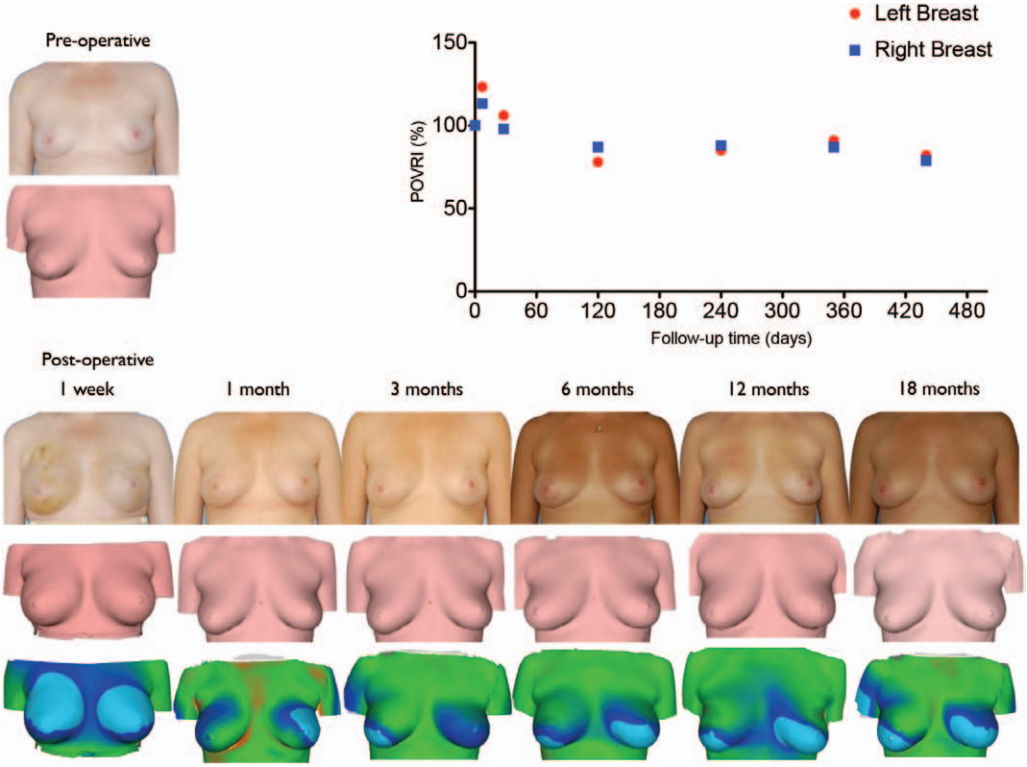

Fig. 6. An 18-month follow-up on a representative asymmetric cell-enhanced lipoaugmentation. A, Preoperative photograph and the corresponding 3D-scan mesh. Left breast had smaller volume (120 mL). Correction was attempted by grafting 225 mL to the right and 375 mL. Cell enhancement at 520,000-SVF cells/mL fat graft. B, Each photograph (upper row) finds the corresponding 3D-scan mesh at a given postoperative time (middle row). However, although total volumes remain constant over time, the volume distribution within the breast changes: color contour maps (lower row) show that large volumetric differences (light blue areas) migrate from the upper pole to the lower breast quadrants. C, POVRI(%) is plotted over time demonstrating correction of asymmetry and its stability over time.

For grafts prepared with the methods described, it is 16±3%. Another potential explanation for the finding of IVRI < 1 relates to the concept of packing density, which is further influenced by graft particle shape and compressibility. In short, the IVRI is a quantitative measure of what others have referred to as "graft-to-capacity ratio."[12] To our knowledge, however, these are the first data that objectively illustrate this concept in a systematic, quantitative manner.

Our data also reflect the postoperative volume retention over time of a single fat grafting session using SVF cell-enhanced fat grafts prepared in real time at the point of care. We consider the SVF cell dose used in our low cell-enhancement group as inconsequential, making the low cell-enhancement group functionally equivalent to a "sham" control. In fact, our results using low cell enhancement are similar to those obtained by other authors using fat grafting techniques without any cell enhancement. For instance, a modified Coleman method, when quantified by similar mammometric systems, obtains retention values around 30–50% 5 months postoperatively[13] or 1 year postoperatively.[14]

When using a high SVF dose, our results correlate with those previously published by Kakudo et al[15] using a similar amount of SVF cells per milliliter of fat graft in an animal model (300,000 nucleated cells). Other recent studies have also reported high volume retention levels (80%) after fat grafting using much higher doses of cultured ASCs[16] compared to fat grafts alone (with no cell supplementation). Although cultured autologous adipose-derived stem cells unequivocally help retain fat graft volume in humans, they do so at the expense of a massive cell-enhancement dose (2×10E7 cells/mL graft) only attained after costly and lengthy cell amplification in a GMP facility.

Peltoniemi et al[17] have failed to demonstrate a positive effect of SVF cell enrichment on fat graft survival using cell-assisted lipotransfer. This result may be explained by inefficient isolation with low cell yield and resulting low SVF cell enrichment (ie, dose), and/or

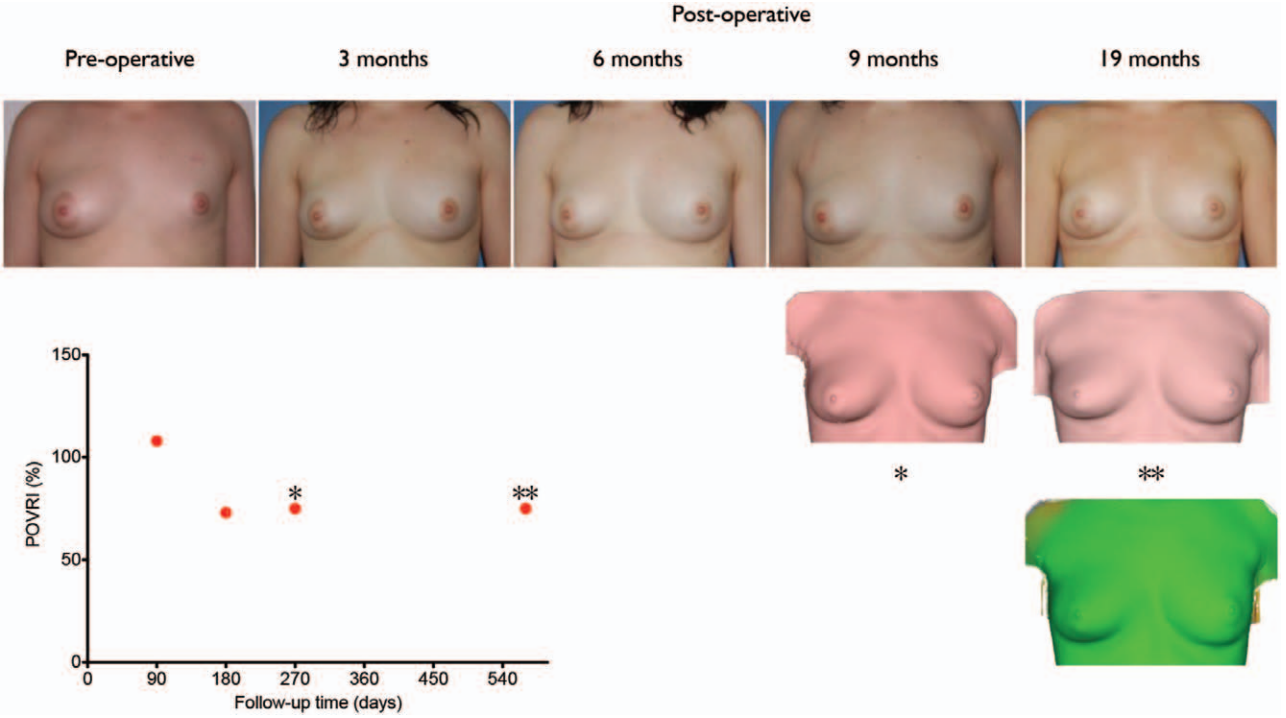

Fig. 7. Congenital left breast aplasia reconstruction with cell-enhanced fat grafting. A, Preoperative condition and photographic follow-up at sequential postoperative times. Postoperative 3D-scan meshes obtained at 9 months (*) and 19 months (**). Color contour profile resulting from superposition of postoperative mesh at 19 months over the mesh at 9 months. Notice lack of changes over a 10 month follow-up interval. B, POVRI over postoperative time, locating long-term follow-ups at 9 months (*) and 19 months (**).

by variable fat graft composition and standardization. The mentioned report does not provide any data in this regard, only the amount of fat used to perform SVF isolation. Because there is a strong variability among patients and age is a very important factor affecting cell yield, we strongly believe that measuring cell yield for each patient is pivotal for quality purposes.

Cell Dose and Anti-inflammatory Effects

Postoperative edema is a well-known effect observed after implant and autologous fat grafting to the breasts.[18] In this study, we theorize that elevated POVRI levels in postoperative weeks 1–3 most likely reflect postoperative swelling. Interestingly, this swelling is clearly less and resolved sooner when using fat grafts supplemented with high doses or more nucleated SVF cells. We speculate that anti-inflammatory properties of MSCs contained within the SVF population might have a role[6]; however, other mechanisms such as early neovascularization potentiated by stromal cells added in combination with host response could be also involved.

Cell Dose and Long-Term Volume Retention

Our data indicate that the volume retention index is constant 3–4 months after the fat transplantation (unless the patient BMI changes significantly), meaning that 3–4 months after surgery, the volume in the breast remains stable. In our study, only patients with slight variations of BMI were analyzed. When using high cell enhancement, long-term POVRI reaches a steady plateau value of 75% at about 3 months. Because we know the fat graft physical composition in this case series, it could be speculated that our aqueous fraction may account for as much as 10–20% of the 25% long-term breast volume loss.

This study supports the idea that the SVF compartment of adipose tissue plays an important role in both adipose tissue survival and graft volume retention over time. Moreover, the effect is dose-related and it appears that a minimal essential number of SVF cells per milliliter of fat seem to be required. At present, this exact minimal dose is not precisely known and, in fact, may differ when different methodologies are used. Our data suggest that at least between 50,000 and 500,000 cells/mL are required when using the methods described.

Several studies have described the positive effect of SVF cells on fat graft tissue survival by increased vascularization and secretion of prosurvival growth factors.[4,19] Similarly, we hypothesize that these factors could explain the improved results found in this study (higher volume retention) when adding a higher number of SVF cells to the fat grafts.

Some of the limitations of this study include the absence of a proper control group without SVF supplementation. However, we believe that due to the great difference in SVF cell dose used, this variable can be statistically analyzed, and hence conclusions about SVF dose on adipose grafting can be drawn based on our data.

Finally, our data objectively demonstrate that the volume of graft administered does not immediately correlate to an increase in recipient site breast volume in a linear fashion. This is true for silicone implants and fat grafts, and underscores the concept of recipient site capacity,[12] which is dependent on the physical properties of the recipient tissue bed and its skin envelope. This last point is important to the field, as it has significant implications for comparing and judging the actual volume maintenance of fat grafts, and emphasizes the need to standardize accurate outcome measures and methods. Moreover, it calls attention to the concept that fat graft efficacy (ie, volume restoration and retention) is highly dependent on the attributes of the recipient site, as well as the many other variables repeatedly highlighted in the literature.

Ramón Llull, MD, PhD
Stem Center SL
USP Clínica Palmaplanas
Camí dels Reis, 308
07010 Palma de Mallorca, Spain
E-mail: ramonllull@mac.com

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