The Journal of Laryngology & Otology

cambridge.org/jlo

Studying the result of underlay myringoplasty using platelet-rich plasma

S P S Yadav[1], J S Malik[1], P Malik[1], P K Sehgal[2], J S Gulia[1] and R K Ranga[1]

Departments of [1]Otolaryngology and [2]Blood Transfusion, Post-Graduate Institute of Medical Sciences ('PGIMS'), Rohtak, India

Abstract

Objective. Perforations of the tympanic membrane are treated with various surgical techniques and materials. This study aimed to determine the efficacy of platelet-rich plasma during underlay myringoplasty.

Methods. The study included 40 patients. Autologous platelet-rich plasma was applied in-between temporalis fascia graft and tympanic membrane remnant during underlay myringoplasty in group 1 ($n = 20$). The outcome was evaluated after three months and compared with group 2 ($n = 20$), a control group that underwent routine underlay tympanoplasty.

Results. After three months' follow up, graft uptake was 95 per cent in group 1 and 85 per cent in group 2 ($p < 0.03$). Mean hearing threshold gain was 18.62 dB in group 1 and 13.15 dB in group 2. This difference was statistically significant ($p < 0.01$).

Conclusion. Platelet-rich plasma, with its ease of preparation technique, availability, low cost, autologous nature and good graft uptake rate, justifies its use in tympanoplasty type I procedures.

Main Article

Dr P Malik takes responsibility for the integrity of the content of the paper

Cite this article: Yadav SPS, Malik JS, Malik P, Sehgal PK, Gulia JS, Ranga RK. Studying the result of underlay myringoplasty using platelet-rich plasma. *J Laryngol Otol* 2018;**132**: 990–994. https://doi.org/10.1017/S0022215118001846

Accepted: 28 May 2018
First published online: 29 October 2018

Key words:
Platelet-Rich Plasma; Tympanoplasty; Otitis Media, Suppurative; Myringoplasty; Platelet-Derived Growth Factor

Author for correspondence:
Dr Priya Malik,
Room no. 211, Girls Hostel, PGIMS,
Rohtak 124001, Haryana, India
E-mail: drpriyamalik09@gmail.com

Introduction

Chronic suppurative otitis media (CSOM) represents one of the most common infections of the ear. It is a worldwide health problem that is still prevalent in the modern antibiotic era.[1] Chronic suppurative otitis media is characterised by chronic purulent discharge through a perforated tympanic membrane, which is intermittent or persistent, and which can be associated with cholesteatoma.[2]

Tympanoplasty type I, or myringoplasty, is reconstructive surgery limited to tympanic membrane perforation repair. The key objectives of myringoplasty are to restore tympanic membrane integrity and hearing improvement.[3] Fundamental to the successful closure of any perforation is an adequate area of contact between the graft and tympanic membrane remnant. Other factors influencing successful tympanoplasty include perforation location, ear canal geometry and bleeding during surgery, which can affect optimal graft placement. Tympanic membrane graft migration due to natural events taking place in the reconstructed tympanic membrane can explain some in situ failures, and this remains a significant problem.

It can be inferred that any substances which promote rapid and effectual growth could prevent graft migration because of their direct cohesion with the tympanic membrane. There is a constant search for biomaterials that meet certain requirements, including being safe, biocompatible, low in cost and as structurally similar to human tissue as possible.[4,5]

Platelet-rich plasma is defined by a portion of the plasma fraction of autologous blood having a platelet concentration above baseline, with the full complement of clotting and growth factors like platelet-derived growth factor, vascular endothelial growth factor and transforming growth factor.[6,7] Platelet-rich plasma serves as a growth factor agonist, and has both chemotactic and mitogenic properties.[7–9] It functions as a tissue sealant and an adhesive, surgical haemostatic agent. Platelet-rich plasma is safe, being biocompatible, and is effective. It accelerates the regeneration of endothelial, epithelial and epidermal layers. It enhances collagen synthesis, angiogenesis and soft tissue healing, and decreases dermal scarring by reversing glucocorticoids that mediate the inhibition of wound healing.[10] Platelet-rich plasma can be prepared with ease, with minimal effort, and is available at the point of care.[11]

Cases of CSOM with large, total or subtotal perforations of the tympanic membrane are common in our country. Given the encouraging results of platelet-rich plasma in underlay myringoplasty reported in a small number of previous studies, the present study was undertaken to evaluate the efficacy of platelet-rich plasma in underlay tympanoplasty using temporalis fascia.

Materials and methods

Study population and pre-operative evaluation

The present randomised, prospective study was conducted in the Department of Otorhinolaryngology, Pandit Bhagwat Dayal Sharma Post-Graduate Institute of Medical

Sciences, Rohtak, India, from January 2014 to February 2015. It included 40 patients of both sexes, aged 16–50 years, who had unilateral or bilateral chronic inactive (mucosal) otitis media with a dry ear over a period of at least 4 weeks. Patients had not used topical or systemic antibiotics, had a good cochlear reserve, and an air–bone gap of more than 25 dB on pure tone audiometry. Patients for whom there was the slightest doubt of cholesteatoma, revision cases and those with sensorineural hearing loss were excluded from study.

All patients underwent a detailed evaluation that involved history-taking, a general physical examination and a complete ENT examination. Tuning fork tests and a pure tone audiogram were conducted to record pre-operative type and degree of hearing loss.

Institutional review board approval was acquired. Informed consent was obtained and available alternative treatment was discussed.

All procedures were conducted under local anaesthesia. Patients were divided randomly into two groups: group 1 patients ($n = 20$) underwent underlay myringoplasty using a temporalis fascia graft with platelet-rich plasma; group 2 patients ($n = 20$) underwent underlay myringoplasty using a temporalis fascia graft without platelet-rich plasma.

Platelet-rich plasma acquisition

First, 10 cc of a patient's venous blood was taken into a tube that contained an anti-coagulant to avoid platelet activation and degranulation. Then, centrifugation, involving a 'soft spin', separated the blood into three layers. The bottom-most layer comprised red blood cells (55 per cent of total volume), the top-most layer consisted of acellular platelet-poor plasma (40 per cent of total volume) and the intermediate layer contained platelet-rich plasma (5 per cent of total volume) called the 'buffy coat'.

Using a syringe, platelet-poor plasma, platelet-rich plasma and some red blood cells were transferred into another tube without an anti-coagulant. A second 'hard spin' centrifugation was conducted, which was longer and faster. Platelet-rich plasma was allowed to settle at the bottom. The acellular plasma (80 per cent of the volume) found at the top, was removed with a syringe. The platelet-rich plasma was mixed with bovine thrombin and calcium chloride at the time of application, which results in the gelling of this platelet concentrate.

Myringoplasty

During underlay myringoplasty, the graft was advanced under the tympanomeatal flap and the malleus handle to the anterior-most extent of the perforation. The edges of the graft were tucked under the de-epithelised margin of the drum remnant. The platelet-rich plasma was applied, with a tuberculin syringe fitted with a needle, onto the graft, visible through the perforation. Some of the platelet-rich plasma seeped in-between the graft and tympanic membrane remnant. Small pledgets of Gelfoam soaked in platelet-rich plasma were used to overlap the junction of the rim and graft circumferentially. Gelfoam pledgets soaked in antibiotic solution were placed lateral to the tympanic membrane. An umbilical tape coated with framycetin ointment was placed in the external auditory canal.

Table 1. Age and sex distribution of CSOM patients

	Group 1		Group 2	
Age group	Males	Females	Males	Females
15–25 years	9	8	11	3
26–35 years	1	0	2	1
36–50 years	1	1	0	3
Total (*n* (%))	11 (55)	9 (45)	13 (65)	7 (35)

Data represent numbers of patients. CSOM = chronic suppurative otitis media

Table 2. Ear involvement

Ear involved	Group 1	Group 2
Unilateral		
– Right	6 (30)	7 (35)
– Left	4 (20)	8 (40)
Bilateral	10 (50)	5 (25)

Data represent numbers (and percentages) of cases

Table 3. Duration of ear discharge

Duration (years)	Group 1	Group 2
0–3	12 (60)	6 (30)
4–6	6 (30)	7 (35)
7–8	2 (10)	7 (35)

Data represent numbers (and percentages) of cases

Table 4. Audiological assessment

		Patients (*n* (%))	
Degree of hearing loss	Hearing loss (dB)	Group 1	Group 2
Mild	25–40	9 (45)	17 (85)
Moderate	41–55	11 (55)	3 (15)
Moderate to severe	56–70	0 (0)	0 (0)
Severe	71–90	0 (0)	0 (0)
Profound	>90	0 (0)	0 (0)

Statistical analysis

The collected data were statistically analysed using the student's *t*-test. For qualitative data, the chi-square test was used. Normally distributed data are presented as means and standard deviations. All tests were performed at a 5 per cent level of significance; thus an association was significant if the *p*-value was less than 0.05.

Results

In group 1, 11 of 20 patients were male (55 per cent) and 9 were female (45 per cent), with age ranging from 16 to 48 years. In group 2, 13 of the 20 patients were male (65 per cent) and 7 were female (35 per cent), with age ranging from 15 to 45 years (Table 1). In both groups, most of the cases showed unilateral ear involvement (Table 2).

https://doi.org/10.1017/S0022215118001846 Published online by Cambridge University Press

Table 5. Relationship between perforation size and graft uptake

	Group 1		Group 2	
Perforation size	Total cases	Graft uptake	Total cases	Graft uptake
Grade I	–	–	–	–
Grade II	–	–	–	–
Grade III	5 (25)	5/5 (100)	12 (60)	11/12 (91.6)
Grade IV	9 (45)	9/9 (100)	5 (25)	4/5 (80)
Grade V	6 (30)	5/6 (83.33)	3 (15)	2/3 (66.6)
Overall	20	19/20 (95)	20	17/20 (85)

Data represent numbers (and percentages) of cases

All the patients had a history of ear discharge. In group 1, 12 patients (60 per cent) complained of ear discharge for 0–3 years, 6 (30 per cent) for 4–6 years and 2 (10 per cent) for 7–8 years. In group 2, 6 patients (30 per cent) complained of ear discharge for 0–3 years, 7 (35 per cent) for 4–6 years and 7 (35 per cent) for 7–8 years (Table 3).

Patients also complained of varying degrees of deafness. In group 1, 20 patients (100 per cent) were affected; in group 2, 11 patients (55 per cent) complained of deafness. The chi-square test value was 4.85 with 2 degrees of freedom. The *p*-value was 0.08, which was statistically insignificant. In both groups, most of the cases showed unilateral ear involvement. In group 1, 45 per cent of patients had hearing loss in the range of 25–40 dB, with 55 per cent in the range of 41–55 dB. In group 2, 85 per cent of patients had hearing loss in the range of 25–40 dB, with 15 per cent in the range of 41–55 dB (Table 4).

Regarding tympanic membrane perforation, in group 1, five patients (25 per cent) had a medium perforation (up to half the size of the tympanic membrane; grade III), whereas nine patients (45 per cent) had a large central perforation (grade IV) and six patients (30 per cent) had a grade V perforation. In group 2, 3 patients (15 per cent) had a subtotal perforation, whereas 5 patients (25 per cent) had a large central perforation and 12 patients (60 per cent) had a grade III perforation (Table 5). The chi-square test value was 5.02, with 2 degrees of freedom. The *p*-value was 0.081, which was statistically insignificant.

All patients presented themselves for regular follow up. In group 1, discharge was observed in two patients, which resolved following a change of antibiotic, and only one patient had graft rejection. In group 2, discharge was observed in three patients, which resolved with a change of antibiotic. The overall graft take-up rate in group 1 was 95 per cent; in group 2 the graft take-up rate was 85 per cent. The chi-square test value was 4.32, with 1 degree of freedom. The *p*-value was 0.03, which was statistically significant. In group 1, the graft take-up rate was 100 per cent both for grade III and grade IV perforations, and was 83.33 per cent for grade V perforations. In group 2, the graft take-up rate was 91.6 per cent for grade III perforations, 80 per cent for grade IV perforations and 66.6 per cent for grade V perforations (Table 5).

Hearing levels were assessed at three months. In group 1, the mean pre-operative hearing threshold was 42.13 dB, while the mean post-operative hearing threshold was 23.51 dB, resulting in a mean gain of 18.62 dB. The mean pre-operative air–bone gap was 30.78 dB, while the mean post-operative air–bone gap was 12.16 dB, giving a mean gain of 18.62 dB. Statistical comparison of pre- versus post-operative air–bone gaps revealed a highly significant difference ($p < 0.001$).

Table 6. Overall follow-up results

Parameter	Group 1	Group 2
Cases operated on	20 (100)	20 (100)
Cases followed up	20 (100)	20 (100)
Residual perforation	–	–
Graft rejection	1 (5)	3 (15)
Anterior sulcus blunting	–	–
Graft lateralisation	–	–
Hearing improvement		
– 0–10 dB	2 (10)	6 (30)
– 11–20 dB	12 (60)	13 (65)
– 21–30 dB	5 (25)	1 (5)
– 31–40 dB	1 (5)	0 (0)

Data represent numbers (and percentages) of cases

In group 2, the mean pre-operative hearing threshold was 37.64 dB, while the mean post-operative hearing threshold was 24.49 dB, resulting in a mean gain of 13.15 dB. The mean pre-operative air–bone gap was 26.94 dB, while the mean post-operative gap was 13.79 dB, giving a mean gain of 13.15 dB. The chi-square test value was 8.27, with 3 degrees of freedom. The *p*-value was 0.04 for the gain in hearing threshold (air–bone gap gain) between both groups, which was statistically significant (Table 6).

Discussion

At present, autologous temporalis fascia is the material of choice as a graft for tympanoplasty. This is because of its many qualities, such as being rich in collagen fibres and lacking the elastic elements that make the graft easy to handle. In addition, its ease of availability, even in revision surgery, makes it a popular choice for myringoplasty. Since Heerman described its use in 1961, various authors have utilised the temporalis fascia in tympanoplasty.[12] Temporalis fascia has been successfully used both in underlay and overlay tympanoplasty methods.[13]

However, temporalis fascia grafts are not immune to problems. The graft has a tendency to 'pull off' the malleus handle, and its use can lead to blunting of the anterior sulcus. This is associated with thickening of the tympanic membrane in this

https://doi.org/10.1017/S0022215118001846 Published online by Cambridge University Press

area, resulting in partial fixation of the malleus to the anterior canal wall. These problems can, to a certain extent, be solved by placing the graft under the handle of the malleus.[14]

Platelet releasates have been used to treat wounds since 1985. The application of platelet-rich plasma has been documented in many fields, such as dentistry, orthopaedics, and cosmetic and plastic surgery. In otology, the efficacy of platelet-rich plasma has been demonstrated in tympanoplasty procedures.[15]

Soumekh *et al.* assessed the efficacy of a platelet releasate in the treatment of chronic, non-healing perforations in 34 chinchillas with bilateral perforations, with 1 treated ear and 1 control ear. After three months, a comparison of both sides suggested that the platelet releasate was not effective.[16] Yeo *et al.* investigated the effect of platelet-derived growth factor-AA on the healing process of tympanic membrane perforation in rats.[17] The results revealed a speeding up of the healing process of the tympanic membrane defect, an improved rate of healing, and fewer atrophic changes in the healed tympanic membrane. This is explained by the fact that platelet-derived growth factor-AA promotes connective tissue growth.[17] Erkilet *et al.* created a traumatic tympanic membrane perforation in 44 rats, and found that platelet-rich plasma was effective in accelerating tympanic membrane perforation healing.[18] Farrag *et al.* studied the effect of platelet-rich plasma and fibrin sealant on facial nerve regeneration in 49 male adult rats, and found platelet-rich plasma be to effective when added to sutures.[19]

- Conventional underlay myringoplasty using temporalis fascia with platelet-rich plasma was compared with temporalis fascia graft without platelet-rich plasma
- Platelet-rich plasma is an autologous material and serves as a growth factor agonist
- It has mitogenic, chemotactic, tissue adhesive, haemostatic and tissue ceiling properties
- Platelet-rich plasma thus improves the overall success rate of graft uptake and can substantially decrease graft failure
- Platelet-rich plasma is an immediate surgical haemostatic agent that is biocompatible, safe, effective and improves graft uptake in myringoplasty

Mehmet *et al.* studied the effect of platelet-rich plasma on the healing of traumatic tympanic membrane perforations.[20] After 1 month, 9 out of 14 cases treated with platelet-rich plasma had total closure; in the control group, 4 out of 18 cases had total closure. However, after two months there was no significant difference in the ratio of closure between the study group and control group ($p > 0.05$).[20] Navarrate Alvaro *et al.* conducted a pilot study on the efficacy of autologous platelet-rich plasma growth factors in tympanoplasty type I procedures; the perforation closed in all three subjects.[21] Gopalakrishnan *et al.* studied the efficacy of autologous platelet-rich plasma during myringoplasty in terms of tympanic membrane perforation closure.[22] Out of 25 cases, 24 had complete tympanic membrane closure and only 1 failure was observed. In contrast, the tympanic membrane perforation failed to close in 5 out of 25 control subjects. The findings suggested that the use of platelet-rich plasma accelerates graft uptake, and thereby the tympanic membrane perforation closure, following myringoplasty.[22] Similar results were observed in the present study.

Platelet-rich plasma accelerates the healing of tympanic membrane perforation following myringoplasty. It prevents graft displacement or shrinkage, especially in wet grafts, with its sealant property. Platelet-rich plasma improves the overall success rate of myringoplasty. Furthermore, it has no noticeable side effects.[6] Given the efficacy of platelet-rich plasma in myringoplasty, as shown here and in larger samples, its use for tympanic membrane perforation repair would result in lower intra-hospital and out-patient costs.[23]

Platelet-rich plasma is a platelet concentrate with enriched growth factors that can be obtained in a cost-effective manner and which is easy to prepare. In a two-step process, whole blood from the patient is first centrifuged to separate the plasma from packed red blood cells, and then centrifuged further to separate platelet-rich plasma from platelet-poor plasma. This concentrate is then activated with the addition of thrombin or calcium chloride, resulting in a gelatinous platelet gel. Potential candidates for treatment with platelet-rich plasma should undergo a pre-treatment haematological evaluation to rule out coagulopathies and platelet function disorders. Anaemic patients and those with thrombocytopenia may be unsuitable candidates for treatment with platelet-rich plasma. Other potential contraindications include haemodynamic instability, severe hypovolaemia, unstable angina, sepsis, and anti-coagulant or fibrinolytic drug therapy.[8,9,23]

Conclusion

Underlay myringoplasty using platelet-rich plasma is a simple, safe, quick and economical procedure conducted under local anaesthesia. It has a good success rate as compared to fascia alone, with minimal or no morbidity, and no complications. Platelet-rich plasma acts as an immediate surgical haemostatic agent that is biocompatible, safe and effective. It improves overall graft uptake rate in myringoplasty by preventing graft shrinkage. Platelet-rich plasma also functions as a tissue sealant, and plays an important role as a host defence mechanism at the wound site, thus improving the survival of the graft, with no adverse effects.

Competing interests. None declared

References

1 Lee YP, Auo HJ, Kang JM. Loop overlay tympanoplasty for anterior or subtotal perforations. *Auris Nasus Larynx* 2010;**37**:162–6
2 Gopen Q. Pathology and clinical course of inflammatory disease of the middle ear. In: Gulya AJ, Minor LB, Poe DS, eds. *Glasscock–Shambaugh Surgery of the Ear*, 6th edn. Shelton, CT: Peoples Medical Publishing House-USA. 2010;425–36
3 Maroto PD, Gutiérrez JJ, Jiménez MC, Morente JC, Rodríguez VP, Benítez PN. Functional results in myringoplasties [in Spanish]. *Acta Otorrinolaringol Esp* 2010;**61**:94–9
4 Jung TT, Park SK. Mediolateral graft tympanoplasty for anterior or subtotal tympanic membrane perforation. *Otolaryngol Head Neck Surg* 2005;**132**:532–6
5 Yoo J. Clinical application of tissue adhesives in soft-tissue surgery of the head and neck. *Curr Opin Otolaryngol Head Neck Surg* 2008;**16**:312–17
6 Carter MJ, Fylling CP, Parnell LK. Use of platelet rich plasma gel on wound healing: a systematic review and meta-analysis. *Eplasty* 2011;**11**:e38
7 Mehta S, Watson JT. Platelet rich concentrate: basic science and current clinical applications. *J Orthop Trauma* 2008;**22**:432–8
8 Marx RE. Platelet-rich plasma (PRP): what is PRP and what is not PRP? *Implant Dent* 2001;**10**:225–8

https://doi.org/10.1017/S0022215118001846 Published online by Cambridge University Press

9 Gasling VL, Acil Y, Springer IN, Hubert N, Wiltfag J. Platelet-rich plasma and platelet-rich fibrin in human cell culture. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2009;**108**:48–55
10 Nikolidakis D, Jansen JA. The biology of platelet-rich plasma and its application in oral surgery: literature review. *Tissue Eng Part B Rev* 2008;**14**:249–58
11 Everts PA, Brown Mahoney C, Hoffmann JJ, Schönberger JP, Box HA, van Zundert A *et al.* Platelet-rich plasma preparation using three devices: implications for platelet activation and platelet growth factor release. *Growth Factors* 2006;**24**:165–71
12 Gulia JS, Yadav SP, Khaowas A. Medio-lateral myringoplasty versus medial myringoplasty: a comparative study. *Otolaryngol Head Neck Surg* 2013;**149** (2 suppl):P231
13 Fiorino F, Barbieri F. 'Over-under' myringoplasty with umbus-anchored graft. *J Laryngol Otol* 2008;**122**:854–7
14 Sakagami M, Yuasa R, Yuasa Y. Simple underlay myringoplasty. *J Laryngol Otol* 2007;**121**:840–4
15 Driver VR, Hanft J, Fylling CP, Beriou JM; Autologel Diabetic Foot Ulcer Study Group. A prospective, randomized, controlled trial of autologous platelet-rich plasma gel for the treatment of diabetic foot ulcers. *Ostomy Wound Manage* 2006;**52**:68–70,72,74
16 Soumekh B, Hom DB, Levine S, Juhn SK, Antonelli PJ. Treatment of chronic tympanic-membrane perforations with a platelet-derived releasate. *Am J Otol* 1996;**17**:506–11
17 Yeo SW, Kim SW, Suh BD, Cho SH. Effects of platelet-derived growth factor-AA on the healing process of tympanic membrane perforation. *Am J Otolaryngol* 2000;**21**:153–60
18 Erkilet E, Koyuncu M, Atmaca S, Yarim M. Platelet-rich plasma improves healing of tympanic membrane perforations: experimental study. *J Laryngol Otol* 2009;**123**:482–7
19 Farrag TY, Lehar M, Verhaegen P, Carson KA, Byrne PJ. Effect of platelet rich plasma and fibrin sealant on facial nerve regeneration in a rat model. *Laryngoscope* 2007;**117**:157–65
20 Platelet Rich Fibrin for the Repair of Tympanic Membrane. In: https://www.researchposters.com/Posters/AAOHNSF/aao2012/SP398.pdf [27 September 2018]
21 Navarrete Alvaro ML, Ortiz N, Rodriguez L, Boemo R, Fuentes JF, Mateo A *et al.* Pilot study on the efficiency of the biostimulation with autologous plasma rich in platelet growth factors in otorhinolaryngology: otologic surgery (tympanoplasty type I). *ISRN Surg* 2011;**2011**: 451020
22 Gopalakrishnan S, Venkatasamy P, Vivekanandamurthy K. A study on efficacy of autologous platelet rich plasma in myringoplasty. *Online J Otolaryngol* 2013;**3**:36–51
23 Gonshor A. Technique for producing platelet-rich plasma and platelet concentrate: background and process. *Int J Periodontics Restorative Dent* 2002;**22**:547–57

https://doi.org/10.1017/S0022215118001846 Published online by Cambridge University Press