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ORIGINAL ARTICLE

Application of nanofat for treatment of traumatic faecal incontinence after sphincteroplasty – A pilot study

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Abstract

Aim: The aim of this study was to investigate whether the application of nanofat containing stem cells improves continence in women who had previously undergone anal sphincteroplasty with unsatisfactory long-term outcomes.

Method: This prospective pilot study included nine women with various degrees of anal incontinence who had previously undergone anal sphincteroplasty due to obstetric trauma. In all patients, the Wexner Incontinence Score (WS) and Faecal Incontinence Quality of Life Score (FIQLS), as well as anal manometry and endoanal ultrasound measurements, were performed before the procedure and during follow-up. In all patients, liposuction was performed and 50 ml of raw lipoaspirate was obtained and processed using a NanoFat Kit device. Approximately 20 ml of the mechanically emulsified and filtrated fat was obtained and the anal sphincter complex was infiltrated with it. Patient follow-up was conducted in person or via telephone 6 and 12 months after the procedure.

Results: The squeeze pressure was significantly increased 6 months after the procedure ($p = 0.01$). The external anal sphincter measured at the 12 o'clock position was significantly thicker ($p = 0.04$). A significant decrease in the WS was observed both 6 and 12 months after the procedure compared with baseline values ($p < 0.05$ for both).

Conclusion: This study is the first to show that the application of nanofat as an injectable product improves continence in patients with unsatisfactory results after sphincteroplasty, suggesting it to be a promising and effective therapeutic tool. The procedure is safe and can be easily performed as an ambulatory procedure.

KEYWORDS
adipose-derived stromal cells, regenerative cell-based medicine, faecal incontinence, sphincteroplasty, liposuction, nanofat

INTRODUCTION

Faecal incontinence is the inability to control the passage of gas, liquid or solid stool, causing involuntary bowel discharge [1]. It affects up to 11% of the general population and has severe psychological implications and crippling consequences for quality of life [2]. The aetiology of faecal incontinence is diverse, including alterations in bowel motility, stool volume and consistency, rectal sensitivity, neural pathways and anal sphincter function. The most frequent cause is traumatic faecal incontinence due to obstetric or iatrogenic injury. Surgical treatment of traumatic faecal incontinence includes sphincteroplasty, different muscle transposition and reconstruction procedures, sacral nerve stimulation, an artificial bowel sphincter and injection of biomaterials. Sphincteroplasty is the treatment of

choice for patients who are incontinent with an external anal sphincter defect. Shortly after sphincteroplasty, approximately 60%–75% of patients have a satisfactory outcome and 15%–20% have some improvement; however, in the remaining 15%–20% unchanged or worse outcome is reported [3–6]. Long-term outcomes of sphincteroplasty are limited since the number of incontinent patients rapidly increases over time [7–11].

Adipose tissue is known to be a biological agent involved in the repair and regeneration of different tissues [12,13]. Adipose-derived stromal cells (ADSC) have already been used in proctology for the treatments of complex Crohn's perianal fistula [14–16] and anal fissures [17], with satisfactory results.

Adipose tissue is structurally complex, harbouring various cells within its lobulated fibrous septal network. This network can be disintegrated through enzymatic digestion, and the so-called stromal vascular fraction (SVF) may be isolated. Adipose-derived stem/stromal cells are important SVF residents with mesenchymal multipotency. The importance of these cells and their therapeutic potential has been documented [18].

Despite the clinical potential of ADSC, there are some clinical difficulties due to the regulation and costs of cell therapies. That is why different nonenzymatic procedures for obtaining SVF cells, such as the nanofat procedure, are gaining in popularity [19].

The SVF obtained after emulsification of the fat with the nanofat technique, described by Tonnard and Verpaele, contains functional ADSC [20]. ADSC isolated with this technique fulfil the functional criteria and have immunosuppressive properties characterized by the capability to decrease the proliferation of human T cells [21]. Functional ADSC, through secretion of paracrine factors, can modulate inflammation and favour angiogenesis, two factors that play an essential role in tissue regeneration and possibly in muscle regeneration [22].

This study aimed to investigate whether the application of mechanically disaggregated fat, i.e. nanofat, improves continence in women who had previously undergone anal sphincteroplasty with unsatisfactory long-term outcomes.

What does this paper add to the literature?

Maintaining continence after sphincteroplasty performed due to anal sphincter injury remains a significant challenge. This pilot study is the first to show that autologous nanofat transplantation into the anal sphincter improves continence in patients with unsatisfactory long-term results after sphincteroplasty, suggesting it to be a promising therapeutic tool.

METHOD

Study design and clinical protocol

This prospective study was conducted at the Clinic for Digestive Surgery – First Surgical Clinic of the University Clinical Center of Serbia between December 2019 and July 2021.

Recruitment of patients was conducted by telephone using the database on anal sphincter repair procedures and a questionnaire based on the Wexner Incontinence Score (WS) (Table S1). Inclusion criteria were women over 18 years of age with anal incontinence who had previously undergone anal sphincteroplasty due to obstetric trauma. In all included cases, continence function deteriorated over time with no visible defect in the anal sphincter on baseline ultrasound, meaning that repaired sphincter ends were not disrupted. The exclusion criteria were severe comorbidity (American Society of Anesthesiologists score >3), malignant tumour, autoimmune disease, inflammatory bowel disease, subjectively satisfactory continence and incontinent patients with disrupted sphincter ends after sphincteroplasty (visible defect on endoanal ultrasound). Pudendal nerve terminal motor latencies (PNTMLs) were not measured for technical reasons.

Ethical approval for the study was obtained from the Ethics Committee of the University Clinical Center of Serbia (approval number 788/7). All patients gave informed consent before the procedure.

After hospital admission, a detailed medical history was obtained and a physical examination was performed, including a digital rectal examination. The primary endpoint is subjective improvement in continence measured with the WS and Faecal Incontinence Quality of Life Score (FIQLS), as previously described [23–25]. The secondary endpoint is an objective estimation of anal sphincter function measured as basal pressure (BP) and squeeze pressure (SP), as well as internal anal sphincter (IAS) and external anal sphincter (EAS) thickness values. The WS, FIQLS and anal manometry and endoanal ultrasound measurements were all registered before the procedure. IAS and EAS thicknesses were measured at the 12 and 9 o'clock positions using endoanal ultrasound (BK Medical Pro Focus ultrasound system, type 2202). BP and SP were measured by anal manometry, using a nonperfusion water sphincterometer (MSM ProMedico GmbH, STM-0234-SM). Endoanal ultrasound and anal manometry were always performed by the same investigator, while WS and FIQLS were obtained by others.

The surgery was done under general anaesthesia. All patients received one prophylactic intravenous dose of ceftriaxone 2 g. After preparation of the operative field, liposuction was performed in the same manner in all subjects. The subcutaneous layer of the abdominal area was infiltrated with a fluid containing 100 ml of normal saline and 0.1 ml of epinephrine (1:1,000,000). Following that, a 3 mm blunt harvesting cannula (Tulip® Medical Products, San Diego, USA) was introduced through a 4 mm stab incision on the right lateral side of the abdomen into the fat tissue and 50 ml of raw lipoaspirate was obtained into 50 cm^3 syringes with a Johnnie Lock (Tulip Medical) installed to hold suction. The lipoaspirate was then processed using the NanoFat Kit device (Tulip® Medical Products, San Diego, USA) according to the manufacturer's protocol. In short, the fat was transferred into 20 cm^3

Luer Lock syringes and was sequentially passed 30 times first through a 2.4 mm Luer-to-Luer transfer followed by 1.2 mm transfer. The obtained product was connected to a sterile NanoTransfer device (Tulip Medical) and transferred by a single pass through a 0.6–0.4 mm mesh screen into a 20 cm^3 syringe. We obtained approximately 20 ml of mechanically emulsified and filtrated fat (so-called nanofat) for transplant.

After liposuction, the wound was sutured, the incision was dressed and the patient was set in the lithotomy position. Then, the anal sphincter complex was infiltrated with the nanofat transplant in a volume of 3 ml at both the 3 and 9 o'clock positions (at the edges of overlapped anal sphincter muscles) and with 9 ml at a position between 10 and 2 o'clock into the new anal sphincter complex (overlapped EAS and IAS). Since the injection was not performed under endoanal ultrasound guidance, nanofat was applied in both the EAS and IAS at the predefined areas. Patients were discharged the next day with no therapy prescribed and follow-up was conducted in person or via telephone. Endoanal ultrasound and anal sphincter BP and SP were remeasured 6 months postsurgery by the same investigator who was blinded regarding the outcome, while WS and FQLS were determined 6 and 12 months after the procedure (the 12 month time point evaluation was conducted via telephone).

Statistical analysis

Statistical analyses were performed using IBM SPSS version 18.0 (IBM Corp., Armonk, NY, USA). Due to the small sample size, the Shapiro–Wilk test was performed for the assessment of data distribution. The Mann–Whitney *U*-test was performed for unrelated group comparison and Friedman's nonparametric repeated measures analysis of variance with Wilcoxon's paired post hoc test for related group comparison. For correlation assessment, Spearman's nonparametric correlation test was employed and *p*-values less than 0.05 were considered statistically significant.

RESULTS

Demography and clinical characteristics of the nine patients included in this study are presented in Table 1. After nanofat injection, patients reported minimal local discomfort in the anal region which resolved after 48 h and localized bruising in the liposuction area that lasted for 2 weeks, on average. Other procedure-related or other types of complications were not observed in any of the participants.

The values of BP, SP and IAS and EAS thickness before and 6 months after the procedure are presented in Table 1. The SP was significantly increased 6 months after the procedure ($p = 0.01$) (Figure 1A), while the increment in BP had marginal significance ($p = 0.09$). The EAS measured at the 12 o'clock position was significantly thicker ($p = 0.04$) (Figure 1B), while its thickness at the 9 o'clock position did not change significantly. IAS thickness measured at the 12 o'clock position was increased but not statistically significantly ($p = 0.08$); the same measurement at the 9 o'clock position also did not change significantly.

WS and FIQLS were measured in three time points, before and 6 and 12 months after the procedure (Table 1). A significant decrease of WS was observed at both 6 and 12 months after the procedure compared with baseline values ($p < 0.05$ for both) (Figure 1C). All FIQLS scales remained unchanged 6 months after the procedure compared with the baseline values. FIQL summary score (FIQLSS) increased after 6 months with borderline significance ($p = 0.05$). Twelve months after the procedure, FIQL 1, FIQL 3 and FIQLSS increased significantly ($p < 0.05$ for all parameters) compared with the baseline value (Figure 1D–F). There was no significant difference in any of the scores 6 and 12 months after the procedure. Also, there was no significant correlation of time elapsed between sphincteroplasty and inclusion into the study with WS and FQLS 6 and 12 months after the procedure.

To estimate the overall improvement of continence 12 months after the procedure, we calculated the change of WS and FIQLSS between baseline and 12 months (ΔWS and ΔFIQLSS, respectively). The decrease in WS ranged from 0 to 10 points, with a median value of 5. The observed WS decrease was ≤5 points in five patients and >5 points in four patients. The change of FIQLSS ranged from −2.03 to +5.26 points, with a median value of 0.89. The same five patients with a WS decrease of ≤5 points had a FIQLSS difference of <1 point, and the four patients with a WS decrease >5 points had a FIQLSS difference ≥1 point. Based on these cut-off values, we divided patients into two groups: those who benefited more (ΔWS > 5 and ΔFIQLSS ≥ 1) and less (ΔWS ≤ 5 and ΔFIQLSS < 1) from treatment. All measured variables were compared between these two groups, and four parameters were found to differ significantly. SP values, both baseline and 6 months after the procedure, were significantly higher in the group of patients who benefited more from the treatment. On the other hand, values for FIQLS scales 1 (baseline) and 4 (6 months after the procedure) were significantly lower in the group of patients who benefited more from treatment (Figure 2).

A significant positive correlation was revealed between WS values at baseline and 6 months after the procedure ($\rho = 0.747$, $p < 0.05$), as well as between FIQLSS values at the same time points ($\rho = 0.783$, $p < 0.05$). A significant negative correlation was detected between WS values at baseline and FIQLSS values at baseline, 6 and 12 months after the procedure ($\rho = -0.869$, $\rho = -0.895$ and $\rho = -0.682$, respectively; $p < 0.05$), as well as between WS and FQLSS values 6 months after the procedure ($\rho = -0.831$, $p < 0.05$). There was no significant correlation between WS values 12 months after the procedure and any other score values.

DISCUSSION

Although not a life-threatening condition, faecal incontinence has significant social and psychological implications and severely affects quality of life.

TABLE 1 Patients' demographic characteristics and measurements at baseline and 6 and 12 months' posttreatment

Patient number		1	2	3	4	5	6	7	8	9
Gender		F	F	F	F	F	F	F	F	F
Age (years)		56	59	41	33	51	28	38	53	27
BMI (kg/m^2)		26.6	29.8	38.1	31.4	24.7	21.6	30.9	28.1	31.6
Incontinence aetiology		OI	OI	OI	OI	OI	OI	OI	OI	OI
Repair type		AOS	AOS	AOS	AOS	AOS	AOS	AOS	AOS	AOS
Time elapsed between injury and repair (years)		16	35	3	1	8	1	1	22	1
Time elapsed between repair and nanofat injection (years)		2	3	4	2	20	5	3	7	6
Associated conditions		–	Rectocele	–	–	–	–	–	Perineal cyst	RV fistula
Baseline values	BP (mm Hg)	30	29	23	32	6	12	12	23	33
	SP (mm Hg)	80	48	69	89	34	52	77	57	47
	IAS 12 o'clock (mm)	1.3	0.8	1.7	1	1.6	1.9	1.3	1.6	1.5
	IAS 9 o'clock (mm)	1.4	1.1	1.7	1.6	1.7	1.6	2.6	2.4	2
	EAS 12 o'clock (mm)	1.5	3.1	2.1	2.2	1.4	1.6	2.4	2.2	1.9
	EAS 9 o'clock (mm)	2.6	3.7	3.6	4.3	4	3.7	4.2	4.1	4.2
	WS	9	12	13	11	9	6	11	7	7
	FIQL Scale 1	3.20	2.60	1.80	2.00	3.90	4.00	3.00	3.90	4.00
	FIQL Scale 2	1.67	1.33	1.22	1.78	3.33	3.78	3.11	3.67	2.78
	FIQL Scale 3	2.86	2.86	1.86	2.14	3.43	3.86	3.57	4.00	2.29
	FIQL Scale 4	2.67	2.33	2.00	2.33	3.67	3.33	1.67	3.67	3.67
	FIQLSS	10.39	9.12	6.88	8.25	14.33	14.97	11.35	15.23	12.73
6 month values	BP (mm Hg)	30	23	28	32	17	26	32	25	38
	SP (mm Hg)	103	59	124	89	45	63	101	100	61
	IAS 12 o'clock (mm)	1.9	2.1	1.4	1.2	1.7	2	1.5	2.4	1.4
	IAS 9 o'clock (mm)	1.5	1.2	1.7	1.7	1.7	1.6	1.9	2.4	1.9
	EAS 12 o'clock (mm)	2.3	2.8	2.4	2.5	1.9	2.5	2.5	3.4	1.9
	EAS 9 o'clock (mm)	3.7	3.8	3.4	4.4	4.2	3.6	4	3.6	4.1
	WS	4	12	6	5	7	3	4	4	2
	FIQL Scale 1	3.1	3.1	2.6	2.9	3.5	4	2.8	3.9	3.8
	FIQL Scale 2	2.56	1.67	1.89	2.89	2.22	3.78	3.00	3.78	3.22
	FIQL Scale 3	3.29	3.29	1.86	3.71	2.57	3.86	3.43	4.00	2.86
	FIQL Scale 4	4.00	1.33	2.67	3.00	4.00	4.00	3.33	4.00	3.33
	FIQLSS	12.94	9.39	9.01	12.50	12.29	15.63	12.56	15.68	13.21

TABLE 1 (Continued)

Patient number		1	2	3	4	5	6	7	8	9
12 month values	WS	3	12	5	3	4	3	1	5	3
	FIQL Scale 1	3.80	2.70	2.70	3.10	4.00	4.00	3.40	3.90	3.80
	FIQL Scale 2	3.89	1.44	2.11	2.56	3.22	4.00	2.78	3.33	2.00
	FIQL Scale 3	3.86	2.86	2.86	3.86	3.43	3.86	3.71	4.00	2.57
	FIQL Scale 4	4.00	2.00	3.00	4.00	4.00	4.00	3.67	4.00	2.33
	FIQLSS	15.55	9.00	10.67	13.51	14.65	15.86	13.56	15.23	10.70

Abbreviations: AOS, anterior overlapping sphincteroplasty; BMI, body mass index; BP, basal pressure; EAS, external anal sphincter; FIQL, Faecal Incontinence Quality of Life; FIQLSS, Faecal Incontinence Quality of Life Summary Score; IAS, internal anal sphincter; OI, obstetric injury; RV, rectovaginal; SP, squeeze pressure; WS, Wexner Score.

Described by Lockhart-Mummary in 1923, sphincteroplasty was the first surgery performed to remedy this condition [26]. Parks further popularized it, introducing the overlapping technique to clinical practice [27]. Since then, this procedure has become the cornerstone in the treatment of anal incontinence caused by mechanical injuries. While the short-term results after sphincteroplasty are promising, maintaining continence over time remains a significant challenge [28]. Worsening of continence over time could be the consequence of disruption of repaired muscle ends, progression of pudendal neuropathy or both. Disrupted muscle ends can be identified by endoanal ultrasonography, while pudendal neuropathy can be detected by PNTMLs measurement. Unfortunately, we were not able to measure PNTMLs and determine if incontinence was a result of pudendal nerve injury during vaginal delivery. In patients with unsatisfactory postoperative results, biofeedback can improve continence [4,29] but the long-term benefits remain unclear. Sacral nerve stimulation may be a promising option for patients with unsatisfactory results after sphincter repair [30]. However, this procedure is not available in all countries.

Adipose tissue has been recognized as an easily accessible and rich source of adipose-derived stem and other regenerative cells [31,32]. ADSC serve as paracrine mediators by delivering trophic factors and enzymes, especially in hypoxic conditions [33]. Additionally, these cells modulate immune reactions [34] and have the potential to differentiate into mature adipocytes, endothelial cells or other cells [35].

There are two common methods to process lipoaspirates to obtain a product with regenerative potential: (a) enzymatic digestion (the product is a stromal vascular fraction pellet) and (b) mechanical disaggregation (the products are cell aggregates, nanofat) [36]. Therefore, nanofat can be defined as an injectable product composed of cell aggregates that is obtained from fat by the process of emulsification and filtration.

In their recently published paper Sese et al. [37] analysed the cell inoculum obtained by the same protocol and device (NanoFat Kit by Tulip Medical) that we used in our study. They found that the aforementioned method of nanofat preparation resulted in an average of 125 million cells from 20 cm^3 of raw lipoaspirate, a cell yield which is 10 times higher than one obtained by enzymatic dissociation from the same amount of fat tissue. The cell content analysis showed no statistical difference between nanofat and lipoaspirate cell burden, suggesting that nanofat comprises a stromal cell population equivalent to the aspirated adipose tissue. Furthermore, this mechanical processing method is easy to perform and does not require enzymes, the manipulation is minimal, it is cost-effective and does not bear the regulatory hurdles of enzymatic digestion [19]. Consequently, it is becoming increasingly popular for use in different surgical treatments and clinical applications [38].

The application of enzymatically isolated ADSC has shown promising results in the treatment of proctological diseases such as perianal fistula [15,16,39] and chronic anal fissure refractory to medication [17].

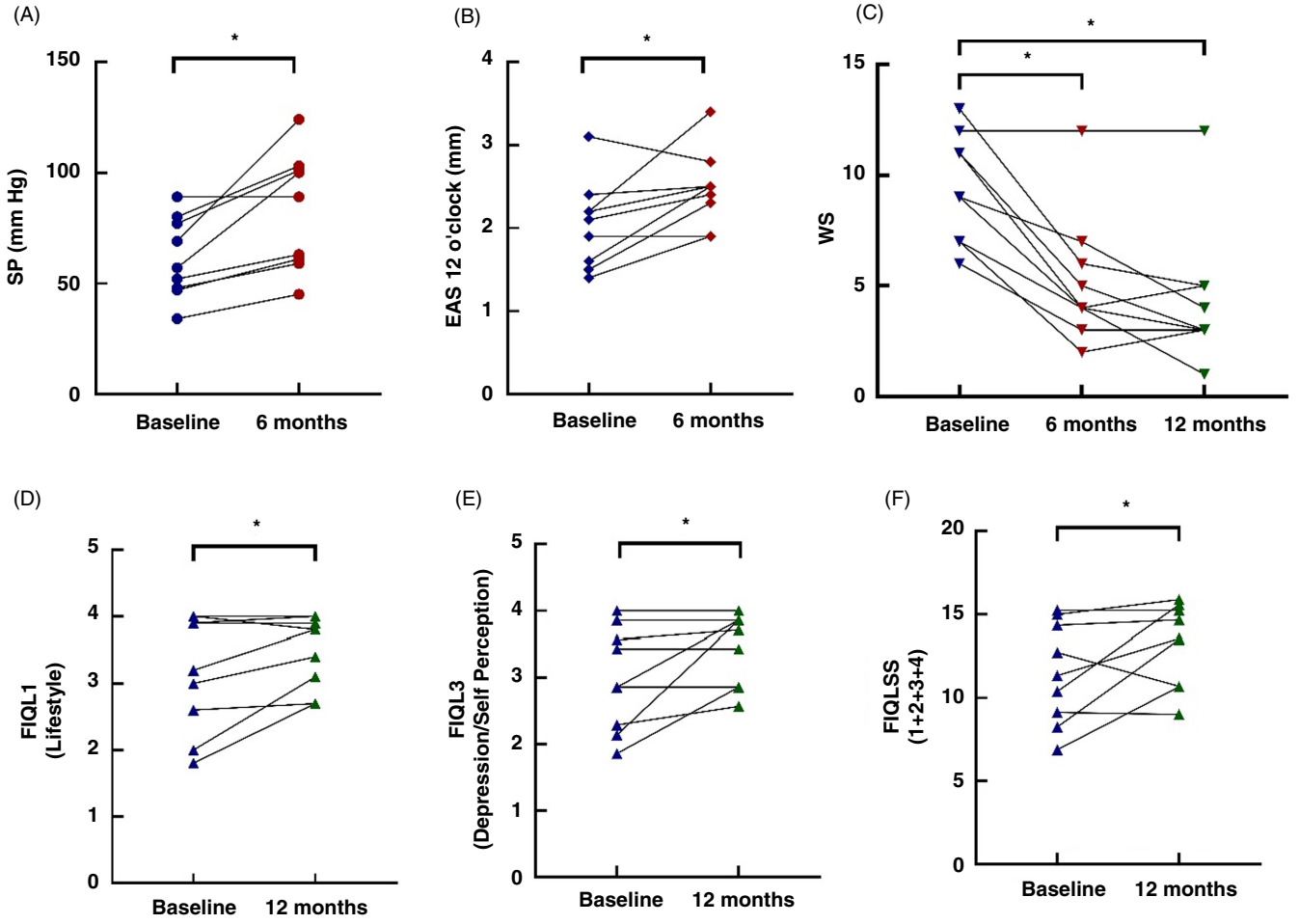

FIGURE 1 Significantly different parameters after nanofat injection. Each point (●, ♦, ▼ and ▲) represents an individual value for a single patient, and a horizontal line connects measures in different time points. *p-value from Wilcoxon's paired test (A and B) and Friedman's test for related samples (C, D, E and F). EAS, external anal sphincter; FIQL, Faecal Incontinence Quality of Life Score; WS, Wexner Incontinence Score

Previous studies in the murine model of anal sphincter injury showed improved anal manometry results following treatment with enzymatically isolated ADSC [40,41]. Inoue et al. histologically detected reproduction of smooth muscle in rats injected with allogeneic rat ADSC sheets after sphincterotomy, while in the control group only collagen fibres were observed at the sphincterotomy site [41]. Conversely, Kuismanen et al. did not find a histological difference between rats injected with human ADSC in repaired anal sphincter after injury and rats injected with saline or bulking agent without cells [40]. However, significant improvement of BP and SP was observed in ADSC-treated rats in combination with both bulking agent and saline compared with bulking agent and saline alone [40]. This indicates that the mechanism of detected functional restoration cannot be attributed to a bulking effect [40].

Using electromyography (EMG), Sarveazad et al. detected higher numbers of motor units in a rabbit model with sphincterotomy treated with human ADSC alone and in combination with laser treatment compared with laser-treated and untreated rabbits [42]. Functional improvement was also detected, with a higher BP in rabbits with the ADSC and laser treatment combination and significant improvement in SP in all treated animals compared with the sphincterotomized animals [42].

In humans, application of allogeneic human ADSC during sphincteroplasty showed better endoanal ultrasound parameters and significantly higher EMG activity compared with sphincteroplasty without ADSC application [43]. Sarveazad et al. identified by ultrasonography an increased ratio of the area occupied by the muscle to the total area in ADSC-treated patients, indicating replacement of fibrous tissue and restoration of action potentials [43]. On the other hand, De la Portilla et al. failed to demonstrate an improvement in measured clinical, manometric and ultrasonographic parameters in patients with structural faecal incontinence treated with autologous expanded mesenchymal stem cells [44]. Although application of ADSC was investigated in animal models and humans as a treatment for anal incontinence of various aetiologies, acute or healed injuries with or without reparation, it has never been investigated in patients years after surgical reconstruction [45]. To our knowledge, this is the first study to evaluate the use of mechanically disaggregated fat, nanofat, in the therapy of faecal incontinence after anal sphincteroplasty in humans.

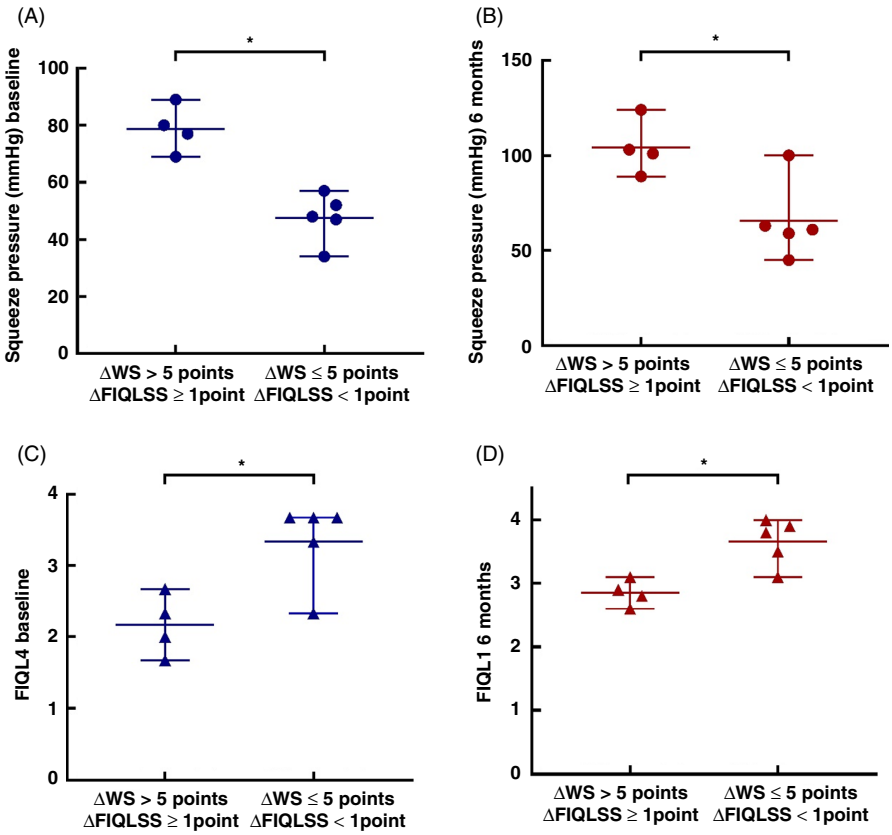

FIGURE 2 Parameters that significantly differ between patients who benefited more (ΔWS > 5 and ΔFIQLSS ≥ 1) and those who benefited less (ΔWS ≤ 5 and ΔFIQLSS < 1) from treatment based on measurements at 12 month. Each point (● and ▲) represents an individual value for a single patient at given time point. *p-value from the Mann–Whitney U-test. FIQL, Faecal Incontinence Quality of Life Score; WS, Wexner Incontinence Score

Anal continence is predominantly maintained by the anal sphincter complex formed from the IAS and EAS. The IAS is a major contributor in providing anal continence, responsible for 75% of basal pressure maintenance [46]. The EAS is a skeletal muscle that allows voluntary control and is responsible for SP. Six months after the procedure, the thickness of the EAS increased significantly at the 12 o'clock position, followed by a significant elevation of SP values. IAS thickness, as well as BP values, increased with marginal significance. This improvement could indicate a successful regeneration of the muscle cells in the position where the thickness was most compromised due to the previous injury. Considering that thickening of the EAS and IAS occurred in conjunction with elevation of SP and BP, as well as results from the above-mentioned previous studies, we can assume that the improvement of continence was a consequence of ADSC differentiation into muscle cells and not just a bulking effect. Histopathological assessment of the injected zone was not performed since obtaining a biopsy specimen could compromise the repaired muscle.

In this study, we decided to employ the WS for estimation of the severity of faecal incontinence and FIQLS for evaluation of quality of life since they have been widely used for their good reproducibility and simplicity [47]. Continence was significantly improved at 6 and 12 months after the procedure according to the WS results. The quality of life after the procedure also improved significantly, as demonstrated by the elevation of FIQLSS after 6 and 12 months, and FIQL 1 and FIQL 3 after 12 months.

After observing improvement in both continence and quality of life, we estimated a degree of change and divided patients according to those who benefited more and less from the treatment. The patients who benefited more had significantly higher SP values at baseline, as well as 6 months after the procedure. This indicates that nanofat application is a more suitable treatment option for patients with better anal sphincter function at baseline. Conversely, patients who benefited more from treatment had significantly higher embarrassment initially (lower FIQL 4) and impact on lifestyle measured after 6 months (lower FIQL 1).

A good correlation between WS and FIQL scores has already been demonstrated, jointly allowing for better patient evaluation [48]. As we presumed, WS and FIQLSS were significantly negatively correlated, demonstrating the validity of using the FIQLSS.

The main study limitation is the small number of enrolled participants. The limited number of patients with previous anal sphincteroplasty can be justified since this procedure is not frequently performed. Additionally, restrictive eligibility criteria were employed to obtain a well-characterized homogeneous patient group. For the same reason, this study design did not include a no-treatment control group.

CONCLUSION

The application of nanofat as an injectable product showed an improvement in continence in patients with unsatisfactory results after sphincteroplasty and presents a promising treatment option and an effective therapeutic tool. The procedure is safe and can be easily performed as an ambulatory procedure without any

concerns regarding regulatory or other issues. Further investigations on a larger cohort are needed for validation of these results and ultimately the establishment of this procedure in routine clinical practice.

CONFLICT OF INTEREST

The authors declare that there have no conflicts of interest.

AUTHOR CONTRIBUTIONS

GB: conception and design, acquisition of data, analysis and interpretation of data, revising the article, final approval of the article. KA: conception and design, acquisition of data, analysis and interpretation of data, revising the article, final approval of the article. JR: acquisition of data, analysis and interpretation of data, drafting the article, revising the article, final approval of the article. MM: acquisition of data, analysis and interpretation of data, drafting the article, revising the article, final approval of the article. JKS: acquisition of data, analysis and interpretation of data, revising the article, final approval of the article. TD: acquisition of data, analysis and interpretation of data, drafting the article, final approval of the article. JJ: acquisition of data, analysis and interpretation of data, drafting the article, final approval of the article. ZK: conception and design, acquisition of data, analysis and interpretation of data, revising the article, final approval of the article.

FUNDING INFORMATION

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ETHICS APPROVAL AND PATIENT CONSENT STATEMENTS

Ethical approval for this study was obtained from the Ethics Committee of the University Clinical Center of Serbia (approval number 788/7). All patients signed informed consent before the procedure.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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REFERENCES

1. Lee J, Vogel S, Madoff R. Diagnosis and management of fecal incontinence. In: Yeo CJ, editor. Shackelford's Surgery of the Alimentary Tract. 7th ed. Philadelphia, PA: Elsevier/Saunders; 2013.
2. Nelson RL. Epidemiology and Incidence of anal Incontinence: magnitude of the Problem. Seminars in Colon and Rectal Surgery. Philadelphia, PA: WB Saunders Co.; 1997 c1990-.
3. Gilliland R, Altomare DF, Moreira H, Oliveira L, Gilliland JE, Wexner SD. Pudendal neuropathy is predictive of failure following anterior overlapping sphincteroplasty. Dis Colon Rectum. 1998;41(12):1516–22.
4. Jensen LL, Lowry AC. Biofeedback improves functional outcome after sphincteroplasty. Dis Colon Rectum. 1997;40(2):197–200.
5. Zetterström J, López A, Holmström B, Nilsson BY, Tisell Å, Anzén B, et al. Obstetric sphincter tears and anal incontinence: an observational follow-up study. Acta Obstet Gynecol Scand. 2003;82(10):921–8.
6. Barisić G, Krivokapić Z, Marković V, Popović M, Saranović D, Marsavelska A. The role of overlapping sphincteroplasty in traumatic fecal incontinence. Acta Chir Iugosl. 2000;47(4 Suppl 1):37–41.
7. Karoui S, Leroi A, Koning E, Menard J, Michot F, Denis P. Results of sphincteroplasty in 86 patients with anal incontinence. Dis Colon Rectum. 2000;43(6):813–20.
8. Halverson AL, Hull TL. Long-term outcome of overlapping anal sphincter repair. Dis Colon Rectum. 2002;45(3):345–8.
9. Malouf AJ, Norton CS, Engel AF, Nicholls RJ, Kamm MA. Long-term results of overlapping anterior anal-sphincter repair for obstetric trauma. Lancet. 2000;355(9200):260–5.
10. Gutierrez AB, Madoff RD, Lowry AC, Parker SC, Buie WD, Baxter NN. Long-term results of anterior sphincteroplasty. Dis Colon Rectum. 2004;47(5):727–32.
11. Barisic GI, Krivokapic ZV, Markovic VA, Popovic MA. Outcome of overlapping anal sphincter repair after 3 months and after a mean of 80 months. Int J Colorectal Dis. 2006;21(1):52–6.
12. Hu MS, Borrelli MR, Lorenz HP, Longaker MT, Wan DC. Mesenchymal stromal cells and cutaneous wound healing: a comprehensive review of the background, role, and therapeutic potential. Stem Cells Int. 2018;2018:1–13.
13. Bartczak A, McGilvray I, Keating A. Mesenchymal stromal cell therapy to promote cardiac tissue regeneration and repair. Curr Opin Organ Transplant. 2017;22(1):86–96.
14. Voswinkel J, Francois S, Simon JM, Benderitter M, Gorin NC, Mohty M, et al. Use of mesenchymal stem cells (MSC) in chronic inflammatory fistulizing and fibrotic diseases: a comprehensive review. Clin Rev Allergy Immunol. 2013;45(2):180–92.
15. Lee WY, Park KJ, Cho YB, Yoon SN, Song KH, Kim DS, et al. Autologous adipose tissue-derived stem cells treatment demonstrated favorable and sustainable therapeutic effect for Crohn's fistula. Stem Cells. 2013;31(11):2575–81.
16. Herreros MD, Garcia-Arranz M, Guadalajara H, De-La-Quintana P, Garcia-Olmo D. Autologous expanded adipose-derived stem cells for the treatment of complex cryptoglandular perianal fistulas: a phase III randomized clinical trial (FATT 1 fistula Advanced Therapy Trial 1) and long-term evaluation. Dis Colon Rectum. 2012;55(7):762–72.
17. Andjelkov K, Sforza M, Barisic G, Soldatovic I, Hiranyakas A, Krivokapic Z. A novel method for treatment of chronic anal fissure: adipose-derived regenerative cells–a pilot study. Colorectal Dis. 2017;19(6):570–5.
18. Feng J, Doi K, Kuno S, Mineda K, Kato H, Kinoshita K, et al. Micronized cellular adipose matrix as a therapeutic injectable for diabetic ulcer. Regen Med. 2015;10(6):699–708.
19. Mashiko T, Wu SH, Feng J, Kanayama K, Kinoshita K, Sunaga A, et al. Mechanical micronization of lipoaspirates: squeeze and emulsification techniques. Plast Reconstr Surg. 2017;139(1):79–90.
20. Tonnard PL, Verpaele AM. Discussion: mechanical micronization of lipoaspirates: squeeze and emulsification techniques. Plast Reconstr Surg. 2017;139(1):91–2.
21. Chaput B, Bertheuil N, Escubes M, Grolleau JL, Garrido I, Laloze J, et al. Mechanically isolated stromal vascular fraction provides a valid and useful collagenase-free alternative technique: a comparative study. Plast Reconstr Surg. 2016;138(4):807–19.
22. Bertheuil N, Chaput B, Ménard C, Varin A, Garrido I, Grolleau JL, et al. Adipose-derived stromal cells: history, isolation, immunomodulatory properties and clinical perspectives. Ann Chir Plast Esthet. 2015;60(2):94–102.
23. Jorge JMN, Wexner SD. Etiology and management of fecal incontinence. Dis Colon Rectum. 1993;36(1):77–97.
24. Rockwood TH, Church JM, Fleshman JW, Kane RL, Mavrantonis C, Thorson AG, et al. Fecal incontinence quality of life scale. Dis Colon Rectum. 2000;43(1):9–16.

25. Lin YH, Chen HP, Liu KW. Fecal incontinence and quality of life in adults with rectal cancer after lower anterior resection. J Wound Ostomy Continence Nurs. 2015;42(4):395–400.
26. Mummery JPL. Diseases of the Rectum & Colon and Their Surgical Treatment. Toronto: MacMillan; 1923.
27. Parks AG, McPartlin JF. Late repair of injuries of the anal sphincter. Proc R Soc Med. 1971;64(12):1187–9.
28. Johnson E, Carlsen E, Steen TB, Backer Hjorthaug JO, Eriksen MT, Johannessen HO. Short-and long-term results of secondary anterior sphincteroplasty in 33 patients with obstetric injury. Acta Obstet Gynecol Scand. 2010;89(11):1466–72.
29. Ghahramani L, Mohammadipour M, Roshanravan R, Hajihosseini F, Bananzadeh A, Izadpanah A, et al. Efficacy of biofeedback therapy before and after sphincteroplasty for fecal incontinence because of obstetric injury: a randomized controlled trial. Iran J Med Sci. 2016;41(2):126–31.
30. Ong K, Bordeianou L, Brunner M, Buntzen S, Collie MHS, Hanly A, et al. Changing paradigm of sacral neuromodulation and external anal sphincter repair for faecal incontinence in specialist centres. Colorectal Dis. 2021;23(3):710–5.
31. Strem BM, Hicok KC, Zhu M, Wulur I, Alfonso Z, Schreiber RE, et al. Multipotential differentiation of adipose tissue-derived stem cells. Keio J Med. 2005;54(3):132–41.
32. Pikuła M, Marek-Trzonkowska N, Wardowska A, Renkielska A, Trzonkowski P. Adipose tissue-derived stem cells in clinical applications. Expert Opin Biol Ther. 2013;13(10):1357–70.
33. Suga H, Eto H, Shigeura T, Inoue K, Aoi N, Kato H, et al. IFATS collection: fibroblast growth factor-2-induced hepatocyte growth factor secretion by adipose-derived stromal cells inhibits postinjury fibrogenesis through a c-Jun N-terminal kinase-dependent mechanism. Stem Cells. 2009;27(1):238–49.
34. De Miguel MP, Fuentes-Julián S, Blázquez-Martínez A, Pascual CY, Aller MA, Arias J, et al. Immunosuppressive properties of mesenchymal stem cells: advances and applications. Curr Mol Med. 2012;12(5):574–91.
35. Planat-Benard V, Silvestre JS, Cousin B, André M, Nibbelink M, Tamarat R, et al. Plasticity of human adipose lineage cells toward endothelial cells: physiological and therapeutic perspectives. Circulation. 2004;109(5):656–63.
36. Sherman LS, Condé-Green A, Kotamarti VS, Lee ES, Rameshwar P. Enzyme-free isolation of adipose-derived mesenchymal stem cells. Methods Mol Biol. 2018;1842:203–6.
37. Sesé B, Sanmartín JM, Ortega B, Matas-Palau A, Llull R. Nanofat cell aggregates: a nearly constitutive stromal cell inoculum for regenerative site-specific therapies. Plast Reconstr Surg. 2019;144(5):1079–88.
38. Tonnard P, Verpaele A, Peeters G, Hamdi M, Cornelissen M, Declercq H. Nanofat grafting: basic research and clinical applications. Plast Reconstr Surg. 2013;132(4):1017–26.
39. Zhou C, Li M, Zhang Y, Ni M, Wang Y, Xu D, et al. Autologous adipose-derived stem cells for the treatment of Crohn's fistula-in-ano: an open-label, controlled trial. Stem Cell Res Ther. 2020;11(1):1–13.
40. Kuismanen K, Juntunen M, Girish NN, Tuominen H, Huhtala H, Nieminen K, et al. Functional outcome of human adipose stem cell injections in rat anal sphincter acute injury model. Stem Cells Transl Med. 2018;7(3):295–304.
41. Inoue Y, Fujita F, Yamaguchi I, Kinoe H, Kawahara D, Sakai Y, et al. Improvement of anal function by adipose-derived stem cell sheets. Digest Surg. 2018;35(1):64–9.
42. Sarveazad A, Babahajian A, Yari A, Rayner CK, Mokhtare M, Babaei-Ghazani A, et al. Combination of laser and human adipose-derived stem cells in repair of rabbit anal sphincter injury: a new therapeutic approach. Stem Cell Res Ther. 2019;10(1):367.
43. Sarveazad A, Newstead GL, Mirzaei R, Joghataei MT, Bakhtiari M, Babahajian A, et al. A new method for treating fecal incontinence by implanting stem cells derived from human adipose tissue: preliminary findings of a randomized double-blind clinical trial. Stem Cell Res Ther. 2017;8(1):1–10.
44. De la Portilla F, Guerrero JL, Maestre MV, Leyva L, Mera S, García-Olmo D, et al. Treatment of faecal incontinence with autologous expanded mesenchymal stem cells: results of a pilot study. Colorectal Dis. 2021;23(3):698–709.
45. Balaphas A, Meyer J, Meier RPH, Liot E, Buchs NC, Roche B, et al. Cell therapy for anal sphincter incontinence: where do we stand? Cell. 2021;10(8):2086.
46. Jones JFX. Fecal incontinence. In: Robertson D, Biaggioni I, Burnstock G, Low PA, Paton JFR, editors. Primer on the Autonomic Nervous System. 3rd ed. San Diego: Academic Press; 2012. p. 601–2.
47. Bols EMJ, Hendriks HJM, Berghmans LCM, Baeten CGMI, de Bie RA. Responsiveness and interpretability of incontinence severity scores and FIQL in patients with fecal incontinence: a secondary analysis from a randomized controlled trial. Int Urogynecol J. 2013;24(3):469–78.
48. Ciriza de los Ríos C, Ruiz de León A, García Durán F, Tomás Moros E, Carneros Martín JA, Muñoz Yague T, et al. Calidad de vida en pacientes con incontinencia fecal y su relación con la gravedad de la misma. Gastroenterol Hepatol. 2010;33:621–8.

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