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Research Paper

Efficacy and feasibility of stromal vascular fraction containing adipose derived stem cells obtained from freshly prepared lipoaspirate for treatment of complex anorectal fistulas- A novel approach

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ABSTRACT

Background: Anal fistulas are relatively common. Despite diverse treatment modalities and tremendous advance and optimization of surgical approaches, considerable recurrence and high failure rates are not uncommon especially after surgery for complex anal fistulas. Therefore, autologous adipose-derived stem cell therapy for the treatment of complex perianal fistula can be an innovative and promising option with potential high cure rate and long-term healing.

Aim: To assess the efficacy and feasibility intralesional administration of stromal vascular fraction containing autologous, adipose-derived stem cells prepared from fresh lipoaspirate for management of complex and recurrent anal fistula.

Methods: Totally, 28 patients with complex perianal fistulas were enrolled in this prospective case-series study from March 2018 and August 2022. They were 22 male and 6 females patient) with 38.8 years average ages (ranges from 26 to 54 years). All patients were assigned to receive intralesional injection with autologous freshly prepared adipose-derived stem cells from lipoaspirate suction in a concentration of 1×10^7 cell/ml. The healing of fistulas, confirmed by clinical assessment and magnetic resonance imaging was evaluated at week 6 weeks, 12 weeks and 6 months after treatment. If no evidence of healing was not seen at 6 weeks, a second dose of same concentration was administered. In addition to fistula healing, adverse effect (functional outcomes), complications and procedure safety were also evaluated.

Results: The overall healing rate of fistulas treated by injection of ADSCs was 60.7% (17/28), while healing rate after first injection was 50% (14/28) and after second injection was 30% (3/10).Three patients were lost after first injection. During the follow up period of 6 months none of the patients who showed complete healing, recorded relapse of their fistulas (recurrence 0%). Further, none of the patients recorded complications or adverse reactions related to stem cell therapy during the study and none of them had worsening fecal incontinence during follow-up period. Postoperative pain assessed by.

VAS score was significantly low (>3). Fecal continence assessed by Wexner score of all patients was not changed or affected.

Conclusions: Injection of complex perianal fistulas by freshly prepared autologous ADSCs could be a valid and effective treatment modality. It is safe, feasible and minimally invasive procedure that lacks the risk of injury to anal sphincter complex, unlike other surgical procedures that endanger the anal continence.Further, second dose of ADSCs injection could improve the rate of healing, without adding adverse effects.

1. Introduction

By definition, Perianal fistula is an abnormal inflammatory tract that connects anal canal or rectum and perianal skin. Most these fistulas are believed to develop as a result of infection of anal crypt glands with resultant perianal abscess whose extension determine the type of anal fistulas [1]. Anal fistulas are categorized into either simple type including intersphincteric and low transsphincteric fistulas or complex

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type including high transsphincteric fistulas, suprasphincteric fistulas, extresphincteric fistulas, fistula with multiple and branching tracts, horseshoe fistulas and anterior fistula in female. Fistulas secondary to malignancy, irradiation and Crohn's disease are also considered as complex fistulas [2] (see Fig. 1)

Surgery remains the treatment of choice for anal fistulas with aim of draining and control sepsis, eradicating the fistula tract(s) while preserving anal continence and avoiding recurrent disease. Although simple fistulas can be treated efficiently by fistulotomy with excellent prognosis and low recurrence rate management of complex anal fistula remains challenging and real surgical dilemma [3]. Different options have been practiced to eradicates complex fistula, but none of these options results in a definitive cure with recurrence rate varies from 7% to 65% and some degree of incontinence in approximately 10%–15% [4]. Treatment options include seton (loose draining and cutting types), endorectal advancement flap, fibrin sealants with adhesives, fistula plug, Laser ablation including FiLaC (Fistula Laser Closure) and VAAFT (Video Assisted Anal Fistula Treatment) as well as LIFT procedure(Ligation of Intersphincteric Fistula Tract) but with insufficient healing rates. Antibiotics mainly metronidazole and ciproflxacine have a limited and temporary effects on symptoms. The presence of abscesses mandates drainage in order to control sepsis followed by fistulotomy with or without draining Seton [5]. Some patients, however, with complex and refractory fistluas, fecal diversion by proximal stoma may be required. Repeated procedures are usually required for more than 25% of complex perianal fistulas; however, such interventions carry poor functional outcomes due to high risk of incontinence and profound alteration in the quality of life [6].

In recent two decades, a new novel approach using allogenic or autologous mesenchymal stem cells derived from adipose tissue has

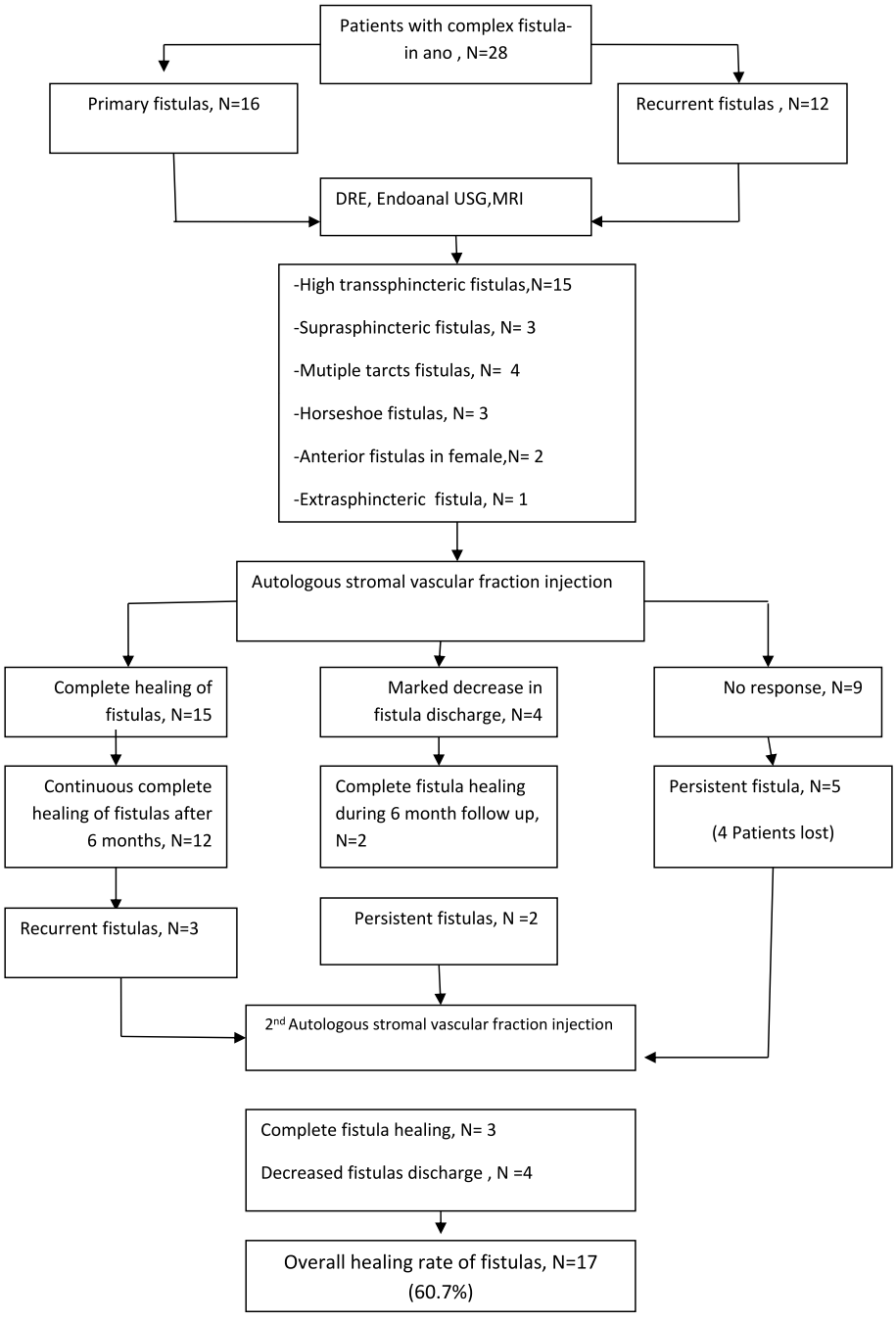

Fig. 1. Flow chart of complex anal fistula management using autologous freshly prepared stromal vascular fraction containing ADSC.

been tried as new modality to treat complex perianal fistulas with no effect on continence [7–9]. Adipose derived stem cells have been largely used since they are easier to harvest than other types of mesenchymal stem cells while maintaining all the properties of multipotency, self-renewal, anti-inflammatory effects, immune-modulation and pro-angiogenesis. Thus, this approach is acted not only on the removal of the damaged tissue but also on a regeneration of new tissue [9]. Adipose-derived stem cells have intrinsic immunomodulatory and some anti-inflammatory properties, with paracrine signaling to adjacent cells to maintain the insitu anti-inflammatory environment. Therefore, they can repair damaged tissues resulting in long-term healing of fistulas. Adipose-derived stem cells are purified from adipose tissue which can be safely harvested by liposuction. The process produces 100 times more stem cells than bone marrow aspirates and the process are quite safe. Initial application of these stem cells in perianal Crohn's disease was promising and found to be safe and effective, while studies on their role for complex anal fistulas are still limited [10].

The aim of the this study is to evaluate the efficacy, safety and feasibility of local injection of autologous adipose -derived stem cells (ADSCs) obtained from freshly prepared aspirate of liposuction for complex cryptoglandula anal fistulas.

1.1. Patients and methods

This is a prospective case series study conducted for the period between March 2018 and August2022, in which in which 28 adult patients (22 males, 6 females) with mean age 38.8 ± 6 years ages (ranges from 26 to 54 years) and mean body mass index26.8 ± 5.5 (average 20.6–30.4). Inclusion criteria were those adult patients with complex anal fistulas including those with high transphincteric, suprasphincteric, and extrasphincteric fistulas, horseshoe fistula, recurrent fistulas with tract more than 4 cm, multiple tract fistulas and anterior fistula in female patients. Exclusion criteria were patients with intersphincteric and low transphincteric fistulas, presence of abscess, fistulas secondary to inflammatory bowel diseases, tuberculosis and malignancy.

Physical examination was performed for patient in knee elbow or left lateral positions. All patients were proceeded for three dimensional endoanal ultrasound with rotating 10–16 MHz endoprobe with an internal puller giving a 360° 3D image and allowing the longitudinal distance to be measured. Identifications of internal fistula opening was done according to Cho criteria which appeared as submucosal breach connected to an internal sphincter defect or as a budding which is in contact or inside the internal sphincter. Anal continence especially for those patients who had previous anal fistulas surgery were also studied using Wexner score to assess ability of the patient to hold stool, liquid stool, gases, and soiling. Previous anal surgery for anal fistulas, number and type of intervention were also recorded. Pelvic perineal MRI were performed for all patients consisting of T1-weighted and high -spatial resolution T2 images for delineation of fistula tract, muscles group and fat plane, thus giving the most detailed information about the anatomical characteristics of fistula and its relationship to the anal sphincter complex. The fistulas then were classified acceding Park's classification [11] depending mainly on pelvic and perineal MRI reports. Patient's characteristics and demographics are illustrated in Table 1.

Regarding the 28 perianal fistula cases, 16 cases were high transphincteric (Parks type 2), 6 were suprasphincteric (Parks type 3),and 6 of them presented a complex nonCrohn's cryptoglandular fistulas including horseshoe fistula, fistulas with multiple tracts and anterior fistula in female patients (see Table 2). Nineteen of twenty eight had previous failed surgeries (persistent or recurrent fistulas). Apparent fecal incontinence was not observed in any patient at the moment of enrollment in this study (mild soiling was recorded by 4 patients). Three patients had previous closed colostomy after failed fistula surgery.

Procedure: These were day case ones procedure. All patients arrived on the same day of surgery. No bowel preparation or antibiotics were given prior to the surgery. The procedure was fully explained and written informed consent was obtained from all patients before intervention.

Adipose-derived stem cells are extracted by liposuction performed by a plastic surgeon using a thin hollow tube or cannula introduced through small incision under diluted local anesthesia to suck a loosen fat in particularly from the lower abdomen, buttocks or inner and outer thighs using a controlled and careful back and forth motion resulting in mechanical disruption of fatty tissue producing micro-fragmented adipose tissue. The lipoaspirate is then suctioned out using either a syringe attached to the cannula or a surgical sucker.

Techniques to purify the lipoaspirate in brief involves repeated and vigorous washing of the aspirate obtained from liposuction with phosphate buffered saline containing 5%Penicillin/Streptomycin and mechanically disrupted using a gentleMACS Octo Dissociator with heaters.

Table 1
Demographics and baseline characteristics of 28 patients with complex perianal fistulas.

Patients	Total	Primary fistula	Recurrent fistula
Total	28	16	12
Male	22	12	10
Female	6	4	2
Age (mean)	38.3 ± 6		
BMI	26.8 ± 5.5		
VAS[1]	1.8 ± 1.0		
Wexner[2]	2.2 ± 1.7		
Previous intervention			
Negative	10		
Drainage of perianal abscess	6		
Fistulotomy ± seton	4		
Fistuloectomy ± seton	3		
Drainage and seton	6		
Other procedures*	6		
Fistula type (By perIneal MRI)			
High transsphincteric	15		
Fistulas with multiple tracts	4		
Suprasphincteric	3		
Horseshoe fistula	3		
Anterior fistula in female	2		
Extrasphincteric	1		
Anal incontinence	4		
Co-morbidities	13 (46.4%)		

Score ranges from 0 to 10; higher scores suggest more severe pain. [2]Score ranges from 0 to 20; higher scores suggest severe anal incontinence.

Table 2
Outcomes of Autologous ADSCs injection in 28 patients with complex anal fistulas.

Results	NO.	%
Overall success rate	17	61.7
Complete Closure after first injection	14	50
Closure after second injection	3	30
Recurrence after apparent healing	3	
Failed closure (total)	11	
Mean length of follow up period in weeks	24 ± 4	
Complications		
- urine retention	0	
- infection (abscess)	2	
- Bleeding	0	
VAS[a]	2.4 ± 1.4	
Fecal incontinence (Wexner)[b]	2.6 ± 1.2	
Patient satisfaction[c]	7.8 ± 4	
Recurrence (after 6 months follow up)	0	0

[a] Score ranges from 0 to 10; higher scores suggest more severe pain.
[b] Score ranges from 0 to 20; higher scores suggest severe anal incontinence.
[c] Score ranges from 0 to 10; higher scores suggest high degree of satisfaction.

The isolation of stromal vascular fraction (SVF) containing the stem cells from lipoaspirate is performed by digestion and processing of the fatty portion of the lipoaspirate with collagenase separating the contents into two distinct layers: the floating mature adipocytes fraction, and the stromal vascular fraction containing the stem cells in the lower aqueous fraction which is obtained by centrifuging the sample at 2000 rpm for 5 min at room temperature. The stromal vascular fraction is cultured after series of washing and centrifugation in the medium overnight at 37 °C in an atmosphere with 5% CO2. The sample is taken out of the centrifuge and shaken vigorously to disrupt the pellet and to mix the cells. After further centrifugation and spinning, all the collagenase solution above the pellet is aspirated.The stromal vascular fraction containing the stem cells will be apparent as a dark red cells pelleted on the bottom. The supernatant is aspirated and filtered through special strainer. The concentrate is then collected in 1 ml syringes.

All patients included in this study were well prepared prior to injection of stromal vascular fraction containing stem cells. The preparations entailed a careful examination of perianal region under general anesthesia and proper identification and debridement and cleansing of all fistula tracts and internal fistula openings by extensive curettage and washing using hydrogen peroxide diluted in 0.9% normal saline to remove devitalized tissue in particularly in those patients with previous history or presence of seton used to treat or drain anal fistulas. Hence, all the epithelial tissues of the fistulas were removed. Closure of internal fistula opening then were done using 2-0 Vicryl or 0 chromic catgut sutures. After these measures, the solution containing adipose-derived stem cells using long fine spinal needle (23 G) were injected circumferentially around the internal fistula opening and the through the external opening into the fistula walls along its all length avoiding entrance to the lumen of fistula using several passes around the fistulas tract from the internal openings to external openings. The volume of the injected solution ranges between 20 ml and 80 ml (average 60 ml) depending on the length, size and numbers of fistula tracts. The external fistula opening then closed with the same absorbable suture. Rubbing along the whole fistula tract was done for even distribution of implanted cells.

Patients were discharged few hours after recovery with prescription of metronidazole 500 mg three times daily and ciprofloxacin 750 mg twice daily given orally for 5–7 days. Postoperative pain was managed using oral paracetamol 1g and ibuprofen tablet 200 mg twice daily. All patients were informed about the postoperative instructions and possible complications such as pain, urine retention, fever and bleeding. All procedures were done by specialist's expert surgeons in colorectal surgery.

Patients were seen initially 2 weeks postoperatively for clinical assessment and then, they were followed and checked at 6 weeks, 3months and 6 months as an outpatients setting. During these visits, patients were evaluated carefully regarding ceased or persisting discharge both spontaneous and after squeezing of fistulous tract. If the clinical healing of perianal fistula was not occurred six weeks after the injection of ADSCs, a second injection of same concentration was offered. The primary endpoint was to assess healing rate after ADSCs injection. A fistula was deemed completely clinically healed if the patient has no visible discharge and there was closed external fistula opening. For those patients who showed clinical healing, MRI was repeated at 6 months after last injection to ensure that the treatment resulted in complete healing of fistula tracts and not only partial closure of external or internal fistula openings. The perineal MRI healing was defined as complete healing of the internal fistula opening, the inersphincteric portion of the fistula tract, and tracts in the ischiorectal fossa. The secondary outcomes were evaluation of time required to healing of the fistula (both clinical and radiological healing), occurrence of infection, abscess and bleeding, fistula recurrence and lastly, evaluation of functional changes in continence according to Wexner incontinence sco re during follow-up period Postoperative pain was also assessed using VAS score as a reference. Post procedure complications and patient satisfaction were also recorded and evaluated. The work has been reported in line with Consolidated Standards of Reporting Trials (CONSORT) criteria [12]. The study was registered at Researcregistry: http://www.researchregistry.com.

2. Results

A total of 28 consecutive patients with newly diagnosed or recurrent complex perianal fistula were seen between March 2018 and August2022, with male predominance (male: female: 22:6),mean age 38.3 ± 6 years (range26–64 years) and mean BMI was 26.8 ± 5.5. None of the patient had inflammatory bowel diseases. Based on perineal and pelvic MRI findings; 15 fistulas (53.6%), were high transphincteric type, 3 (10.7%)were suprasphincteric, 4 (14.3%) fistulas with multiple tracts, 3 (10.7%) horseshoe fistula, 2 (7.1%)anterior fistula in female patients and 1 (3.6%) extrasphincteric type. Recurrent fistulas submitted to one or more previous various interventions regardless the type were 12 (42.9%). The average number of operations before local infiltration of ADSC was 2.8.The average Preoperative pain score assessed by visual analogue score (VAS) and anal continence assessed by Wexner score were 1.8 ± 1.0 and 2.2 ± 1.7 respectively. One or more co-morbidities were observed in 13 patients (46.4%), six (21.4%) patients had DM, 4 patients (14.3%) were hypertensive on regular treatment and 3 patients (10.7) had ischemic heart diseases. Patient's characteristics and demographics are shown in (Table 1).

Eighteen patients (64.3%) gave history of previous intervention including abscess drainage (6, 21.4%) and prior one or more fistula operations (12, 42.9%). The average length between the external fistula opening and the anal verge was 5.6 cm (range, 3–10 cm). The fistulas had been present for a median of 18 months (range 2–36 months) at the time of intervention of this study. Mild anal incontinence (soiling) was recorded in 4 patients.

In our clinical trial, a total of 28 patients (22 men and 6 women)with complex crypotoglandular anal fistulas, both primary and recurrent type, were treated by local injection of autologous freshly prepared stromal vascular fraction solution containing adipose derived stem cells after closure of internal fistulas opening with 2/0 vicryl suture with curettage and washing of all fistula tracts.

Considering all the cases treated, six weeks after first injection of stromal vascular fraction; 15/28 patients (53.6%) showed complete healing of the fistula and had a complete cessation of the discharges. Further, 4(%) additional patients noticed marked reduction in fistula discharge. Two of these 4 patients had stopped fistula discharge and had complete healing of fistula tract 12–18 weeks after initial treatment. Three of the 15 patients who had shown initial complete healing had a recurrence of fistula during the 6 months follow-up period. Hence, 14 (50%) patients of the total 28 patients had completed healing of the fistulas after the first injection of autologous ADSCs within the follow up period of this study. Perineal MRI showed complete healing of all 14 fistulas. Most of these fistulas were cured in a mean time of 10.5 weeks (range: 2–24 weeks).

Regarding the remaining 14 patients who did not achieve complete healing of their fistulas after the first infiltration of ADSCs, 10 patients agreed to receive a second injection and 4 patients were lost. Three of these 10 patients, the second injection resulted in complete fistula closure within 10 weeks, and this healing was confirmed by pelvic MRI. The healing of these fistulas was continued throughout the whole follow up period of 6 months. Further, three other patients of the 10 patients who received a second injection showed cessation of fistula discharge by 4–6 months. The average postoperative pain assessed by VAS score anal continence assessed by Wexner score were 2.4 ± 1.4 and 2.6 ± 1.2 respectively. Most patients were satisfied (mean 7.8 ± 4).

Thus, our results showed overall all healing rate of % 60.7 (17 out of 28 patients). The healing rate after first and second injections of ADSCs was 50%, 30% respectively. During the overall follow up period of 6 months of these patients who showed healing and closure of their

fistulas,(17 patients) none of them recorded relapse of the fistulas (recurrence 0%). Further, none of the patients recorded complications or adverse reactions related to stem cell therapy during the study and none of them had worsening fecal incontinence during follow-up period.

3. Discussion

The management of low and simple perianal fistulas is straightforward by fistulotomy with excellent healing and functional outcomes. However, the management of complex anal fistulas still represents a major surgical challenge with high rate of failure and poor functional outcomes The repair of anal fistulas can be challenging, largely due to the persistence or recurrence of fistulas even after several attempts of repair [3,12]. Up to 30% of fistulas persist after surgery despite advancements in treatment [13,14]. It is reported that the incidence of anal incontinence after fistulotomy of complex CPAF is up to 40% [15]. Thus, management of these complex fistulas is commonly faced by two main challenges, these are: risk of anal incontinence and high recurrence rate. Therefore, the introduction of new sphincter-sparing procedures has become an urgent necessity.

According to the American Society of Colon and Rectal Surgeons (ASCRS), complex perianal fistulas included those fistulas with more than 30% of the external sphincter, fistulas with multiple tracts including horseshoe type, anterior fistula in female patient, recurrent fistula, and those fistulas associated with fecal incontinence, Crohn's disease or radiation [16]. Seton technique (tight or loose) has not proven to be effective. Other options such as endoanal advancement has been tried as standard approach for complex fistulas but two major drawbacks are frequently encountered: fistula recurrence and anal incontinence [17]. Fibrin glue, fibrin plugs and LIFT procedure as sphincter saving treatment modalities are commonly not effective with efficacy range between 10 and 70%. Temporary fecal diversion by diverting colostomy is sometimes performed for refractory complex anal fistulas aiming to divert the fecal matter away from anorectum to control the septic process to be closed later when the fistula has healed. This approach, however, represents a big burden to the patients and has a major effect on the quality of life. Further, there is no certain that the fistulas would not recur [18].

We report the treatment outcomes of a cohort of 28 patients who presented with complex anal fistula, both primary and recurrent type, after injection of stromal vascular fraction containing ADSCs prepared from fresh aspirate of liposuction. The overall success rate over a mean follow-up time of 6 months was 60.7%. The healing rate after first and second injections of ADSCs was 50%, 30% respectively. We found that patient characteristics and co-morbidities, type the fistula, and number of prior operations were the main factor that determines the outcome success. The procedure was safe and with limited adverse effects in this cohort, with only 2 cases of postoperative infection, and no cases of fecal incontinence. Postoperative pain was mild and most patients were satisfied. The recurrence rate of those patients who showed complete healing of their fistulas after 6–12 months follow up period was zero, thus no relapse was detected.

It was found that adipose-derived stem cells can promote healing and inhibit inflammation by significantly increasing the production of anti-inflammatory cytokines such as IL-10β and decreasing the secretion of pro-inflammatory cytokines such as interleukin (IL)-1β and tumor necrosis factor (TNF) [18,19]. Adipose-derived stem cells (ADSCs) have all the properties of stem cells such as multipotency, anti-inflammatory effects, immuno-modulation, pro-angiogenesis and self-renewal, besides. ADSCs are easy to harvest and collect than other types of mesenchymal stem cells [20–22]. ADSCs can selectively migrate to the site of inflammation promoting tissue regeneration and repair through its paracrine effects, which migrates to the site of tissue inflammation selectively and reduces the degree of fibrosis by inhibiting the inflammatory response [23,24]. The main characteristic properties of an adult stem cell population to be efficient in treatment of chronic inflammation are that: the cell population must be immature, have high proliferative potential and be generally multipotent, [25] and therefore capable of regenerating its tissue of origin and lastly they are capable of self-renewal [8,26].Our findings showed that, treatment of complex anal fistulas with freshly prepared stromal vascular fraction containing ADSCs has a promising future and could be a valid and feasible treatment modality for complex and recurrent perianal fistulas of cryptoglandular origin. Our findings were consistent with the results of other several similar studies [26–29].

Zhang Y et al. [30] reported in their comparative study that the closure rate of ADSC of complex perianal fistulas treated with ADSCs group at week 12 were 54.55% without any significant adverse effects. VAS score in ADSC group was significantly lower than those patients treated by endorectal advancement flap. Wexner score of all patients was not increased with no significant differences between the two groups and they conclude that treatment of complex fistulas by ADSCs was not inferior to that of endorectal advancement flap (ERAF). Similar conclusion was reported by Garcia-Arranzet al [31]. We believed that treatment failure of some cases with ADSC in this study may be caused by inadequate control of sepsis and infection. Similar findings were reported by Choi et al. [29] and Cho et al. [32].

Management of complex fistulas by ADSCc injection is minimally invasive procedure with rapid recovery and excellent functional protection (preserved anal continence) since the procedure is not associated with tract excision and the injection of ADSCs does not injure the anal sphincter. On the other hand, our results showed that the healing rate can be further enhanced and increased by second injection of the same concentration of ADSCs with no any harm or sever adverse event. The healing rate in this study was further increased by 30% after second injection. Our findings are consistent to that of Garcia-Olmo et al. [33] who recorded in their phase II, randomized clinical trial, that the addition of ADSCs to fibrin glue was more effective than fibrin glue alone in promoting healing of some refractory complex anal fistulas and the procedure was safe and efficient and second injection of ADSCs increased the healing rate also.

Our results showed that even for those patients who didn't notice healing of their fistulas, their discharge was significantly decreased and their quality of life was remarkably improved, as it minimally invasive procedure, and the injection of ADSCs was not associated with anal distention postoperatively and almost all patients felt comfortable since there was no excessive manipulation, retraction, or excision. It is worth mentioning that autologous ADSC can avoid autoimmune rejection and ensure the safety of treatment since these cells are autologous, immature and freshly prepared. Topal U et al. [34] and Simona A et al. recorded in their similar studies that curettage of fistula tract and closure of internal fistula opening, and injection of autologuos micro-fragmented adipose tissue injection is feasible, safe inexpensive and reproducible procedure depending on early promising results in complex idiopathic perianal fistulas. The healing rate was significantly higher in experimental group received autologous centrifuged adipose tissue injection compared with control group. No major adverse effect or complications were recorded and postoperative anal pain was significantly lower in the injection group. Besides, time taken to return to daily activities was significantly shorter in experimental group. Their findings were similar to ours. Our results showed that fecal incontinence score 6 months follow up period after surgery was similar to the preoperative score. These results were consistent with other similar studies [10,30,23,and34]].

4. Conclusion

Treatment of complex anal fistula with intralesional injection of ADSCs obtained from freshly prepared lipoaspirate is feasible, safe and effective treatment modality with accepted healing rate. It is relatively painless and sphincter-saving procedure with no risk of fecal incontinence and any adverse events or complications. The second injection of ADSCs could further enhance healing rate and decrease the recurrence

rate. Administration of ADSCs for complex anal cryptglands fistulas is promising treatment modality that worth further studies and investigations.

Ethical approval

The study was approved by ethical committee of college of medicine, university of Basrah, Department of surgery. Ref. No.123/2019.

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Author contribution

The surgical and imaging aspect of this study was done by prof. dr. Ibrahim Falih Noori and dr.Ahmed Falih Noori,The tables,results and statical analysis of this study were done by.dr. Qais Khadim.

Research registration unique identifying number (UIN)

Research Registration: Researcregistry: http://www.researchregistry.com.resea **Unique Identifying Number (UIN)**:9023.Hyp Hyperlink: https://www.researchregistry.com/browse-the-registry#home/

Guarantor

The study is under the authors responsibility.

Consent

Written informed consent was obtained from the patient for publication of this randomized controlled study and accompanying images. A copy of the written consent is available for review by the Editor-in-Chief of this journal on request".

Declaration of competing interest

Authors declare no any conflicts of interest.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ijso.2023.100686.

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