

# Comparative study between Platelet Rich Fibrin and Temporalis Fascial Graft in Type 1 Tympanoplasty

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## Comparative study between Platelet Rich Fibrin and Temporalis Fascial Graft in Type 1 Tympanoplasty

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### ABSTRACT

**Background.** Myringoplasty is the surgical procedure that aim to restore a barrier between epithelial lining external auditory canal and mucosal lining middle ear cavity. Grafting materials for this procedure are variable and includes: fascia, cartilage, fat and paper patches, all of which act as a scaffold for cells

migration. Platelet rich fibrin (PRF) is strictly autologous material and its role in accelerating tissue healing had been established in various branches for its high content of growth factors. The aim of study is to evaluate role of PRF in the healing of chronic central tympanic membranes perforation and to comparing the results with that of temporalis fascial graft in type I tympanoplasty.

**METHODS.** A prospective comparative study enrolled fifty six cases attended ENT department. Cases were distributed into two groups, group A underwent myringoplasty with platelet rich fibrin alone under local anesthesia, and group B underwent myringoplasty with temporalis facial graft under general anesthesia. PRF was prepared from patient own venous blood in which 9 ml collected in plain glass tube and centrifuged immediately at 2700 rpm/12 min. Cases were followed up for 6 months postoperatively.

**RESULTS.** The main age in group-A was (30-39yr) as 35.7%, while in group B was (20-29yr) as 32.1%. Cases were divided upon size of perforation into three groups:  $\leq 25\%$ ,  $>25\%-50\%$ , and  $>50\%$ -subtotal. The overall success was in group-A 71.4% (20 of 28 patients) and 82.1% in group-B (23 of 28 patients) ( $P=0.342$ ). The healing for perforation  $\leq 25\%$  was 83.3% in group-A and 100% in group-B, for perforation of 25%-50% the healing in group-A was 63.6% and in group-B was 76.9%, while in perforation larger than 50% , the healing in group-A was 60% and in group-B was 71.4%.

**CONCLUSION.** PRF is a grafting material acting as a scaffold that stimulates the healing process of perforated tympanic membrane. It lacks the harvesting site morbidity and is relatively more abundant material than fascial graft. The success rate decreases with increase in perforation size and repeated sessions are more likely to be required.

**Keywords:** platelet rich fibrin, tympanic membrane perforation, tympanoplasty, temporalis fascial graft

## INTRODUCTION

Ears function are an early warning system via detection and localization potentially threatening environmental sounds. They play a major part in the balance system, give important information about sudden change in an environment, both self and external, to avoid falling and injuries. In addition, they forming a major parts of a communication system [1]. Abnormal tympanic membrane (TM) might be seen as part of the disease that affect the EAC as in otitis externa, or the disease affecting the middle ear as chronic otitis media or it might be seen alone as in chronic myringitis [2].

The diagnosis of COM implies a permanent abnormalities of the pars tensa or flaccida. It is started with episode of acute otitis media (AOM) or otitis media (OM) with effusion in pediatrics [2]. The etiology of tympanic membrane perforation (TMP) are infection as acute otitis media, and trauma (including: a

direct blow to the ear can result in rupture TM, head trauma or iatrogenic from middle ear surgery or extrusion of grommet) [3]. It may be asymptomatic, but sometimes may be associated with normal hearing whereas a near-total perforation may cause a 35- to 40-dB CHL. This amount of hearing loss in the setting of a small perforation should raise concern for a disrupted or fixed ossicular chain [4]. A mucoid discharge from the ear, be it simple mucus or mucopurulent, should always alert the clinician to the likelihood of a tympanic membrane defect [3]. According to the four quadrants of tympanic membrane, Saliba classified the perforation size into four groups (I-IV) [5]. It is management by myringoplasty [3], and tympanoplasty [6].

Myringoplasty is defined as the surgical repairing of TM. In a majority of cases the damage is likely to be a persisting perforation of the drum but there are also situations where a thin or retracted drum may need to be reinforced (reinforcement myringoplasty) [3].

Tympanoplasty refer to surgical repairing of TM in combination within the cleft, like ossiculoplasty, middle ear exploration, or lysis of adhesions. Wullstein's classification of tympanoplasty depending on the underlying features of acoustic mechanics connected with each tympanoplasty, and it is classified from I to V types [6]. Myringoplasty indicated in prevention of recurrent otorrhea and/or recurrent ear Infections, lift water restrictions, attempt to improve hear and reinforcement of atelectatic TM [4,7].

Temporalis fascia is a common used material for repairing of TMP [8].

1 Platelet-rich plasma (PRP ) is a platelet concentrate used to fasting soft- tissue and hard-tissue healing. 1 PRF was first described by Choukroun et al., in France [9]. It refer to second -generation platelet concentrate, that shown to have several advantages over traditionally prepared PRP. These including 20 easy preparation and lack of biochemical handling of blood (autologous) [9]. PRF serve as a vehicle in carrying cells involve in tissue regeneration and a very significant slow sustain releasing of key growth factor for one week and up to 4 weeks [10]. It has been used in many fields including maxillofacial, sport medicine and otorhinolaryngology and plastic surgery as surgical adjuvants and tissue engineering tools [11].

18 Here, we try to assess the role of PRF in the healing of chronic central TMP. In addition, a comparison between the results of PRF and temporalis fascial graft in type 1 tympanoplasty.

## **METHODS**

### **Study setting and design**

A prospective-comparative study was conducted after an approval by committee board institute, the current study was conducted at Al-Yarmouk Teaching Hospital, Department of Otolaryngology, from

February 2018 to October 2019. A total of 56 cases with chronic TMP, 29 females (51.8%) and 27 males (48.2%) had been enrolled. All cases had routine ENT assessment including history, examination, audiological (pure tone audiometry and tympanometry) and radiological assessment (computed tomography) when indicated.

Patients were selectively distributed into two groups, group A (the study group) included those underwent myringoplasty with PRF alone, and group B (the control group) included those underwent underlay myringoplasty with temporalis fascial graft. The success had been determined by complete closure of the perforated tympanic membrane after three months.

#### **Inclusion criteria**

1. Chronic perforation of at least 3 months
2. Central perforation
3. Dry perforation of ( 4 weeks)
4. Air bone gap equal or less than 40 dB

#### **Exclusion criteria**

1. Inactive and active squamous COM or suspected middle ear pathology
2. Diabetic and immunocompromised patient
3. Patients on antiplatelet and anticoagulant
4. Patient with narrow EAC

#### **Preparation**

The procedures and their suspected outcomes had been explained to the patients and their relatives including the possible need for multiple sessions groups and informed consents were taken. Preoperative investigation included complete blood count, bleeding profile, renal function test, virology, chest X-ray and ECG in control group . In study group complete blood count and virology were ordered. All smoker patients had been instructed to quit smoking at least 2 weeks preoperatively.

#### **Procedure**

##### **Study group**

PFR was prepared from each patient own blood in which 9 ml of venous blood was collect in 10 ml plain glass tube and immediately centrifuging for 12 minutes at 2700 round per minutes. The patient ear was prepared with antiseptic( povidone iodine 10%), local anesthesia was infiltrated into EAC (2% lidocaine) in four quadrants and small wick soaked with 10% lidocaine inserted in EAC. After 5 minutes, via a transcanal approach, zero degree 2.7 mm rigid endoscope was introduced, and

refreshment of perforation edges were done with micro ear pick. PRF clot was divided by scissors. The cylinder and piston were used to prepare the plug while the thin compressor was used for membrane preparation. For perforations less than 50% PRF plug larger than the perforation is placed through the perforation in dumbbell fashion with most of plug in middle ear and covered with a short piece of PRF membrane in the EAC then with gelfoam. For perforation larger than 50% , the middle ear is filled with gelfoam pieces, one piece of PRF membrane placed under the tympanic membrane and another piece over it in sandwich fashion, then the EAC is filled with gelfoam.

### **Control group**

The procedure was done under GA with endotracheal tube. The involved ear prepared with shaving of 2 cm post auricular hair, antiseptic ( povidone iodine 10%), and patient draping was done. Under microscope the perforation edges refreshing were done. Post-auricular incision done with blade No.15, subcutaneous tissue and muscle dissected with monopolar diathermy. A retractor was used to elevate the skin. The layer above temporalis muscle was identified (loose areolar) and the dissection gone superiorly, keep the superficial to the muscle. Number 15 blade is used to make incision at lower border of temporalis muscle, then with a Freer elevator the fascia is elevated off the underlying muscle. About 2x2 cm piece of true fascia is harvested if possible. The graft left to dry and any visible muscle fibers were removed. Elevation of periosteal flap followed by placement of self-retaining retractors. Under microscope tympanomeatal flap elevated, and Chorda tympai was identified and preserved. The graft placed between handle of malleus and tympanic membrane remnant. Posteriorly, it was anchored under the annulus. <sup>10</sup> The middle ear filled with gelfoam and the flap unfolded. <sup>10</sup> The external auditory canal filled with gelfoam and small piece of bismuth iodoform paraffin paste (BIPP). Wounds closed in layers and mastoid dressing applied till the stiches are removed.

### **Post-operative and follow-up**

Post-operatively, the antibiotics and analgesia were described. Follow-up of group A in one month if the perforation still persist another session is performed up to three sessions. In patient with complete closure, PTA is performed after three months from the last session. Follow-up of group B at day 1 the dressing is changed and the patient discharged to be followed up at day 3, 5 and 7 for further dressing change and wound assessment. Stiches are removed after 7 days with instruction to keep the ear dry and avoid nose blowing. IPP is removed after one to two weeks. The next visit is arranged after one month and any remnant of gelfoam are suctioned for assessment. The patients are followed up every month up

to three months. If graft up take occurs, PTA is ordered. Successful cases in both groups are followed after 6 months for further evaluation.

### Statistical analysis

Data analysis by SPSS-25 (IBM, NY, USA, version 25). Data were presented as frequency, percentage, mean, SD and range. The different means were tested by Students-t-test or Paired-t-test for difference of paired observations. The different percentages were tested by Pearson Chi-square test or Fisher Exact test. Statistical significance considered when P value was  $\leq 0.05$ .

### RESULTS

Fifty six patients with chronic dry central perforation were included in the current study, they were categorized in two groups: (A study group, and B control group). The mean age (range) of group A was 35.2 (18-57yr), while in group B, it was 30.4 (16-55yr). Most of cases in group A were in the age group (30-39yr) 35.7%, while in group B the most common age group was (20-29yr) 39.3%. In group A (53.6%) of the patients were males (15 patients) while in group B (57.1%) were females (16 patients).

[Table 1]

**Table 1. Distribution of age and sex**

		Platelet rich fibrin		Temporalis facial graft		P value
		No	%	No	%	
Age (years)	<20 y	2	7.1	3	10.7	0.786
	20-29	8	28.6	11	39.3	
	30-39	10	35.7	9	32.1	
	40-49	6	21.4	3	10.7	
	$\geq 50$ y	2	7.1	2	7.1	
	Mean $\pm$ SD (Range)	35.2 $\pm$ 9.9 (18-57)		30.4 $\pm$ 10.3 (16-55)		
Sex	Male	15	53.6	12	42.9	0.422
	Female	13	46.4	16	57.1	

In group-A most of cases were right ear (15 patients 53.6%), as well as for group B (19 patients 67.9%). The mean duration of perforation was 2.8 years in group-A and 2.4 years in group-B. The perforation located anteriorly in 14 cases (50%) in group-A and in 8 cases (28.6%) in group-B. Posteriorly located perforations were 9 patients (32.1%) in group-A and 13 cases (46.4%) in group-B. The perforation was large in 5 cases (17.9%) in group-A and in 7 cases (25%) in group-B. The perforation size was  $\leq 25\%$  in 12 cases (42.9%) from group-A, as well as 8 cases from group B(28.6%), between 25- 50% in 11 cases from group-A (39.3%) and 13 cases from group-B (46.4%), and between 50- subtotal in 5 cases

from group-A (17.9%) and 7 (25%) from group-B. In both groups the most likely cause of perforation was infection, 19 cases in group-A (67.9%) and 24 cases in group B (85.7%) [Table 2].

**Table 2. Distribution of the affected side ,duration of perforation, size, and most probable cause**

		Platelet rich fibrin (A)		Temporalis facial graft (B)		P value
		No	%	No	%	
Side	Right	15	53.6	19	67.9	0.274
	Left	13	46.4	9	32.1	
Duration since perforation (years)	<1 y	5	17.9	5	17.9	0.798
	1-4	16	57.1	18	64.3	
	≥5 y	7	25.0	5	17.9	
	Mean± SD (Range)	2.8±2.1 (4m-7y)		2.4±1.7 (5m-6y)		
Site of perforation	Anterior	14	50.0	8	28.6	0.065
	Posterior	9	32.1	13	46.4	
	Large	5	17.9	7	25.0	
Size of perforation	≤25%	12	42.9	8	28.6	0.522
	>25-50%	11	39.3	13	46.4	
	>50- subtotal	5	17.9	7	25.0	
Cause of perforation	Trauma	9	32.1	4	14.3	0.114
	Infection	19	67.9	24	85.7	

There were 7 smoker cases in group-A and 5 (17.1%) in group-B. Regarding the previous ENT surgeries, 5 (17.9%) patients in each group had history of tonsillectomy, adenolectomy in group-B two cases only (7.1%) and septoplasty in one case (3.6%) from group-A and 2 (7.1%) from group-B [Table 3].

**Table 3. Previous surgical history and history of smoking.**

		Platelet rich fibrin		Temporalis facial graft		P value
		No	%	No	%	
Smoking	Current & Ex-smoker	7	25.0	5	17.9	0.515
	Not	21	75.0	23	82.1	
Previous ENT surgeries	Tonsillectomy & Adenolectomy	5	17.9	5	17.9	0.552
	Adenolectomy	-	-	2	7.1	
	Septoplasty	1	3.6	2	7.1	
	No	22	78.5	19	67.9	



Regarding the number of the sessions, 16 patients (57.1%) in group-A had only one session, 2 cases (7.1%) had two sessions and 10 had three sessions (35.8%), while all the patients in group B 28 (100%) patients had only one procedure. The healing was successful in 20 cases (71.4%) in group-A and 23 patients (82.1%) in group-B (P=0.342). The duration of the procedure in group-A from the time anesthetic infiltration till gelfoam insertion in EAC was between 15-30 minutes (mean 21.1 min). In group-B, the duration from induction of anesthesia till wound closure was between 90-120 minutes (mean 105.7min). Post-operative complications in group-A was one case of tympanosclerosis (3.6%). Other complications of sudden sensorineural hearing loss, taste disturbance, wound infection, lateralization of the graft and cholesteatoma had not been recorded [Table 4].

The mean air bone gap closure for group-A was 8.3 dB, and for group-B 10.3 dB. [Table 5]

**Table 4. Number of sessions, healing, duration of procedure and post-operative complications**

		Platelet rich fibrin (A)		Temporalis facial graft (B)		P value
		No.	%	No.	%	
Number of sessions	1	16	57.1	28	100.0	-
	2	2	7.1	-	-	
	3	10	35.8	-	-	
Healing	Positive	20	71.4	23	82.1	0.342
	Negative	8	28.6	5	17.9	
Duration of procedure	Mean ±SD(range)	21.1 ± 4.4 (15-30min)		105.7± 3.7 (90-120min)		0.0001*
Complications	Sclerosis	1	3.6	-	-	-
	No	27	96.4	28	100	

**Table 5. Pre-operative and post-operative air bone gap**

	Air-Bone Gap	Procedure applied			
		Platelet rich fibrin (A)		Temporalis facial graft (B)	
		Healing		Healing	
		Positive	Negative	Positive	Negative
Pre-op		15.1±11.1	21.4±9.7	19.1±9.4	27.6±7.7
Post-op	ABG	6.8±5.5	21.4±9.7	8.8±5.3	27.6±7.7

P value	0.0001*	-	0.014*	-
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The healing rate was higher in the right ear in group-A (80%) while in group-B it was higher in left ear (100%). In group-A the higher successful rate was for perforation of less than one year (80%), while in group-B it was higher for perforation of  $\Rightarrow$  5 years (100%). In both groups the success rate was higher in posteriorly located perforation. In group-A 88.9% and in group-B was 92.1%. The healing for perforation  $\leq$ 25% was (83.3%) in group-A and (100%) in group-B, for perforation of  $>$ 25% $\leq$ 50% the healing in group-A was (63.6%) and in group-B was (76.9%), while in perforation larger than 50%, the healing in group-A was (60%) and in group-B was (71.4%). In both groups the success was higher for perforations caused by trauma, 77.8% (seven from nine cases) in group-A and 100% (four patients) in group-B [Table 6].

**Table 6. The healing percentage according to different variables**

		Healing				P value
		Positive		Negative		
		No	%	No	%	
<b>Platelet rich fibrin</b>						
Side	Right	12	80.0	3	20.0	0.281
	Left	8	61.5	5	38.5	
Duration since perforation (years)	<1 y	4	80.0	1	20.0	0.889
	1-4	11	68.8	5	31.3	
	$\geq$ 5 y	5	71.4	2	28.6	
Site of perforation	Anterior	9	64.3	5	35.7	0.538
	Posterior	8	88.9	1	11.1	
	Large	3	60.0	2	40.0	
Size of perforation (%)	$\leq$ 25%	10	83.3	2	16.7	0.477
	$>$ 25-50%	7	63.6	4	36.4	
	$>$ 50-subtotal	3	60.0	2	40.0	
Cause of perforation	Trauma	7	77.8	2	22.2	0.609
	Infection	13	68.4	6	31.6	
Number of sessions	1	16	100	-	-	-
	2	2	100	-	-	
	3	2	20.0	8	80.0	
<b>Temporalis facial graft</b>						
Side	Right	14	73.7	5	26.3	0.090
	Left	9	100.0	-	-	
Duration since perforation (years)	<1 y	4	80.0	1	20.0	0.513
	1-4	14	77.8	4	22.2	

	≥5 y	5	100.0	-	-	
Site of perforation	Anterior	6	75.0	2	25.0	0.578
	Posterior	12	92.3	1	7.7	
	Large	5	71.4	2	28.6	
Size of perforation (%)	≤25%	8	100.0	-	-	0.282
	>25-50%	10	76.9	3	23.1	
	>50-subtotal	5	71.4	2	28.6	
Cause of perforation	Trauma	4	100.0	-	-	0.314
	Infection	19	79.2	5	20.8	
Number of sessions	1	23	82.1	5	17.9	-
	2	-	-	-	-	

## DISCUSSION

Its ease of preparation from small amount of venous blood and ease of use under local anesthesia, being strictly autologous, and being available at the grafting site for up to 4 weeks followed by complete resorption, PRF can act as a scaffold for tympanic membrane regeneration with its content of growth factors.

The current study recorded high frequency of chronic inactive mucosal disease in female 29 patients (51.7%) which is consistent to study done by Saliba (2008) reported a greater prevalence in women in comparison to men [5].

The majority of cases in study group were in the age between 30-39 years 10 of 28 patients in contrast to previous study done by Zaman and Omar (2019), myringoplasty using temporalis fascia, in which the main age group was 18-27 years which was consistent with control group main age distribution 39.3% 11 of 28 patients [12].

In current study the success rate for both group was higher in small size perforation =25% [ A 83.3% ,B 100%] and decrease with increase in perforation size in both groups 25-50% [ A 63.3%, B 76.9 % ], > 50% [A 60%, B 71.4 %], in agreement with a study done by Ezzat et al. (2019), who declared a success rate for small size 93.3 %, for medium size 87.5% and for large size 71.4% [13].

In group A, The decrease in success rate with increased size of perforation may be explained by rapid resorption of the graft in comparison to the large size of perforation, this causes a deprivation of perforation margin from the scaffold and its content of growth factors.

In this study, the success rate for posterior perforation in both groups was greater than anteriorly located perforation (88.9% for group A, 92.3% for group B), in agreement with Das et al., (2015) study, in which the success rate for posterior perforation was higher than anterior perforation [14].

In current study the air bone gap gain for both groups (group A 8.3 dB, group B 10.3 dB) were higher than study done by Yegin et al., (2016) in which the air bone gap gain for temporalis facial graft was 4.68 dB [15].

El-Anwar et al. said that the effect of PRF in chronic small size perforation only (2 mm which is half of one quadrant according to Saliba classification) and showed a success rate 84% in 25 ears. In agreement with the current study for perforation of  $\leq 25\%$  in which the success was 83.3% in 12 patients. A larger sizes of perforation had not been included in El-Anwar study [16].

Saliba, (2008) used a combination of fat and hyaluronic acid as a minimal invasive surgery for repairing of TMP in 21 patients with successful closure in 17 patients (81%) and only one patient required a second section after 2 months, in compare to (71.4 %) success rate in current study with PRF.

Two unsuccessfull repair were attributed to post-operative graft infection, a complication did not observed in current study for PRF which may be attributed to activated WBC in PRF graft, a procedure that also has advantage of absence harvesting site scar [5].

The growth factors had been evaluated by Kanemaru et al (2011). They used basic fibroblast growth factor (b-FGF) with gelatin sponge and fibrin glue in 53 patients with chronic perforation. The treatment could be repeated up to 4 times at 3 weeks interval with success rate of 98.1% (52:53) which was higher than current study success rate (71.4%).

However the preparation of b-FGF is much more complicated process and more expensive than PRF [17].

Sharma et al., (2018) compared the results of adding PRF as a sealant over temporalis facial graft in 50 patients and 50 without PRF. They concluded that postoperative graft uptake was better in cases with PRF (success rate of 92%) while without PRF the success was 64%. The postoperative infection rate was lower in the same group. This was higher than current study with success rate of temporalis facial graft alone was 82.3% [18].

## CONCLUSION

The success rate with temporalis fascia (82.3%) is higher than with PRF (71.4%). The resulting tympanic membrane with PRF is more transparent and has a natural appearance than the with temporalis fascial graft, which support the role of growth factors within the scaffold. PRF lacks the harvesting site morbidity, can be done under local anesthesia and is relatively more abundant material than fascia or cartilage. The success rate decreases with increase in perforation size in both groups and repeated sessions are more likely to be required with PRF. PRF is recommended as a graft in myringoplasty for

small perforation size, for patients in whom general anesthesia carry a risk, and patients who refuse the surgery under general anesthesia. For perforation size between 25-50%, the use of sandwich fashion of PRF is advisable in further researches which may provide a prolonged time of contact with perforation margins.

Disclosure

None

## REFERENCES

1. Valentine P, Wright T. Anatomy and Embryology of the External and Middle Ear . In Watkinson JC and Clarke RW (editors), Scott-Brown's Otorhinolaryngology, Head and Neck Surgery, 8<sup>th</sup> edition, CRC Press,2018. 525-539.
2. Dornhoffer JL, Gluth MB. Reconstruction Of The Tympanic Membrane And Ossicular Chain . In Johnson JT and Rosen CA (editors), Baily's Head and Neck Surgery, Otolaryngology. 5th edition, Lippincott Williams and Wilkins, 2013. 2465-2466.
3. Browning GG, Weir J, Kelly G, Swan IRC. Chronic Otitis Media. In John C Watkinson JC, Clarke RW (editors). Scott-Brown's Otorhinolaryngology, Head and Neck Surgery, 8<sup>th</sup> edition, CRC Press 2018. 977-978 .
4. Huins C, Lavy J. Myringoplasty. In Watkinson JC and Clarke RW (editors), Scott-Brown's Otorhinolaryngology, Head and Neck Surgery, 8<sup>th</sup> edition, CRC Press 2018.1021-1023.
5. Chiffer R, Chen D: Myringoplasty And Tympanoplasty. In Myers EN and Snyderman CH. Operative Otolaryngology-Head and Neck Surgery, 3<sup>rd</sup> edition, Elsevier 2018. 880-881.
6. Saliba I. Hyaluronic Acid Fat Graft Myringoplasty: How We Do It. Clinical Otolaryngology;2008;33:610-614.
7. Chole RA. Chronic Otitis Media, Mastoiditis, and Petrositis. In Flint et al (editors). Cumming Otolaryngology, head and Neck Surgery, 6<sup>th</sup> edition, Saunders, 2015. 2140.
8. Adams ME, El-Kashlan HK. Tympanoplasty and Ossiculoplasty. In Flint et al (editors). Cumming Otolaryngology, Head and Neck Surgery, 6<sup>th</sup> edition, Saunders, 2015. 2178-2179.
9. Raja VS, Naidu EM. Platelet-Rich Fibrin: Evolution of A Second-Generation Platelet Concentrate Indian J Dent Res, 2008.19:42-46.
10. Chang YC, Zhao JH: Effects Of Platelet-Rich Fibrin On Human Periodontal Ligament Fibroblasts And Application For Periodontal Infrabony Defects. Australian Dental Journal. 2011.56 :365-371.

11. Bielecki T, Dohan Ehrenfest DM. Platelet-rich plasma (PRP) and Platelet-Rich Fibrin (PRF): Surgical Adjuvants, Preparations For In Situ Regenerative Medicine And Tools For Tissue Engineering. *Curr Pharm Biotechnol*, 2012;13:1121–1130.
12. Zaman A, Omar S. Myringoplasty Using Temporalis Fascia And Its Clinical Outcome. *International Journal of Otorhinolaryngology and Head and Neck Surgery*. 2019;5:1539-1542 .
13. Saleh EM, Salem MAM, Nemr SHA. Impact Of Site And Size Of Pars Tensa Tympanic Membrane Perforation On The Success Rate Of Myringoplasty .*Egyptian Journal of Otolaryngology* , 2019;35:339-346.
14. Das A, Sen B, Ghosh D, Sengupta A. Myringoplasty: Impact of Size and Site of Perforation on the Success Rate. *Indian Journal Otolaryngol Head Neck Surgery*. 2015;67:185–189.
15. Yegin Y, Yaz ZM, Çelik M, Güneş S, Sayın I, Kayhan FT. Comparison of Temporalis Muscle Fascia And Full-Thickness Cartilage Grafts In Type 1 Tympanoplasty. *Nternational Journal of Clinical and Experimental Medicine* 2016;9:8731-8736.
16. El-Anwar MW, Elnashar I, Foad YA. Platelet-rich plasma myringoplasty: A New Office Procedure For The Repair Of Small Tympanic Membrane Perforations. *Ear Nose Throat Journal*. 2017;96:312-326.
17. Kanemaru SI, Hiroo U, Yoshiharu K, Tatsuo N, Shigeru H, Juichi I. Regenerative Treatment for Tympanic Membrane Perforation. *Otology & Neurotology*. 2011;32:1218-1223.
18. Sharma D, Mohindroo S, Azad RK. Efficacy Of Platelet Rich Fibrin In Myringoplasty. *International Journal of Otorhinolaryngology and Head and Neck Surgery*. 2018;4:677-681.