

AUTOLOGOUS PLATELET-RICH PLASMA VERSUS SETON INSERTION FOR HIGH PERIANAL FISTULA TREATMENT: COMPARATIVE STUDY

By

EMBABY ADEL AHMED EID¹, MOHAMMED KAMAL ALDIN ELHADARY² AHMED HASSAN EL TAWDY³, MOHAMMED ADEL ELSAYED⁴

And AHMED HASSAN SOLIMAN⁵

^{1,3}Department of Surgery, Military Medical Academy, Cairo 11291 and Department of General Surgery^{2,4,5}, Faculty of Medicine, Suez Canal University Faculty of Medicine - Suez Canal University, Ismailia, Egypt (*Correspondence: em-papy_2005@yahoo.com

Abstract

The study compared outcomes of autologous platelet-rich plasma and loose Seton insertion in treating high perianal fistulas and demonstrated the effectiveness and safety.

This was conducted from October 2017 to February 2023 as a prospective randomized controlled clinical trial for patients suffering from high perianal fistula managed in department of surgery, Suez Canal Hospitals and department of surgery, Armed Forces Military Hospitals. A total of 102 patients were divided into two groups of 51 patients each. GA was treated by Loose Seton insertion and GB was treated by applying Autologous Platelet Rich Plasma (APRP) into fistulous track. Patients were assessed with clinical examination, MRI fistulogram, or endo-anal U/S on follow-up if needed. Patients were followed every two weeks and until six months post-operatively. The primary outcome measure was the fistula-healing rate (complete and non-healing) with secondary outcome measures assessing fistula recurrence, continence status, quality of life, and pain scores.

The results showed that overall healed cases were 69/102 (67.6%) during follow up, which was 37/51(72.5%) underwent Loose Seton insertion and 32/51(62.7%) for APRP injection with little higher healing rate in loose Seton group, without significant differences (P 0.397).

Keywords: High anal fistula, Loose Seton, APRP, Endoanal U/S. MRI Fistulogram.

Introduction

The term "perianal fistula" refers to an abnormal tract that runs from the anal canal's opening to the skin's surface. Males are two to four times more likely than females to be impacted by this condition, which affects about 10 in 100,000 persons (Khera *et al*, 2010).

Perianal fistulizing illness has a poor influence on quality of life due to its association with localized pain, discharge, and significant morbidity, including the destruction of the sphincter and perineal tissue (de Groof *et al*, 2016).

Approximately 40–50% of patients treated with the incision of an acute ano-rectal abscess will develop or have an underlying anal fistula (Vasilevsky and Gordon, 1984).

Historically, surgeons used digital rectal examination and anesthesia-induced examinations to detect fistulous tracts and their internal openings. However, this technique

frequently fails to recognize complex fistulas and their branches, resulting in incorrect classification and insufficient therapy (Sharma *et al*, 2016).

MRI is the preferred imaging technique for detecting perianal fistulas due to its 100% sensitivity and 86% specificity (Sing *et al*, 2014). Because it may delineate the extent and secondary implications of the fistula tract as well as identify anal and cutaneous openings and accompanying abscesses, MRI is therefore very helpful in preoperative planning. In 12 of 56 patients (21%), it is proposed that careful preoperative review of the MRI findings may provide additional critical information for surgery (Baluret *et al*, 2019).

Endo-anal ultrasound also gives excellent imaging of the internal and external sphincters, the inter-sphincteric plane, the position of the internal orifice, the rectal wall, and

the flexibility of the muscles, all of which are necessary for surgical techniques that reduce the risk of incontinence (Pellino *et al*, 2014).

The treatment of high anal fistulas needs to strike a balance between cure and continence. There are many surgical treatment options available for high anal fistulas. The ideal surgical operation for high anal fistulas is difficult to define because they have varying cure and incontinence rates. Two main groups of surgical techniques; involved laying open techniques and sphincter preserving techniques and a colorectal surgeon should become familiar with various new techniques for treating an anal fistula.

First treatment (laying open techniques) achieved higher rates of healing, but leads to higher rates of fecal incontinence and other complications. In the second treatment which involves sphincter-preserving techniques, various approaches have been proposed for the conservative treatment of fistulas that affect a large fibers volume within sphincter apparatus, based on sealing the fistula with materials such as fibrin (Van der Hagen *et al*, 2011), stem cells (Fathallah *et al*, 2021), or bioprosthetic plugs (Corman *et al*, 2008).

The present study aimed to evaluate efficacy of loose-Seton placement as compared to local application of APRP for high perianal fistulas.

Patients and Methods

In this study, autologous platelet-rich plasma (APRP) was used and obtained cure rates that were compared to Seton insertion cure rates. The study was carried out on 102 Egyptian patients suffering from high perianal fistula. Patients were managed in the department of surgery at Suez Canal Hospital and the department of surgery at Armed Forces Military Hospital.

Study design: The study was conducted between October 2017 and February 2023 as a prospective randomized controlled clinical trial study, which included 102 patients divided into two groups, each with 51 patients.

The local Surgery Department Ethics Committee granted its ethical approval after the International Declaration of Helsinki (2008) Patients who fulfill the eligibility criteria will be subjected to: History of the present complaint, Operative details of the previous operation, Clinical examination, Investigations which will include (Routine laboratory investigations that include, Complete blood count, Renal function tests, which include Urea and Creatinine levels, Liver function tests, which include AST and ALT levels, INR, PT, PTT), and Radiological investigations, which include End anal ultrasound or MRI.

Inclusion criteria: 1- Male and female patients ages ranged from 18 to 60, 2- Patients with high perianal fistula (inter-, trans-, supra-, or extra-sphincteric fistula), 3- Patients with a history of anal surgeries or trauma, but without affected continence anal canal, 4- Patients fit for anal operations and accepted by anesthetic team, 5- Agreed to sign the consent, & 6- Absence of any exclusive criteria.

Exclusion criteria: 1- Unfit for operations (impaired renal function, respiratory problems, and cardiac disease affecting its functions), 2- Patients with recent anal surgeries less than 6 months, 3- Advanced anal carcinoma, 4. Patients are approved to have inflammatory bowel diseases (Crohn's disease or ulcerative colitis), 5- Patients under the age of 18 and over 60 years, 6- Patients with known abdominal malignancies, 7- Patients refuse to undergo surgical intervention or sign consent, & 8- More than 75% of patients had anal sphincter injury defects.

Exclusion criteria: 1- Patients with an active purulent infection at the time of surgery, 2- Patients with a large diameter fistula (>5mm), 3- Patients with a superficial fistula or low perianal fistula, 4- Patients with a short fistulous tract (1cm in length).

One hundred and two consecutive patients were treated for high perianal fistulas by dividing them into two groups, and each group includes 51 patients. The first group (GA)

was treated by loose Seton insertion and left in the fistulous tract for 6–8 weeks, with follow-up every two weeks until healing occurred. The second group (GB) was treated by applying (APRP) into the fistulous tract after curettage of the tract and activation of APRP by calcium chloride. A second and third injection can be administered in an outpatient clinic until healing occurs.

Preoperatively for both groups: Patients are admitted to the hospital on the day of the procedure for both groups, all patients received preoperative antibiotic prophylaxis (intravenous 2g amoxicillin-clavulanate and one dose of 500mg metronidazole) early morning an hour prior to the surgery. Anal enema was then given to the patients by the nurse staff at the ward to clean the anal canal and the rectum.

Intraoperatively for both groups: Procedures were performed under general or loco-regional anesthesia (spinal or epidural) according to the patient condition and anesthesiologist's decision. All patients were positioned in the lithotomy position on the theatre table, and shaving of excess hair at perianal areas was performed before sterilization and draping.

Surgical technique for loose Seton insertion in GA: The operation was started with perianal and anal examinations intraoperatively; this careful examination was for confirmation of the diagnosis, and any missed pathology may be discovered at the time of the operation, for example, missed anal or rectal masses, ulcers, branched anal fistula tracts, active perianal abscess, inflammatory bowel diseases, which presented with anal fistulas and thick fistulous tract.

Surgeon tried to locate external and internal openings of the fistulous tract in order to excise external opening and to remove granulation tissue that may increase the recurrence rate. Finally, attempt to easily probe the tract and avoid encountering any resistance during probing, without force insertion to prevent false passage of probe and formation of a new incorrect tract. Alterna-

tively, if there was resistance and the probe didn't pass, the surgeon tried to inject (hydrogen peroxide, methylene blue, betadine) under anal speculum vision to determine internal opening tract side and o'clock. Thus, after probing in an accurate direction with a grooved Lockhart-Mummery probe to which any cheap suture (silk, prolene) was attached, the probes draw the suture through the track.

The suture was used to guide one of the following techniques (using a piece of gauze or a long strip of prolene mesh) that can pass in the tract using the already inserted suture. On the gauze second side, another suture can be connected, or long gauze can be used. At this stage, gauze or prolene mesh can be moved within the tract backwards and forwards repeatedly until no further resistance was encountered. Regardless curettage techniques, it was crucial to put a finger just within the inner orifice to help the curette-guide pass through the tract. To avoid pressure being applied to sphincters below tract, which could be partially sectioned, by maintained of assembly as longitudinal as feasible. Tract can then be repeatedly washed with hydrogen peroxide until no more bleeding, depending on available; wash tract with regular saline or sterile water. Finally, probe was reinserted and attached to Seton material that loosely tightened, hemostasis confirmed and dressing.

Surgical technique for APRP (GB): Before procedure, all patients stopped taken any non-steroidal anti-inflammatory drugs (NSAIDs), steroids, anticoagulants, and/or anti-platelets at least 10 days preoperatively, to avoid interference with platelet functions.

APRP preparation: After admission, at least two hours before surgery, blood 20–30ml were taken in citrated tube and fractionated using centrifugation. Platelets were concentrated in the platelet-rich plasma at levels generally 6–8 times the baseline levels. The resulted APRP was stored at room temperature until needed whenever 10,000 units of bovine thrombin powder are com-

bined with 10% calcium chloride (Rachita *et al*, 2014). After that, the APRP was drawn into 10ml syringe, the thrombin/calcium chloride mixture is then aspirated into a 1 ml syringe, and both syringes are mounted in a mixing applicator. At the tip of the applicator, the two preparations are mixed to activate the APRP. Within 5 to 30 seconds. A gel was formed as the citrate neutralized and the thrombin activates polymerization of the fibrin and degranulation of platelets. The gel was then inserted into the surgical field with a fine probe to allow the fibrin to be applied to the entire tract (Kucharczyk *et al*, 2014).

APRP Technique: Intraoperative perianal examinations, identification of internal and external opening, tract curettage, hemostasis and washing as in GA. Closing the internal opening (simple suturing or a patch of mucous membrane). The tract was sealed using APRP Fig. 3. That was prepared before and administered using a 14-gauge cannula color orange. After this procedure, the remnant of APRP was deep freeze (-18°C) to be use in a second application as an outpatient treatment.

Postoperatively for both groups: Patients were transferred to the ward after fully recovering and having stable vital signs. In the ward, staff nurses encouraged patient to move (stand) as soon as possible after procedure and anesthesia recovery. This helped the recovery improvement and reduced the complications' risk. Within two to four hours of the operation, the patients can get up and walk around.

Operative findings were thoroughly recorded in post-operative notes, as well as discussed with the patient and statistically analyzed. All patients were allowed to eat and drink normally, with high-fibre diet and fluid intake of at least six to ten glasses of water daily to avoid constipation. The hospital discharge was on the same day (for planned day-case surgery) or the following day. Resuming normal activities, including work, after a few days, provided the patient feels comfortable, there are no restrictions on ac-

tivity, and all patients may lift, drive, and go back to work. Patients submitted for APRP were neither allowed to receive NSAIDs, anti-platelets nor steroids in the postoperative period. The only allowed analgesics are oral paracetamol and/or pethidine.

Follow-up for both groups: At each visit (every two weeks), patients were assessed for wound healing, anal pain, discharge, and/or infection, detection of any recurrence, and fecal incontinence problem.

In GA Seton was removed after 6–8 weeks in an outpatient clinic, which was successful if the patient became asymptomatic (no pain, discharge, or recurrence with a closed external opening). Failure of surgical procedure was considered if pain, suppuration, and/or an external opening persisted after 6 months. In GB APRP, a second or third injection was needed at an outpatient clinic four to six weeks after the first one, depending on follow-up findings.

Statistical analysis: Using statistical program for social sciences, version 23.0, recorded data was examined (SPSS Inc., Chicago, Illinois, USA). Quantitative data, means, standard deviations, and ranged were reported. Qualitative variables were also shown as percentages and numbers. The independent-samples t-test of significance was used to compare two means, Chi-square (χ^2) test of significance was used to compare proportions between two qualitative components. Allowable margin of error was set at 5%, and the confidence interval was set at 95%. As a result, the p-value was deemed significant. Probability: P-values less than 0.05 were deemed significant, P-values less than 0.001 were very significant, and P-values greater than 0.05 were deemed insignificant.

Results

The evaluation included perianal and anal examination for discomfort, discharge, tenderness, erythema, incontinence, digital rectal examination, radiographic assessment, and radiographic assessment. Depended on the patient's history and health condition, an

endoanal U/S or an MRI fistulogram may be performed. Mean age of patients was 37.72 (9.82 SD) with a range of 20–61 but without significant difference $P= 0.542$. They were 16 (15.7%) females and 86 (84.3%) males.

100 (98.0%) of patients who under-went an MRI Fistulogram examination had a perianal fistula that could be reliably detected.

Healed cases was 69 (67.6%) after follow-

up, were 37 (72.5%) for loose Seton insertion and 32 (62.7%) for APRP injection, Seton group slight high healed rate without significant (0.397), 33 (32.4%) of followed-up cases unhealed, of which 14 (27.5%) involved loose Seton insertion and 19 (37.3%) involved APRP injection

Details were given in tables (1, 2 & 3) and figures (1, 2, 3 & 4).

Table 1: Comparison between Loose Seton (n=51) and APRP (n=51) according to demographic data.

| Demographic data | Total (n=102) | | Loose Seton group | | APRP group | | Test value | p-value |
|-----------------------|------------------|-------|-------------------|-------|------------------|-------|-----------------|---------|
| | No. | % | No. | % | No. | % | | |
| Age groups: <30 years | 27 | 26.5% | 12 | 23.5% | 15 | 29.4% | $\chi^2: 0.828$ | 0.661 |
| 30-40 years | 34 | 33.3% | 19 | 37.3% | 15 | 29.4% | | |
| >40 years | 41 | 40.2% | 20 | 39.2% | 21 | 41.2% | | |
| Range | 20-61 | | 20-60 | | 20-61 | | $t: 0.375$ | 0.542 |
| Mean \pm SD | 37.72 \pm 9.82 | | 38.31 \pm 10.23 | | 37.12 \pm 9.47 | | | |
| Female | 16 | 15.7% | 8 | 15.7% | 8 | 15.7% | $\chi^2: 0.000$ | 1.000 |
| Male | 86 | 84.3% | 43 | 84.3% | 43 | 84.3% | | |

Table 2: MRI Fistulogram MRI 9 (n= 102).

| Fistulogram MRI | Total | | Loose Seton insertion (n=51) | | APRP injection (n=51) | | χ^2 | p-value |
|-----------------|-------|-------|------------------------------|-------|-----------------------|-------|----------|---------|
| | No. | % | No. | % | No. | % | | |
| Positive | 100 | 98.0% | 50 | 98.0% | 50 | 98.0% | 0.000 | 1.000 |
| Negative | 2 | 2.0% | 1 | 2.0% | 1 | 2.0% | | |
| Total | 102 | 100% | 51 | 100% | 51 | 100% | | |

Table 3: Comparison between Loose Seton insertion and APRP injection as to outcome for healing fistula follow up.

| Outcome for healing fistula to complete follow up | Total | | Loose Seton insertion | | APRP injection | | χ^2 | p-value |
|---|-------|-------|-----------------------|-------|----------------|-------|----------|---------|
| | No. | % | No. | % | No. | % | | |
| Unhealed fistula | 33 | 32.4% | 14 | 27.5% | 19 | 37.3% | 0. | 0.397 |
| Healing fistula | 69 | 67.6% | 37 | 72.5% | 32 | 62.7% | 71 | |
| Total | 102 | 100% | 51 | 100% | 51 | 100% | 7 | |

Discussion

Generally, Fistula-in-ano is one of the commonest anorectal diseases, greater in men than women, with a rate of 12.3 cases/100,000 and 5.6 cases/100,000, respectively (Sainio, 1984). Risk factors for fistula development include obesity, diabetes, smoking, hyperlipidemia, and a sedentary lifestyle (Wang *et al*, 2014). High perianal fistulas require complex treatment, normally with partial division of the sphincter (Wei *et al*, 2019). This technique is associated with an increased risk of incontinence and achieves rates of success that are variable, yet significantly lower than with low fistulas. Conventional laying open technique in high perianal fistulas may involve sacrifice of part or the whole sphincter muscle, impairing continence. The greater the extent of anal muscle division, the greater the degree of anal incontinence (Ho *et al*, 1998).

High-complexity fistulas can be safely treated with only minor loss of continence using the Seton technique (Cintron *et al*, 2000). Another technique for preserving the sphincter was local application of PRP, which may be an alternative to the laying open methods (Moreno-Serrano *et al*, 2016). The ability of enriched plasma to provide high concentrations of platelet-derived growth factors to soft tissues near fistulous tracts, with subsequent acceleration of fistula healing was mentioned in a few experimental reports, including applications in complex (Alves and Grimalt, 1995) and Crohn's anal fistulas (Göttgens *et al*, 2015)

Loose-Seton devices used in this review were placed after curettage of the fistulous tract and granulation tissue at the external opening was excised.

In the present study, male preponderance was (84.3%) of all patients, with the ages

ranged from 30 up to 60 years. This was greater than other age groups for high perianal fistula (McCourtney *et al.*, 1999; Seow-Choen *et al.*, 2003). The difference can be due to environmental cultures and/ or style-life.

In the present study, both groups show the main patients compliant preoperatively is perianal discharge that almost presented in all patients (100%), and second main complaint is perianal pain (60.8%).

In the present study, the MRI proved superior to Endoanal U/S in the fact enabling tissues differentiation, including identification of active inflammatory lesions and their differentiation from scars (recurrent fistulas and posttraumatic lesions) as (98.0%) of examined patient with MRI fistulogram showed accurate diagnosis for perianal fistula. This agreed with Ziech *et al.* (2009) in The Netherlands, who reported that perianal fistulas, cryptoglandular or Crohn's disease-related, have a tendency to recur, and that preoperative imaging helped to identify extensions that otherwise would be missed during surgery and therefore prevent recurrence.

In the present study, healing fistulas in GA was 37(72.5%) after complete follow up period. This agreed with McCourtney *et al.* (1999), UK they reported that Seton techniques have been used in the successful management of fistula in ano for thousands of years. They added that of 33 loose Seton patients the fistula tract eradication was 60-78% of cases.

The present recurrence rate in GA was (27.5%) and disturbance in continence was minor and not lasting for long time. Abbas *et al.* (2008) in USA reported that all recurrences were in patients with left sided fistulae. At last follow-up, all had healed their fistula except for two. They concluded that ERF closed most complex anorectal fistulae with an acceptable complication rate and low recurrence rate.

In the present study, GB reacted complete healing of anal fistulas after APRP applicati-

on was 32 (62.7%), after APRP application was (62%). Moreno-Serrano *et al.* (2015) in Spain reported that there was a good benefit to use of Vivostat PRF[®] as a treatment for complex perianal fistulas. They concluded that a highly reproducible technique with acceptable results and does not produce impairment of continence. Hermann *et al.* (2022) in Poland reported that PRP application of in small, low, and recurrent IBD anal fistulas is effective, simple, and safe with an acceptable rate of healing. This therapy may also precede any further, surgical methods of treatment.

In the present study, as far as APRP number of injections was considered, fistulas were clinically closed after 2 to 3 injections. Healing process took 4 up to 6 weeks, but the average time of recurrence occurred at 8 up to 10 weeks after the final plasma application. Patients (33.3%) needed second APRP injection for healing, and (23.5%) needed third APRP injection for healing. Van der Hagen *et al.* (2011) in The Netherlands who studied explore autologous platelet-rich plasma as an adjunct to the staged mucosal advancement flap in the treatment of perianal fistulae, reported that platelet-rich plasma as an adjunct to a staged mucosal advancement flap for the treatment of perianal cryptoglandular fistulae was a promising treatment modality and seems to establish a high healing rate.

In the present study, loose Seton insertion and APRP gave better results than traditional methods without significant difference in healing ($P = 0.397$). So, treatment is indicated for recurrent as well as primary fistulas, as less invasive, painless, low morbidity, and easier to perform. Feroz *et al.* (2020) in USA found major disadvantage of Seton therapy was the discomfort and to achieve time stability. They hypothesized that the two aspects should be addressed concurrently to increase the fistula healing or closure rate. First, Seton should be used as initial therapy to maintain tract patency to allow abscess drainage and minimize the intestinal

flora colonization within the tract mucosa, thereby leukocytic infiltration and inflammatory propagation within tract. Second one target initial stimulation of Th1/Th17 mediated hypersensitivity instead of a factor/ cytokine in inflammation mediation.

Conclusion

The outcome showed that none of the two techniques had significantly difference. But, there was a marginally higher rate of healing with loose Seton than with APRP injection.

Recommendation

Both Loose Seton and APRP as the first line of defense against high anal fistulas is indicated as patient-preferred, low-risk, easy to repeat, and low-cost.

In addition, antibiotics (metronidazole or ciprofloxacin) are also supplemented, which further promotes healing

References

Abbas, MA, Lemus-Range, R, Hamadani, A, 2008: Long term outcome of Seton for complex anorectal fistulae. *Am. Surg.* 74, 10: 921-4.

Adamo, K, Sandblom, G, Brannstrom, F, Strigard, K, 2016: Prevalence & recurrence rate of perianal abscess, a population-based study, Sweden 1997-2009. *Int. J. Col. Dis.* 31, 3:669-73.

Alves, R, Grimalt, R, 2018 A review of platelet-rich plasma: History, biology, mechanism of action, and classification. *Skin Append. Disord.* 4:18-24.

Balci, S, Onur, MR, Karaosmanoğlu, AD, Karçaaltıncaba, M, Akata, D, et al, 2019: MRI evaluation of anal and perianal diseases. *Diagn. Interv. Radiol.* 25, 1:21-7.

Cintron, JR, Park, JJ, Orsay, CP, et al, 2000: Repair of fistulas-in-ano using fibrin adhesive: Long term follow up. *Dis. Colon Rectum* 43: 944-50.

Corman, M, Abcarian, H, Bailey, HR, et al, 2008: Consensus statement on the use of the bio-prosthetic plug for anal fistulas. *Colorect. Dis.* 10:17-22.

De Groof, EJ, Cabral, VN, Buskens, CJ, Morton, DG, Hahnloser, D, et al, 2016: Systematic review of evidence and consensus on perianal fistula: An analysis of national and international guidelines. *Colorectal Dis.* 8: O119-34.

Fathallah, N, Alam, A, Haouari, M, et al, 2022: Injection of mesenchymal stem cells (darvadstrocel) into perianal fistula of Crohn's disease:

A video vignette. *Colorectal Dis.* Sep. 2022, 24(9):1097. doi: 10.1111/codi.

Faucheron, JL, Saint-Marc, O, Guibert, L, Parc, R, 1996: Long-term seton drainage for high anal fistulas in Crohn's disease: A sphincter saving operation. *Dis. Colon Rectum* 39:208-11.

Feroz, SH, Ahmed, A, Muralidaran, A, Thirunavukarasu, P, 2020: Comparison of the efficacy of the various treatment modalities in the management of perianal Crohn's fistula: A review. *Cureus* 12, 12:e11882. doi:10.7759/cureus.11882.

Göttgens, KW, Smeets, RR, Stassen, LP, Beefts, GL, Pierik, M, et al, 2015: Treatment of Crohn's disease-related high perianal fistulas combining the mucosa advancement flap with platelet-rich plasma: A pilot study. *Tech. Coloproctol.* 19:455-9.

Hermann, J, Cwaliński, J, Banasiewicz, T, 2022: Application of platelet-rich plasma in rectovaginal fistulas in the patients with ulcerative colitis. *Langenbecks Arch. Surg.* 407, 1: 429-33

Ho, YH, Tan, M, Leong, AF, Seow-Choen, F, 1998: Marsupialization of fistulotomy wounds improves healing: A randomized controlled trial. *Br. J. Surg.* 85, 1:105-7.

Khera, PS, Badawi, HA, Afifi, AH, 2010: MRI in perianal fistulae. *Indian J. Radiol. Imag.* 20, 1: 53-7.

Kucharczyk, A, Kołodziejczak, M, Sudół-Szopińska, I, Bielecki, K, 2014: Autologous growth factors used for the treatment of recurrent fistula-in-ano preliminary results. *Dis Colon Rect. Tech. Coloproctol.* 18, 3:317-8.

McCourtney, JS, Finlay, IG, 1995: Setons in the surgical management of fistula in ano. *Br. J. Surg.* 82, 4:448-52.

Moreno-Serrano, A, García-Díaz, JJ, Ferrer-Márquez, M, Alarcón-Rodríguez, R, Álvarez-García, A, et al, 2015: Using autologous platelet-rich plasma for the treatment of complex fistulas. *Rev. Esp. Enferm. Dig.* 108, 3:123-8.

Pellino, G, Selvaggi, F, 2014: Surgical treatment of perianal fistulizing Crohn's disease: From lay-open to cell-based therapy: An overview. *Sci. World J.* 14:6281-6.

Rachita, D, Sukesh, MS, 2014: Principles and methods of preparation of platelet-rich plasma: A review and author's perspective. *J. Cutan. Aesthet. Surg.* 7, 4:189-97.

Sainio, P, 1984: Fistula-in-ano in a defined population: Incidence and epidemiological aspects. *Ann. Chir. Gynaecol.* 73, 4:219-24.

Seow-Choen, F, 2003: Seton insertion for difficult anal fistulas. *Colorectal Dis.* 5:373-7.

Sharma, G, Khandige, G, Mohan, M, 2016: Magnetic resonance imaging in perianal fistulas: A pictorial atlas. *Indian J. Gastroenterol.* 35, 5: 337-42.

Singh, K, Singh, N, Thukral, CL, Singh, KP, Bhalla, V, 2014: Magnetic resonance imaging (MRI) evaluation of perianal fistulae with surgical correlation. *J. Clin. Diagn. Res.* 8, 6: RC0 1-4. Published online 2014 doi:10.7860/JCDR/2014/7328.4417

Van der Hagen, SJ, Baeten, CG, Soeters, P B, van Gemert, WG, 2011: Autologous platelet-derived growth factors (platelet-rich plasma) as an adjunct to mucosal advancement flap in high cryptoglandular perianal fistulae: A pilot study. *Colorectal Dis.* 13:215-8

Vasilevsky, CA, Gordon, PH, 1984: The incidence of recurrent abscesses or fistula-in-ano following ano-rectal suppuration. *Dis. Colon Rect.* 27, 2:126-30.

Wang, D, Yang, G, Qiu, J, Song, Y, Wang, L, et al, 2014: Risk factors for anal fistula: A case control study. *Tech. Coloproctol.* 18, 7: 635-9.

Wei, P-L, Keller, JJ, Kuo, LJ, Lin, HC, 2013: Increased risk of diabetes following perianal abscess: A population-based follow-up study. *Int. J. Colorectal Dis.* 28:235-40.

Williams, JG, MacLeod, A, Rothenberger, A, Goldberg, M, 2001: Seton treatment of high anal fistulae. *Br. J. Surg.* 78:1159-61.

Ziech, M, Felt-Bersma, R, Stoker, J, 2009: Imaging of perianal fistulas. *Clin. Gastroenterol. Hepatol.* 7:1037-45.

Explanation of figures

Fig 1: A probe inserted into a tract from 6 o'clock position before performing Seton insertion.

Fig. 2: Curettage of fistula tract-using piece of gauze between two stings

Fig. 3: Injection of fistulous tract with activated APRP.

Fig. 4: Fistula has healed temporarily but recurs (2 months after Seton removal).

