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Original Article

Safety and feasibility of autologous adipose-derived stromal vascular fraction in the treatment of keloids: a phase one randomized controlled pilot trial

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Abstract: Introduction: Autologous adipose-derived stromal vascular fraction (SVF) has been described to have therapeutic benefits in the treatment of keloids. However, most of the evidence on its efficacy is based on observational studies the majority of which are conducted in high-income countries and yet the highest burden of keloids is in low- and middle-income countries (LMICs). Objectives: We set out to determine the safety and feasibility of using autologous adipose derived stromal vascular fraction in the treatment of keloids in LMICs. Methods: In this phase II randomized controlled pilot clinical trial conducted in the Plastic Surgery Unit of Kirruddu National Referral Hospital in Kampala Uganda, 8 patients were assigned a 1:1 ratio to either SVF or triamcinolone acetonide (TAC) arms. In the SVF arm, a median (Inter quartile range) amount of stromal cell infiltration of 2.7×10^6 (11×10^6) was administered, while the controls received 10 mg/ml TAC at a ratio of 1:1 TAC to keloid volume. Primary endpoints were adverse event development based on the Common Terminology Criteria for Adverse Events (CTCAE) v5.0 tool and feasibility assessment based on ≥ 70% recruitment feasibility and ≥ 80% interventional feasibility rates. Results: The participants' mean age was 27.9 (±6.5) years, with a female predilection of 5 (63%). Overall, no adverse events were reported in the SVF arm, while ulceration in a single patient in the TAC arm, which was a grade II adverse event, was reported. Recruitment feasibility of 80% and interventional feasibility with 100% completion were reported. Conclusion: Based on our findings, an autologous adipose-derived stromal vascular fraction is feasible and safe for the treatment of keloids in LMICs.

Keywords: Stromal vascular fraction, adipose stem cells, keloids, scars, low middle-income countries

Introduction

Keloids are one of the most common benign skin disorders [1], occurring mostly in individuals with coloured skin, especially among Africans. They are estimated to affect up to 16% of the African population [1, 2]. To date, there is no curative treatment for these keloids, as all existing therapies have varied treatment responses and invariably result in recurrence [3-5].

Several therapies for managing keloids exist of which the most widely used is Triamcinolone acetonide (TAC). TAC is a synthetic glucocorticoid with anti-inflammatory properties and is traditionally used either as monotherapy or in combination with other therapies such as surgical excision, cryotherapy often with significant treatment response [6].

Other therapies used in Keloid management such as 5-Fluorouracil, Verapamil, radiothera-

py, Silicone gel among others have been described globally being used in combination or in isolation and with varying dosing strategies [7-9]. These are however not routinely used in Uganda or other Low-income countries.

All the above therapies have been described to have varying degrees of satisfactory response with the majority described to result in keloid volume regression ranging from 30 to 100% [6, 10]. Despite the majority of therapies reporting more than 50% satisfactory volume regression for keloids [11, 12], Triamcinolone acetonide (TAC) is the most widely used therapy in keloid management especially in LMICs. This is partly due to its low cost, universal availability and ease of administration. It can either be used as single or combination therapy [13, 14]. Despite its efficacy in causing keloid volume regression, the biggest limitation of Triamcinolone and the other existing therapies is the high keloid recurrence rates which range between 20 to 100% following successful treatment [10, 14-18]. To compound this limitation, optimal treatment responses for TAC often require multiple sessions of therapies often on a monthly basis with no predefined limit but until the desired outcome is obtained. This coupled with the pain associated with these injections makes TAC are less desirable treatment for keloids.

Because of the high recurrence rates associated with the existing therapies coupled with the several and cumbersome treatment sessions required to achieve the desired treatment outcomes, there is a need to explore other therapies that can augment the existing therapies or provide entirely alternative options that maybe more tolerable.

Autologous adipose derived stromal vascular fraction (SVF) is one new promising therapy that is increasingly being recognised to have therapeutic potential in treating keloids and hypertrophic scars.

Human adipose tissue which is usually obtained through liposuction contains vast amounts of cells collectively referred to as stromal cells or the stromal vascular fraction (SVF) [19]. This heterogenous cell population contains mesenchymal stem cells, pericytes and other cell precursors as well nucleated cells [20]. Characteristically, these cells exhibit multi-lineage differentiation potential and are therefore described to be a rich source of adult stem cells very similar to those obtained from bone marrow [21]. Unlike bone marrow aspirate, the lipoaspirate is a lot easier to extract and provides are richer and yet similar quality of mesenchymal stem cells [19, 21]. Through either enzymatic or mechanical processing, the cells in the adipose tissue are extracted and concentrated through centrifugation [19, 22].

The SVF cells obtained after processing the fat have been demonstrated to possess immense therapeutic and regenerative properties [20, 22]. These cells promote wound healing and hence used in the treatment of chronic wounds [23, 24], arthritis [25], neurological diseases [26] among other conditions. Specifically, SVF has been described to improve scar outcomes [27-30] with mouse models demonstrating significant reduction in the hypertrophic scar volumes.

The SVF cells have been described to exert their therapeutic effects through paracrine based signalling mechanisms that modulate excess inflammatory processes [31] as well as suppression keloid fibroblast proliferation and gene expression [20, 32-34]. Specifically in scar therapy, SVF is described to inhibit scar fibrosis through the suppression of the p38/MAPK signalling pathway [35] as well as fibrosis inhibition via the Transforming growth factor-beta (TGF-β) pathway [36]. This results in SVF mediated inhibition of collagen deposition through the suppression of the col1, col3 and α-SMA (Smooth muscle actin) genes [35, 37]. This is described to result in faster healing with more organised scar tissue.

The SVF is constituted either as a point of care therapy or it's sent to the laboratory for culture expansion to yield more cell volumes [38, 39].

Despite this growing evidence of the effectiveness of SVF in scar healing, the majority of studies have a low level of evidence with paucity of well-organized clinical trials to compare the efficacy of SVF to existing therapies. Similarly, most studies are conducted in high income countries and yet the biggest burden of keloids is in Low-income countries.

In order to benefit from this potential therapy, there is a need to understand the feasibility of

utilising this therapy as well as describe the safety of the therapy in a low-income country setting.

Without designed randomized controlled clinical trials, it is difficult to objectively determine the benefits of SVF therapy in scar management and this may subsequently deter future potential utilization as standard therapy. Point of care stromal vascular fraction is a lot cheaper and safer to use as the cells are processed and used at the same sitting. Utilization of the adipose-derived stromal vascular fraction is highly technical and demanding, and before conducting a clinical trial to assess efficacy, it is important that a pilot study be conducted.

In this study, we assessed the feasibility of conducting a clinical trial using the stromal vascular fraction in keloid treatment. Second, we evaluated the safety of using the SVF in comparison to standard triamcinolone acetonide (TAC).

The findings of this pilot study will be used to develop a phase II clinical trial that will evaluate the efficacy of using the stromal vascular fraction in the treatment of keloids.

Materials and methods

Trial design

This was a parallel group single centre randomized controlled pilot trial with a ratio of 1:1 conducted at the plastic surgical unit of Kirruddu National Referral Hospital in Kampala, Uganda, from March to July 2021. The trial was approved by the "The AIDS Support Organization (TASO) Research Ethics Committee (TASOREC/060/19-UG-REC-009) and the Kirruddu National Referral Hospital Research Ethics Committee and was also registered in the ClinicalTrials.gov under the registration number NCT04553159". The trial reporting followed the CONSORT Extended checklist for pilot clinical trials [40] and TIDier guidelines [41].

Eligibility criteria

We included patients with a single keloid of ≤ 4 cm^3 as these have the highest response to any treatment administered. Patients had to be between the age of 18-65 years. Participants were excluded if they had are BMI less than 18.5. As these would have an insufficient fat pad. Participants who had received an intralesional steroid injection therapy or radiotherapy in the three months prior to the study were also excluded. Any participant with an active or ongoing systemic illness demonstrated by the presence of are fever or comfirmatory laboratory results were also excluded. Likewise patients with ulcerated keloids or infected keloids were also excluded from the study.

Included participants were provided with complete oral and written information ahead of their consent on the clinic visit. Informed consent was then obtained on the scheduled day of the procedure usually not less than 2 days from the booking date.

Intervention

Once participants were included in the study, baseline characteristics were obtained. The participants were then randomly allocated into one of the two treatment groups. Group 1 was the group that received the SVF while group 2 received the Triamcinolone Acetonide as described below.

The autologous adipose-derived stromal vascular fraction (SVF) group

Participants in this arm received a single dose of intralesional infiltration of the autologous adipose-derived stromal vascular fraction. The dosage depended on the total number of viable SVF cells in the cellular suspension and was expressed as total viable cells per ml of suspension. The final cell suspension volume was constituted in a 1:1 ratio of the keloid volume with 1 ml of cell suspension per cubic centimetre of keloid tissue.

To obtain the stromal vascular fraction, the steps described below were followed.

Harvesting adipose tissue: Tumescence liposuction was performed aseptically on the outer thigh following the infiltration of 300 ml of tumescent solution (constituted as 20 ml of 2% lignocaine, 1 ml of 1:1000 epinephrine, 12.5 ml of 8.4% sodium bicarbonate in 1 litre of normal saline). Through a 3-4 mm skin incision, liposuction into a 10 ml Leuer lock syringe attached to a 3 mm Coleman liposuction cannula was performed with 100 to 150 ml of

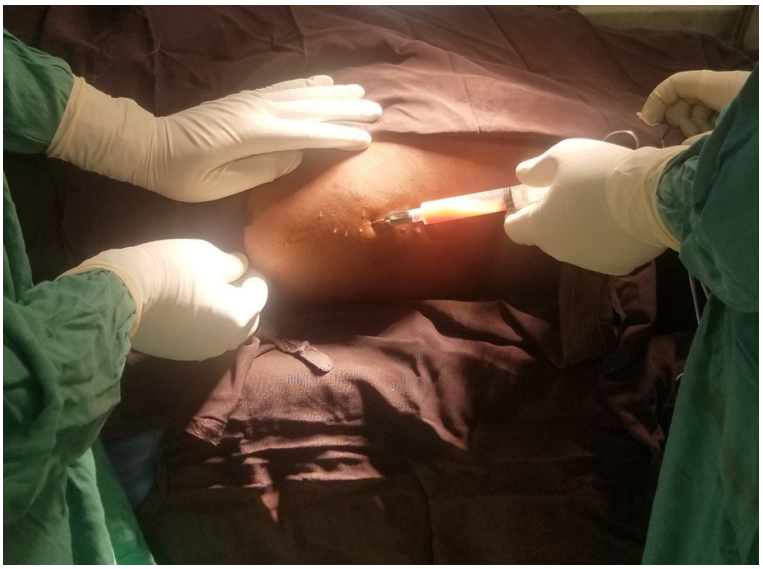

Figure 1. Liposuction process being aseptically performed.

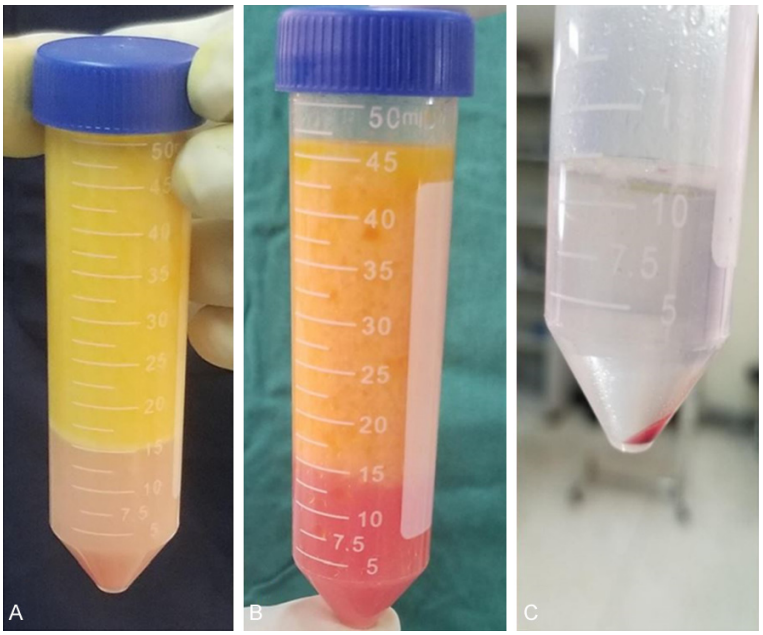

Figure 2. Lipoaspirate processing to obtain the stromal vascular fraction. A. The lipoaspirate after centrifugation. B. Digested lipoaspirate following 0.075% collagensase incubation. C. Stromal vascular fraction pellet (SVF).

lipoaspirate and collected into 50 ml sterile Falcon centrifuge tubes (see **Figure 1**). The liposuction cannula entry incision was closed with a 6/0 Monocryl absorbable suture, and a pressure bandage was placed.

Standard operation theatre aseptic protocols were followed, and the procedures were conducted in a dedicated plastic surgery operation theatre by qualified plastic surgeons.

Extraction of stromal vascular fraction: The harvested lipoaspirate (see **Figure 2A**) was processed aseptically from a designated sterile unit in the operating theatre. The lipoaspirate was washed using 1X Dulbecco's phosphate buffered saline-PBS (Lonza, Walkersville, MD, USA) and then subsequently enzymatically digested using 0.075% Type 1A collagenase (MERCK Millipore, USA). Enzyme stop media comprising 10% foetal bovine serum-FBS (Sigma St. Louis, MO, USA) in Dulbecco's modified Eagle's medium (DMEM)-high glucose (Sigma St. Louis, MO, USA) was used to neutralize the enzymatic process (see **Figure 2B**). The stromal vascular fraction pellet (**Figure 2C**) was subsequently incubated in 10 ml of red cell lysis buffer (Sigma St. Louis, MO, USA) at room temperature for 10 minutes and later washed in 1X PBS. Through a 100 µm nylon cell strainer (BD Falcon, NJ, USA), the mixture was filtered to remove any unwanted tissue debris. Centrifugation at 1200 g resulted in the stromal vascular fraction (SVF) pellet, which was then resuspended in 1.5 mL of Ringer's lactate solution.

To determine the cell count and viability, 10 µl of the cell suspension was added to an equal volume of 0.4% Trypan blue (Sigma St. Louis, MO, USA) and then mounted into a Neubauer counting chamber as per Strober's guidelines [42]. Cells were counted at 40× magnification, as shown in **Figure 3**.

Total nucleated cells per 10 µl were used to determine the final cell dosing, while viability was determined using the Trypan blue exclusion test [42]. The total number of viable cells

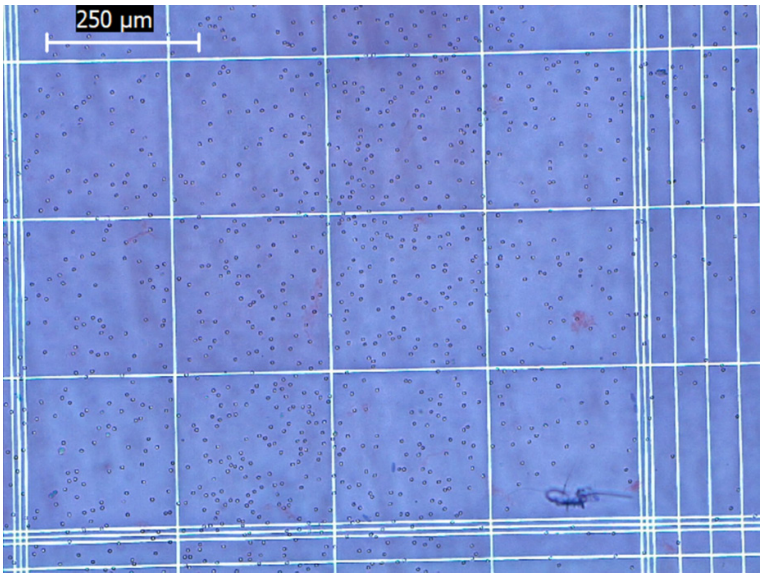

Figure 3. Stromal vascular fraction cells following staining using Trypan blue ready for cell counting. Cell suspension mounted on a Neubauer counting chamber and cells counted at ×40 magnification (Scale bar of 250 µm).

Figure 4. Infiltration of the stromal vascular fraction into keloid tissue.

in the Neubauer counting chamber was used to calculate the total number of cells in the 1.5 ml suspension factoring in the two times dilution factor during trypan blue staining.

The stromal vascular fraction dosing was constituted by diluting the SVF suspension to a 1:1 ratio of SVF volume to keloid volume. Hence, for a 4 cm^3 keloid, 1.5 ml of original SVF suspension was constituted to make 4 ml by the addition of 1X phosphate buffered saline (PBS). The final infiltration cellular dosing calculation per millilitre was computed, and medication was placed into hypodermic syringes ready for infiltration.

The triamcinolone acetonide (TAC) group

For this arm, one ampoule of triamcinolone acetonide (TAC) containing 40 mg in 1 ml was used for each patient. To each ml of triamcinolone, 1 ml of 2% lidocaine and 2 ml of water were added for injection to constitute 4 ml of TAC suspension at a concentration of 10 mg per ml. The suspension was placed in hypodermic syringes for subsequent infiltration into the keloid tissue. For each patient, a maximum of 40 mg of triamcinolone could be infiltrated into the keloid under study.

The standard dosing for triamcinolone for keloid volume was described by Rahban and Ganner [43].

Injection of the SVF and TAC into the keloid

The selected keloid was prepped with 10% povidone iodine and then appropriately draped.

Infiltration into the keloid depended on the keloid morphology, volume and shape.

Each keloid, where applicable, was divided into four quadrants, and the infiltration dose was equally divided into the four quadrants. An infiltration volume of 0.1 ml per cubic millimetre or 1 ml for every cubic centimetre was targeted. This was based on estimates from Rahban [43] and Rables [44] studies. See **Figure 4**.

In some instances, the keloid tissues were thick, and it was impossible to enable infiltra-

Table 1. Common terminology criteria for adverse events (CTCAE) v5.0 tool

Grade		
Grade 1	Mild	Mild symptoms, no intervention required.
Grade 2	Moderate	Minimal or local non-invasive intervention needed.
Grade 3	Severe or medically significant	Severe but not immediately life threatening. Requires hospitalization, disabling.
Grade 4	Life threatening consequences	Urgent intervention indicated.
Grade 5	Death	

tion of the appropriate dosage. In such cases, 'needling' was performed. 'Needling' is when a 22-gauge needle is used to create a mesh/network of interconnecting tunnels 2 mm apart into the keloid. Following this, infiltration of the treatment was instituted into the created tunnels.

General trial procedure

Patients who were included and consented to the trial underwent standard preoperative care, including informed consent for surgery. The research assistants enrolled the participants, obtained baseline characteristics, and then randomized and allocated them into either treatment arm. The intervention was administered by a qualified surgeon who had previously been trained on the standards and practices of the procedures. Following the procedure, postprocedural analgesics were provided to participants based on the hospital guidelines for day-care surgery. At follow-up, patients were reassessed clinically at the plastic surgical clinic. The reviews were scheduled at one week and one month while a final follow-up at the end of three months upon which the participants exited the study. The project principal investigator undertook the follow-up reviews. Participants were provided with a transport refund during the follow-up visits.

Outcome

The primary outcome variables were safety and feasibility of using autologous adipose-derived stromal vascular fraction in the treatment of keloids in comparison to Triamcinolone Acetanoide.

Safety

Adverse events were defined based on the Common Terminology Criteria for Adverse Events (CTCAE) v5.0 tool as any unfavourable and unintended symptom, sign or disease associated with the use of a medical treatment or procedure that may or may not be directly an intended result of the procedure. These adverse events experienced by the participants were documented based on CTCAE v5.0 [45] and were reported on a running daily basis for the entire study period. Specific assessments for adverse event development were conducted on day 0 (immediate post procedure), day 1, days seven, one and three months. The participants were asked to spontaneously report any adverse events to the study principal investigator through a phone call at any time during the follow-up period, and the principal investigator would then follow-up with the necessary intervention.

Adverse events were graded as seen in **Table 1**, and the adverse event grade at each assessment time was recorded. The adverse events looked out for included.

Donor site adverse events: Among patients in the SVF arm. The development of haematoma, bleeding, infections and pain, delayed wound healing, and keloid development at the liposuction site. The nature and severity of the adverse events were graded using the CTCAE v5.0 tool.

Infiltration/recipient site adverse events: In both arms, the keloid and surrounding skin, adverse events were observed, including bleeding, excessive pain, infection, and ulceration. These were also graded using the CTCAE v5.0 tool.

Feasibility

The criteria for feasibility were based on four parameters that had to be met for the procedure to be considered feasible. All four parameters had to be feasible for the procedures to be classified as feasible.

Recruitment feasibility: This referred to the enrolment and acceptability to participate

among those who were found to be eligible. Obtaining consent in 70% of the first 10 respondents was described as feasible for recruitment. For those who declined, reasons for the decline were sought out.

Intervention feasibility: As a day-care procedure: Here we looked at two variables which were the procedure time and the procedure completion rates. The procedure time was defined as the total procedure time it took the whole procedure to be performed in hours. Durations of equal to or less than 5 hours were described as feasible. The procedure completion rates and general anaesthesia conversion rates: The procedure was intended to be conducted under local anesthesia with optional opioid analgesia addition. Completion, as described above without the need for conversion to general anaesthesia or procedural abandonment, was described as feasible.

Suitability of outcome measurements: We intended to describe the keloid thickness measurement as are key outcome measurement. Keloid height/thickness was determined in millimeters using high-frequency ultrasound (Healcerion SONON portable ultrasound model 300 L). This ultrasound scan device has a capacity for length measurement with up to two decimal point accuracy. Suitability was described as the interobserver ability to consistently reproduce three ultrasound thickness measurements at the same site with a variation of ≤ ±0.03.

The feasibility of assessing the primary outcome measures of the main trial of Patients and Observer Scar Assessment Score (POSAS) and keloid volume calculation were also assessed.

Completion rates and challenges in assessing each of these outcomes were evaluated.

Follow up feasibility: We also assessed the feasibility of following up the participants. Specifically, we looked at the assessment for co-intervention, appropriateness of timing for the next intervention. In the first case, participant utilisation of any other treatment during the follow-up period, the timing of use and the reason were determined. For the appropriateness of timing for the next intervention, the study follow-up of three months was used. The appropriateness of this follow-up was evaluated to establish the optimal timing before subsequent follow-up therapy. This was based on the duration of symptom remission or symptom-free duration.

Secondary outcomes

Clinical outcomes were the mean change in the Patient and Observer Scar Assessment Scores (POSAS) from baseline to one month and three months.

Monitoring treatment fidelity

Because this was the first time this kind of study was being conducted, it was important that treatment fidelity be monitored. Specifically, fidelity adherence checklists were developed, and each key step in the study implementation was compared to the described protocol to evaluate the degree of deviation. Deviations from the protocol were identified, and the reasons were reported. For each procedural step, the investigator was asked to rate how compliant they were in keeping with the trial protocols. Second, standardization training of the participants and research assistants was performed to ensure consistency in trial procedures.

Sample size estimation

This was a phase one study intended to explore the safety, feasibility and refine the trial process and not establish an effect. Therefore, hypothesis testing to establish a sample size was not necessary. This was based on the recommendations of Arain [46].

To establish the sample size, we used the formula by Lackey et al where they proposed that the pilot study sample size comprised 10% of the main trial size. Based on this recommendation, a pilot sample size of 8 participants, which is 10% of the main clinical trial sample size, was established with 4 participants allocated to each arm.

Randomization and allocation concealment

Randomization was performed using STATA command ralloc by a statistician who was not directly involved in the study. Block randomization using block size 2 was used to allocate patients following a 1:1 ratio.

Sequentially numbered opaque envelopes were used to conceal the allocation sequences, and these were prepared by the same statistician that generated the randomization. The envelopes were stapled and handed to the recruitment nurse who only interfaced with the patients during the allocation of treatment the day of the procedure and did not interact with the enrolling research assistant.

At the time of allocation into the different study arms, the recruiting nurse would detach the sequential envelope to identify the allocation and subsequently keep the envelope.

Blinding

The nature of the study procedure made it impossible to blind the patients and the surgeons, as liposuction instantly revealed which arm the patient was allocated. The outcome assessors, on the other hand, who followed up patients for review were blinded, as they did not know what intervention the participants had received.

Statistical methods

Data analysis for the participant demographics, baseline characteristics, safety, and feasibility took on a descriptive approach with all data being exported and analysed in STATA 15.0.

Continuous data are reported as the means (± SD), while categorical data are reported as proportions with their percentages.

Categorical data analysis was performed for the first primary endpoint of safety. Proportions with the percentages of the different Common Terminology Criteria for Adverse Events (CTCAE) v5.0 grades were obtained. Broadly, two categories of "*adverse* events" and "*no adverse events*" were analysed and compared between the two treatment arms using chi square tests.

For the second primary endpoint of feasibility, variables were categorized into feasible or not feasible, and the proportions were determined. Comparison between the two treatment arms was performed using chi square tests.

For the secondary outcomes, efficacy endpoints were merely exploratory; therefore, analytical assessments were not performed. These were continuous variables and were reported as the means with standard deviations. We evaluated for the presence of mean differences in the two treatment arms at one week, one month and three months and at baseline.

Patient & public involvement

Patients and participants were not involved in the development of the research question, choice of outcome measures, design of the trial, recruitment of participants or conduct of the trial. The results of the trial were disseminated to the study participants through direct consultation.

Results

Characteristics of the study participants

Eight participants were recruited during the months of March and April 2021 and were followed up for three months with the last follow-up taking place in July 2021.

Their mean age was 27.9 (±6.5) years, while five of the participants were female.

Despite the randomization, all the participants that were allocated into the intervention arm were female, while one female was allocated into the control arm. The mean BMI was 27.9 (±6.0), with a BMI generally higher among the intervention arm. The mean keloid duration was 4.4 (±5.5) years, and none of these participants had received any therapy in the last three months. Details of the keloid distribution can be found in (**Table 2**).

In the SVF arm, participants selectively preferred the liposuction point to be the outer thigh out of three options of the abdominal, inner thigh, and outer thigh. The mean infiltrated volume of tumescence fluid was 387.5 (±114) ml, with a mean lipoaspirate volume of 137.5 (±37.7) ml.

The total cell counts among the participants were found to be skewed. Particularly one participant had an outlier value. Because these data were not normally distributed, we used medians with their interquartile ranges (IQRs) instead of means with standard deviations.

Table 2. Baseline characteristics of study participants

Participant	Age	Sex	BMI	Location of keloid	Duration of keloid (years)	Treatment intervention
001	24	Male	23.3	Sternal	1	TAC
002	26	Female	32.4	Sternal	1	SVF
003	24	Female	35.3	Sub-mandibular	18	SVF
004	39	Female	35.9	Epigastric	2	SVF
005	25	Male	19.8	Sternal	6	TAC
006	20	Female	21.6	Earlobe	5	SVF
007	25	Male	23.9	Lateral infrapatellar	2	TAC
008	36	Female	31.2	Breast	1	TAC
Mean	27.9 (±6.5)	NA	27.9 (±6.0)	Not applicable	4.4 (±5.5)	NA

Table 3. Characteristics of participants in SVF arm

No.	Tumescence fluid infiltrated	Lipoaspirate volume	Processed lipoaspirate	Total viable cell count harvested	Infiltration volume (ml)	Infiltration dose cells/ml	Cellular viability
002	*500*	200	110	4.5×10^6	5	9×10^5	98
004	500	130	70	33.6×10^6	3	11.2×10^6	98
004	300	120	57	6×10^5	3	2×10^5	85
007	250	100	52	8.4×10^5	1.5	5.6×10^5	95
Mean (±SD)	387.5 (±114)	137.5 (±37.7)	72.3 (±22.8)	9.9×10^6 ($\pm13.8\times10^6$)	3.25 (±1.1)	3.2×10^6 ($\pm4.6\times10^6$)	94 (±5.3)
Median (IQR)	400 (212.5)	125 (32.5)	63.5 (24.3)	2.7×10^6 (11×10^6)	3 (0.75)	7.3×10^5 (3×10^6)	96.5 (5.5)

Following processing, the mean total number of viable stromal cells in the entire lipoaspirate was estimated to be 9.9×10^6 ($\pm13.8\times10^6$) cells with a mean dosing of 3.2×10^6 ($\pm4.6\times10^6$) cells/ml of infiltrating suspension. The median total number of stromal cells was 2.7×10^6 cells with an interquartile range of 11×10^6 cells. Overall cellular viability was reported at 94%. For details see **Table 3**.

Comparison of safety profile

On the day of intervention, there were no adverse events that were reported, and all patients were subsequently discharged as had been intended. There were still no adverse events at the 24-hour follow-up. Overall, no serious adverse events were recorded. One participant in the triamcinolone group reported the development of an ulcer at the infiltration site that developed on day five following the intervention. This required administration of oral antibiotics and analgesics and a topical antibacterial cream but no surgical intervention or hospitalization. Subsequently, the ulcer healed by the second week of follow-up. Among the SVF group, there were no reported adverse events in the entire follow-up period.

Feasibility of adipose-derived stromal vascular fraction

Overall, both treatments were feasible with comparable differences.

Recruitment feasibility

Among the first 8 eligible participants who were reached out to participate, two declined to participate in the study after they had received information about the clinical trial. This prompted us to reach out to the next two eligible participants who agreed to participate in the study. Based on this, overall, recruitment feasibility stood at 80%. The reason for the decline was the unwillingness to have another site away from the keloid operated upon, while the second respondent strictly wanted to undergo surgical excision.

Intervention feasibility

Duration of procedure: On average, the procedure time for triamcinolone was 30 minutes to one hour, while that of the SVF took a mean time of 5 hours. All interventions were successfully conducted as day-care procedures, and no patient required hospitalization.

Procedure completion rates: All intended procedures had a completion rate of 100%. This means that no procedure was abandoned once it had been started. Second, there was no conversion to general anaesthesia, as all patients tolerated the procedure.

In the SVF arm, one patient needed supplemental opioid analgesia and was given IV fentanyl 50 mcg, which sufficiently controlled the break-through pain during the liposuction process.

Suitability of outcome measurements

Keloid thickness: Two independent radiologists performed cutaneous ultrasound to determine keloid scar thickness. The interobserver variability was within the stipulated margin of ±0.03.

Surface area estimation using photography when compared to surface area mapping using tracing paper was found to be more unreliable with variability in establishing focal length and angles. Surface area mapping using tracing paper proved a more reliable measurement estimate.

Follow up feasibility

Co-intervention: All patients but one did not seek additional therapy during the first two months of therapy. The patient who developed the ulcer had to seek extra therapy, and she received a topical antibacterial cream.

Secondary outcome: All participants' Patient and Observer Scar Assessment Scores (POSAS) improved (reduced) by a mean of 12.5 (±7.2) and 7.6 (±3.1) at one month and three months, respectively, when compared to baseline. The one-month mean difference in the improvement of POSAS scores between SVF and TAC was 14.8 (±8.6) and 10.3 (±4.6), respectively. At three months, the mean difference in POSAS scores when compared to baseline was 7.8 (±3.3) and 7.5 (±2.9) in the SVF and TAC arms, respectively. All patients in all arms reported resolution of itching and noted scar softening.

Treatment fidelity: To ensure adherence to treatment fidelity, all study staff were trained on strict adherence to the research protocol. In all participants in both arms, adherence to the protocol was followed.

Discussion

To date, there a few clinical trials comparing autologous adipose-derived stromal vascular fractions to other keloid therapies with the majority of existing studies being observational [27, 47-49]. Secondly, most published studies in high-income countries cast uncertainty on the feasibility of conducting similar trials in low- and middle-income countries, where the greatest burden of keloids is found [47, 50]. To the best of our knowledge, no clinical trial has been published, particularly in sub-Saharan Africa exploring this therapy.

This trial therefore provides critical information on the feasibility of conducting this study in a resource-limited setting in addition to describing its safety while also comparing it to the widely used therapy of triamcinolone acetonide.

With the broader goal of conducting a phase II clinical trial to evaluate the efficacy of SVF in keloid treatment, a preliminary phase I trial was required to provide preliminary feasibility and safety data.

Feasibility of SVF

To appropriately determine the feasibility, key steps of the study were identified and evaluated. As a new therapy, the willingness of participants to enrol was unknown, yet this would fundamentally determine the success of the trial. Therefore, recruitment feasibility was assessed. With recruitment feasibility at 80%, the pilot scored well above the minimum feasibility score of 70%. Among the two participants who declined, one had previously been scheduled for surgical excision and had missed surgery and therefore opting for the earlier treatment plan. The second patient expressed her scepticism about being involved in an entirely new therapy that had not been practiced in the country earlier. This recruitment feasibility rate is comparable to studies conducted elsewhere where SVF use is described as feasible [47, 51].

For interventional feasibility testing, key technical aspects of the procedures were assessed, including completion rates. All treatment arms passed the set interventional feasibility test. Specifically, for SVF, a five-hour duration enabled the procedure to be performed as a day care procedure. This was comparable to other

studies with treatment durations of 4 hours [52] and three hours [51]. In one study by Karina, a shorter time of 2-3 hours excluded the infiltration and observation time. The fact that no patients required conversion of anaesthesia to general and the completion rates of 100% clearly demonstrated the feasibility of the trial procedure, especially as a day care therapy. Other centres similarly conduct SVF as a day care procedure with the use of tumescence liposuction [51, 52].

Safety

For the safety of the procedure, overall, patients who received the SVF intervention did not report any adverse events during the entire study period. These findings are in keeping with reports described elsewhere [51, 53, 54]. According to one systematic review by Gentile, there were no particular adverse events associated with SVF treatment in scar tissues [55] besides the pain that comes with the procedure. Overall, SVF therapy for intralesional scar treatment is safe. In contrast, the Triamcinolone arm registered one case of a grade II adverse event. Ulceration following infiltration of TAC has been described and is a well-known side effect [56]. The development of this adverse event highlights the limitation of TAC as the most used keloid treatment. This forms a justification for the need for alternative therapies.

In both treatment arms, there were improvements in the POSAS scores at both one month and three months. The difference was most marked at one month, with the decline in scar improvement possibly explainable by the waning treatment effect. All patients in both arms described symptom resolution of itching in addition to softening of the scars. However, despite the promising improvement in POSAS scores, this study was neither intended nor powered to draw any such conclusions on efficacy, as this is the primary objective of the anticipated phase II trial.

Strengths and limitations

One strength of this trial is that patients were randomized to minimize bias. Second, the real-time evaluation of treatment fidelity ensured adherence to developed protocols. The description of the SVF arm as feasible is a strength of this study, as it forms the foundation for conducting a phase II clinical trial. This is the first randomized clinical trial evaluating stromal vascular fraction in sub-Saharan Africa and will play a role in advancing practice on the continent.

Some limitations in our study included the allocation of one gender into the intervention arm despite block randomization. This was due to the small sample size as well as predominantly female attendance in the plastic surgical clinic. This may be due to the cosmetically disfiguring nature associated with keloids. The gender skew limited the SVF characterisation among the males. This, however, will guide the randomization of the phase II trial with specific incorporation of stratification for sex during the randomization process.

Conclusion

Based on our findings, autologous adipose-derived stromal vascular fraction is feasible and safe as a therapy in the treatment of keloids. The trial demonstrates that SVF therapy can be safely performed as a day care procedure. Although not designed to assess efficacy, the trial describes promising improvement in the POSAS scores and symptom relief for all participants during the follow-up time.

Recommendations

Based on this pilot trial, we recommend that a phase II randomized controlled trial comparing efficacy in SVF and TAC be conducted.

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Disclosure of conflict of interest

None.

Abbreviations

CTCAE, Common Terminology Criteria for Adverse Events; POSAS, Patient and Observer Scar Assessment Score; SVF, Stromal Vascular Fraction; TAC, Triamcinolone Acetonide.

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