The Journal of Laryngology & Otology

cambridge.org/jlo

The role of platelet-rich plasma in microlaryngeal surgery: a randomised, controlled trial

A Gaafar[1], A Eldeghiedy[2], R ElMaghraby[3], I Nouh[1] and M Donia[1]

[1]Department of Otolaryngology, Head and Neck Surgery Unit, [2]Clinical Pathology and [3]Department of Otolaryngology, Phoniatrics Unit, Alexandria Faculty of Medicine, Alexandria University, Alexandria, Egypt

Abstract

Objective. Platelet-rich plasma has gained interest over the two last decades, mainly because of its role in regenerative medicine. This work aimed to assess the role of intra-operative local application of platelet-rich plasma gel in the improvement of quality of voice after microlaryngeal surgery.

Method. This was a prospective comparative study that included 40 patients undergoing microlaryngeal surgery for benign vocal fold lesions. There were two groups divided equally into study group A and control group B. The assessment of voice was performed by videostroboscopy and acoustic analysis pre-operatively and at two weeks and one and three months post-operatively.

Results. The data demonstrated that all the stroboscopic and acoustic parameters showed significant improvement in both groups. Group A showed significant improvement regarding acoustic parameters at the third post-operative follow up when compared with group B.

Conclusion. Platelet-rich plasma has a beneficial effect on voice quality following microlaryngeal surgery based in particular on acoustic parameters.

Introduction

Vocal folds are ultrasensitive structures. The symmetry of their ultrastructure and biomechanical properties is necessary for a normal voice. Therefore, the voice will be affected by different vocal fold conditions.[1] Benign lesions arising in the vocal fold commonly cause dysphonia, which is usually corrected by microlaryngeal surgery. They include polyps, nodules, cysts and Reinke's oedema.[2]

The major causes of dysphonia after unsuccessful microlaryngeal surgery are adhesions and scarring. Once a vocal fold scar has formed, treatment becomes an extremely difficult problem.[3] The medial microflap technique is an ideal method to remove many intracordal lesions. It allows the removal of the pathology with maximum preservation of the mucosa.[4] In the future, resection of vocal fold lesions may be followed with a simple injection of growth factors or stem cells to decrease scarring and to restore the layered architecture of the vocal fold.[5]

Platelet-rich plasma has gained interest over the two last decades, mainly because of its role in regenerative medicine. However, its use in otorhinolaryngology is not yet common, and its application needs further study.[6,7] Platelet-rich plasma has the advantage of being an autologous product. Therefore, there is no risk of cross contamination, disease transmission or immune reaction.[8–10]

Platelets, the main component of platelet-rich plasma, comprise more than 1500 protein-based bioactive factors. Besides its high platelet concentration, platelet-rich plasma contains a high concentration of growth factors that are useful for wound healing and epithelisation.[11–13]

Several studies on animals concluded that platelet-rich plasma has favourable histological outcomes regarding the healing of vocal fold injuries. They recommend further clinical studies to assess the effect of these findings on functional outcomes.[14] Based on recent studies, there has been increasing recognition of the need to study the effect of platelet-rich plasma on human vocal folds.[15]

Materials and methods

This study included 40 patients undergoing microlaryngeal surgery for benign vocal fold lesions (vocal fold polyp, vocal fold nodule, vocal fold Reinke's oedema and vocal fold cyst) at the Alexandria University Hospital in a one year period. Patients who were excluded from the study were those unfit for anaesthesia or with history of previous vocal fold surgery. Ethical approval was obtained from the ethical committee and informed consent was taken from all patients.

Main Article

Dr A Gaafar takes responsibility for the integrity of the content of the paper

Cite this article: Gaafar A, Eldeghiedy A, ElMaghraby R, Nouh I, Donia M. The role of platelet-rich plasma in microlaryngeal surgery: a randomised, controlled trial. *J Laryngol Otol* 2022;**136**:737–741. https://doi.org/10.1017/S0022215121004564

Accepted: 9 September 2021
First published online: 10 February 2022

Key words:
Vocal Cords; Platelet-Rich Plasma; Voice Quality

Author for correspondence:
Dr A Gaafar, Department of Otolaryngology, Head and Neck Surgery Unit, Faculty of Medicine, Alexandria University, Champollion Street, Alexandria 21131, Egypt
E-mail: alaa.gaafar@alexmed.edu.eg

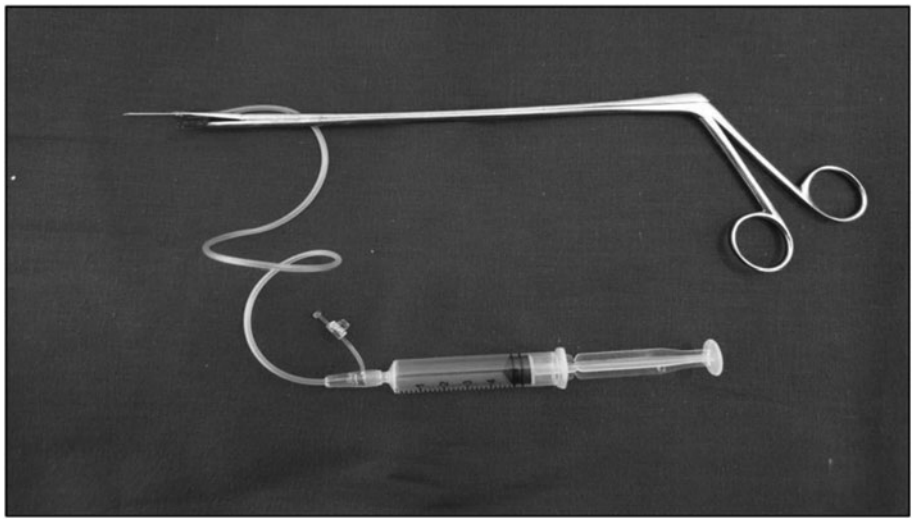

Fig. 1. Injection set assembled for platelet-rich plasma application.

Patients were block-randomised into 2 groups: each group included 20 patients. Group A underwent microlaryngeal surgery for benign vocal fold lesions followed by intra-operative application of platelet-rich plasma gel. Group B was the control group (who underwent microlaryngeal surgery without application of platelet-rich plasma gel).

In group A, platelet-rich plasma was prepared immediately prior to the operation using whole blood by venipuncture in 1 ml acid citrate dextrose in a sterile vacutainer. Centrifugation of blood was done using a soft spin (250 g) for 10 minutes (Hettich zentrifugan protofix 32A, Tuttlingen, Germany). The plasma supernatant containing platelets was transferred into another sterile tube (without anticoagulant, Lab Companion, Daejeon, Korea). The tube was centrifuged at a higher speed (300 g for 10 minutes) to obtain platelet concentrate. At the bottom of the tube, a platelet pellet was formed. Removal of the platelet-poor plasma was performed. The platelet pellet was suspended in 2 ml of platelet-poor plasma by vortex (vortex mixer, Gemmy Industrial, San Chung, Taiwan). The platelet-rich plasma was mixed with calcium gluconate and thrombin (at a tenth the volume of the platelet-rich plasma) intra-operatively to form platelet-rich plasma gel. Vocal fold polyps and nodules were excised using microlaryngeal instruments. Vocal fold cysts and Reinke's oedema were excised using the medial microflap technique.

In group A, platelet-rich plasma gel was applied using a scalp vein infusion set (Figure 1). Its wings were trimmed to allow good exposure during the injection. The main surgeon carried the distal end of the infusion set by laryngeal forceps to the site of injection. The assistant injected the freshly mixed platelet-rich plasma with thrombin in the proximal end of the infusion set. Application of platelet-rich plasma was performed on the raw area in cases of vocal fold polyps and nodules, whereas application of platelet-rich plasma was performed under elevated microflaps in cases of vocal fold cysts and Reinke's oedema (Figure 2).

The assessment of voice was performed by videostroboscopy (using KayPentax TM RLS9100B, Tokyo, Japan) and acoustic analysis (using the following software: Multi-Dimensional Voice Program, model 5105-KayPentax included in Computerised Speech Lab model 4500-KayPentax). One assessment was performed pre-operatively, and three other follow-up points were measured at two weeks, one month and three months.

The following stroboscopic parameters were assessed: mucosal wave (absent, decreased, normal or increased); glottal gap (measured in millimetres); symmetry (asymmetrical or symmetrical); and regularity (irregular or regular). The analysed vocal parameters were: fundamental frequency, jitter percentage, shimmer and noise to harmonic ratio.

Results

Our study included 40 patients: 19 were male (47.5 per cent) and 21 were female (52.5 per cent). The mean age was 35.2 ± 10.3 years.

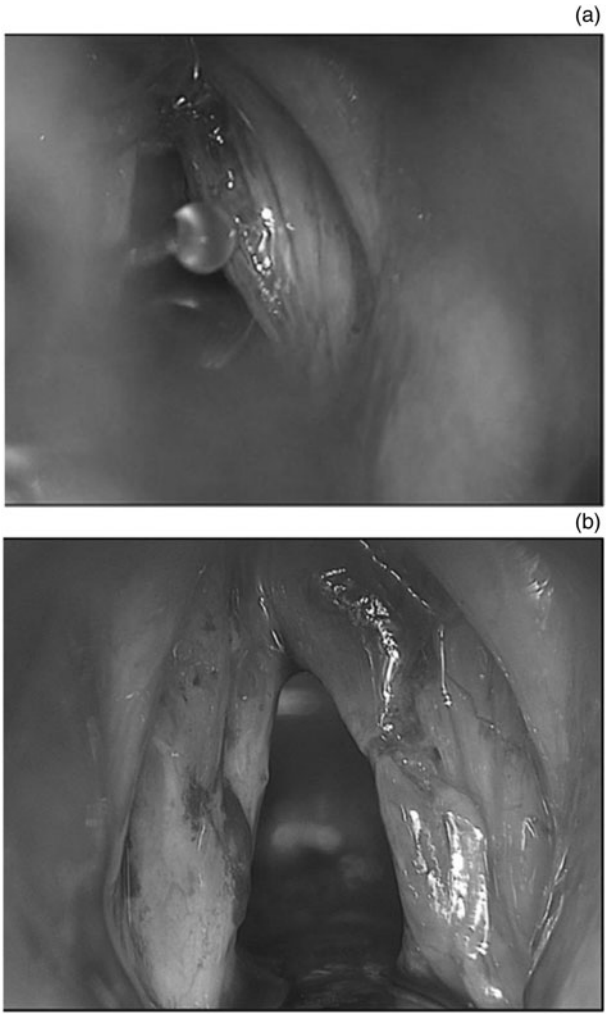

Fig. 2. A patient after excision of a vocal fold cyst using the microflap technique showing: (a) application of platelet-rich plasma under the microflap and (b) properly sealed mucosal flap after the application of platelet-rich plasma.

https://doi.org/10.1017/S0022215121004564 Published online by Cambridge University Press

Most of the patients (31 patients, 77.5 per cent) were non-professional voice users (students, farmers and housewives), whereas 9 patients (22.5 per cent) were professional voice users (teachers and muezzins). Most of the patients (23 patients, 57.5 per cent) were non-smokers; 17 patients (42.5 per cent) were smokers.

The benign vocal fold lesions comprised 16 cases of cysts (40 per cent), followed by 12 cases of polyps (30 per cent), 8 cases of nodules (20 per cent) and 4 cases of Reinke's oedema (10 per cent). During the follow-up period, there were no observed complications associated with the platelet-rich plasma injection or the operation itself.

Statistical analysis

Data were analysed using SPSS® statistical software (version 20.0). Qualitative data were described using number and percentage. The Kolmogorov–Smirnov test was used to verify the normality of distribution. Quantitative data were described using range (minimum and maximum), mean, standard deviation, median and interquartile range. The significance of the obtained results was judged at the 5 per cent level.

Stroboscopic parameters

A comparison between the two groups showed no significant difference in the improvement of the stroboscopic parameters in all follow-up periods.

Fundamental frequency

A comparison between the two groups showed no significant difference in the improvement of the fundamental frequency in all follow-up periods.

Jitter percentage, shimmer and noise to harmonic ratio

When comparing both groups, there were significant differences regarding jitter percentage, shimmer and noise to harmonic ratio at the third post-operative follow up. Group A showed better improvement regarding these parameters (Tables 1, 2 and 3).

When comparing the cases where the microflap technique was used (cysts and Reinke's oedema) in both groups, the group with platelet-rich plasma application showed significant improvement regarding jitter percentage ($p = 0.003$), shimmer ($p = 0.018$) and noise to harmonic ratio ($p = 0.027$) at the third follow up. On the other side, comparing the cases where the microflap technique was not used (polyps and nodules) in both groups, there was no significant difference in all periods (Tables 4–9).

Table 1. Comparison between the two studied groups according to jitter percentage

Jitter	Group A*	Group B†	*P*-value
Pre-operative (mean ± SD; %)	3.0 ± 1.19	3.54 ± 1.03	0.140
Two weeks post-operatively (mean ± SD; %)	2.28 ± 0.86	2.64 ± 0.89	0.213
One month post-operatively (mean ± SD; %)	2.01 ± 0.76	2.28 ± 0.66	0.234
Three months post-operatively (mean ± SD; %)	1.58 ± 0.59	2.12 ± 0.67	0.011‡

*$n = 20$; †$n = 20$; ‡Statistically significant at $p \leq 0.05$. Student's *t*-test was used. The *p*-value shows the comparison between the two studied groups (A and B). SD = standard deviation

Table 2. Comparison between the two studied groups according to shimmer

Shimmer	Group A*	Group B†	*P*-value
Pre-operative (mean ± SD; %)	5.79 ± 1.96	6.19 ± 1.67	0.492
Two weeks post-operatively (mean ± SD; %)	3.92 ± 1.45	4.71 ± 1.81	0.135
One month post-operatively (mean ± SD; %)	3.52 ± 1.28	3.98 ± 1.42	0.290
Three months post-operatively (mean ± SD; %)	2.88 ± 1.09	3.87 ± 1.73	0.037‡

*$n = 20$; †$n = 20$; ‡Statistically significant at $p \leq 0.05$. Student's *t*-test was used. The *p*-value shows the comparison between the two studied groups (A and B). SD = standard deviation

Table 3. Comparison between the two studied groups according to noise to harmonic ratio

Noise to harmonic ratio	Group A*	Group B†	*P*-value
Pre-operative (mean ± SD)	0.19 ± 0.04	0.20 ± 0.05	0.532
Two weeks post-operatively (mean ± SD)	0.16 ± 0.03	0.17 ± 0.03	0.367
One month post-operatively (mean ± SD)	0.16 ± 0.03	0.17 ± 0.02	0.195
Three months post-operatively (mean ± SD)	0.13 ± 0.04	0.16 ± 0.04	0.045‡

*$n = 20$; †$n = 20$; ‡Statistically significant at $p \leq 0.05$. Student's *t*-test was used. The *p*-value shows the comparison between the two studied groups (A and B). SD = standard deviation

Discussion

Phonomicrosurgery is an effective method that results in the improvement of voice in patients with benign vocal fold lesions.[16] Objective evaluation of the voice before and after surgery helps to evaluate the effectiveness of treatment. Post-operative adhesions and scarring alter the mucosal wave dramatically and cause stiffness in lamina propria. As mucosa is the main oscillator in the vocal fold,[17] scarring causes dysphonia following ineffective phonomicrosurgery. Once vocal fold scarring has taken place, it is a very difficult problem to treat.[3] Platelet-rich plasma has recently drawn considerable attention in regenerative medicine as a source for growth factors.[6] The benefit of platelet-rich plasma is attributed to its high growth factor level.

In contrast to animal studies that adopted histological analysis for studying the effect of platelet-rich plasma application, it is not possible in human studies to observe the wound healing histopathologically. Instead, videostroboscopic and

Table 4. Comparison between the two studied groups according to jitter for platelet-rich plasma application under microflaps

Jitter	Group A*	Group B†	*P*-value
0 (mean ± SD; %)	3.55 ± 1.38	4.03 ± 0.94	0.377
1 (mean ± SD; %)	2.62 ± 1.0	3.20 ± 0.85	0.179
2 (mean ± SD; %)	2.26 ± 0.93	2.73 ± 0.61	0.199
3 (mean ± SD; %)	1.60 ± 0.76	2.62 ± 0.57	0.003‡

*$n = 10$; †$n = 10$. Student's *t*-test was used. The *p*-value shows the comparison between the two studied groups (A and B). SD = standard deviation

https://doi.org/10.1017/S0022215121004564 Published online by Cambridge University Press

Table 5. Comparison between the two studied groups according to jitter for platelet-rich plasma application on raw area

Jitter	Group A*	Group B†	*P*-value
0 (mean ± SD; %)	2.46 ± 0.64	3.04 ± 0.90	0.113
1 (mean ± SD; %)	1.95 ± 0.56	2.07 ± 0.50	0.614
2 (mean ± SD; %)	1.75 ± 0.45	1.83 ± 0.33	0.674
3 (mean ± SD; %)	1.56 ± 0.40	1.62 ± 0.29	0.705

*$n = 10$; †$n = 10$. Student's *t*-test was used. The *p*-value shows the comparison between the two studied groups (A and B). SD = standard deviation

Table 6. Comparison between the two studied groups according to shimmer for platelet-rich plasma application under microflaps

Shimmer	Group A*	Group B†	*P*-value
0 (mean ± SD; %)	6.77 ± 2.37	7.29 ± 1.57	0.566
1 (mean ± SD; %)	4.62 ± 1.77	6.09 ± 1.49	0.060
2 (mean ± SD; %)	4.11 ± 1.57	4.89 ± 1.48	0.266
3 (mean ± SD; %)	3.07 ± 1.40	4.95 ± 1.81	0.018‡

*$n = 20$; †$n = 20$; ‡Statistically significant at $p \leq 0.05$. Student's *t*-test was used. The *p*-value shows the comparison between the two studied groups (A and B). SD = standard deviation

Table 7. Comparison between the two studied groups according to shimmer for platelet-rich plasma application on raw area

Shimmer	Group A*	Group B†	*P*-value
0 (mean ± SD; %)	4.82 ± 0.61	5.09 ± 0.85	0.421
1 (mean ± SD; %)	3.21 ± 0.47	3.32 ± 0.65	0.659
2 (mean ± SD; %)	2.94 ± 0.47	3.07 ± 0.51	0.544
3 (mean ± SD; %)	2.69 ± 0.68	2.79 ± 0.66	0.737

*$n = 20$; †$n = 20$. Student's *t*-test was used. The *p*-value shows the comparison between the two studied groups (A and B). SD = standard deviation

Table 8. Comparison between the two studied groups according to noise to harmonic ratio for platelet-rich plasma application under microflaps

Noise to harmonic ratio	Group A*	Group B†	*P*-value
0 (mean ± SD)	0.21 ± 0.05	0.21 ± 0.05	0.820
1 (mean ± SD)	0.17 ± 0.04	0.18 ± 0.03	0.588
2 (mean ± SD)	0.16 ± 0.04	0.18 ± 0.03	0.145
3 (mean ± SD)	0.14 ± 0.04	0.18 ± 0.03	0.027‡

*$n = 10$; †$n = 10$; ‡Statistically significant at $p \leq 0.05$. Student's *t*-test was used. The *p*-value shows the comparison between the two studied groups (A and B). SD = standard deviation

Table 9. Comparison between the two studied groups according to noise to harmonic ratio for platelet-rich plasma application on raw area

Noise to harmonic ratio	Group A*	Group B†	*P*-value
0 (mean ± SD)	0.17 ± 0.02	0.18 ± 0.03	0.289
1 (mean ± SD)	0.15 ± 0.02	0.16 ± 0.01	0.245
2 (mean ± SD)	0.16 ± 0.02	0.16 ± 0.01	0.989
3 (mean ± SD)	0.13 ± 0.04	0.14 ± 0.03	0.539

*$n = 20$; †$n = 20$. Student's *t*-test was used. The *p*-value shows the comparison between the two studied groups (A and B). SD = standard deviation

acoustic parameters are the alternative measures of observation, and these were performed in our study.

Our study was conducted on 40 patients who presented to our institution. Patients were randomly categorised into 2 groups: each group included 20 patients. Group A was the study group that underwent microlaryngeal surgery for benign vocal fold lesions followed by intra-operative application of platelet-rich plasma gel. Group B was the control group without the application of platelet-rich plasma gel.

When comparing both groups, there were significant differences regarding jitter percentage, shimmer and noise to harmonic ratio at the third post-operative follow up. Group A showed greater improvement with regard to these parameters. These results may suggest that platelet-rich plasma has a beneficial effect on voice quality based on vocal parameters in particular.

When comparing the cases where the microflap technique was used (cysts and Reinke's oedema) in both groups, the group with platelet-rich plasma application showed significant improvement. These results show that platelet-rich plasma may have a better effect on voice improvement if it is applied under microflaps rather than on the raw surface. This is perhaps explained by the fact that platelet-rich plasma is applied and not injected, and microflaps may provide a good scaffold for platelet-rich plasma, giving time for platelets to release growth factors and for platelet-rich plasma to solidify and act as a sealant for the mucosal flap. Additionally, the elevation of the microflap creates more trauma because of more dissection in the basement membrane zone. Cutting of anchoring fibres that can be seen running from the lamina densa into the superficial layer of the lamina propria impairs the proper healing process.[18] Furthermore, platelet-rich plasma helps in maintaining the layered architecture of vocal folds as shown in the study by Woo *et al.*[15] This may explain the fact that cases performed with the microflap technique obtain better healing with platelet-rich plasma application with better voice outcomes.

Since we had a short follow-up period post-operatively, this might be a reason that our results were not as dramatic. If the follow up was continued until six months, our results might be better in terms of improvement in stroboscopic findings.

- Platelet-rich plasma has gained interest over the two last decades, mainly because of its role in regenerative medicine
- Platelet-rich plasma is an autologous product with a high concentration of growth factors useful for wound healing and epithelisation
- This study assessed application of the platelet-rich plasma gel in quality of voice improvement after microlaryngeal surgery
- Most of the parameters of acoustic analysis showed significant improvement when platelet-rich plasma was applied

Similar to the study by Kim *et al.*,[19] none of our cases showed any complications associated with the platelet-rich plasma injection during the follow-up period. These results show platelet-rich plasma to be a safe autologous material for injection into vocal folds.

Conclusion

Platelet-rich plasma is an autologous material that can be used safely without the fear of any complications that may threaten voice quality. It has a beneficial effect on voice quality particularly in regard to vocal parameters. It shows better results if applied under microflaps rather than on the surface of vocal folds.

Further studies with a larger sample size and different ethnic groups are required to validate the results of our study. Long-term post-operative evaluation should be performed to evaluate the effect of platelet-rich plasma on stroboscopic parameters. Further studies are needed to evaluate the role of platelet-rich plasma in other vocal fold disorders, such as vocal fold sulcus, vocal fold scar and vocal fold atrophy.

https://doi.org/10.1017/S0022215121004564 Published online by Cambridge University Press

Additionally, platelet-rich plasma may be evaluated as a safe material for injection for vocal fold medialisation in cases with glottal insufficiency.

Competing interests. None declared

References

1 Zhang Z. Cause-effect relationship between vocal fold physiology and voice production in a three-dimensional phonation model. *J Acoust Soc Am* 2016;**139**:1493
2 Cohen SM, Dupont WD, Courey MS. Quality-of-life impact of non-neoplastic voice disorders: a meta-analysis. *Ann Otol Rhinol Laryngol* 2006;**115**:128–34
3 Mortensen M. Laryngeal steroid injection for vocal fold scar. *Curr Opin Otolaryngol Head Neck Surg* 2010;**18**:487–91
4 Gonzalez-Herranz R, Amarillo E, Hernandez-Garcia E, Montojo Woodeson J, Plaza-Mayor G. Amplitude of glottal mucosal wave after vocal fold microflap with or without fibrin glue. *J Voice* 2017;**31**:342–6
5 Morisaki T, Kishimoto Y, Tateya I, Kawai Y, Suzuki R, Tsuji T *et al.* Adipose-derived mesenchymal stromal cells prevented rat vocal fold scarring. *Laryngoscope* 2018;**128**:33–40
6 Padilla S, Orive G, Sanchez M, Anitua E, Hsu WK. Platelet-rich plasma in orthopaedic applications: evidence-based recommendations for treatment. *J Am Acad Orthop Surg* 2014;**22**:469–70
7 Stavrakas M, Karkos PD, Markou K, Grigoriadis N. Platelet-rich plasma in otolaryngology. *J Laryngol Otol* 2016;**130**:1098–102
8 Marques LF, Stessuk T, Camargo IC, Sabeh Junior N, dos Santos L, Ribeiro-Paes JT. Platelet-rich plasma (PRP): methodological aspects and clinical applications. *Platelets* 2015;**26**:101–13
9 Drago L, Bortolin M, Vassena C, Romano CL, Taschieri S, Del Fabbro M. Plasma components and platelet activation are essential for the antimicrobial properties of autologous platelet-rich plasma: an in vitro study. *PLoS One* 2014;**9**:e107813
10 Marx RE. Platelet-rich plasma: evidence to support its use. *J Oral Maxillofac Surg* 2004;**62**:489–96
11 Pomerantz J, Dutton JM. Platelet gel for endoscopic sinus surgery. *Ann Otol Rhinol Laryngol* 2005;**114**:699–704
12 Pavlovic V, Ciric M, Jovanovic V, Stojanovic P. Platelet rich plasma: a short overview of certain bioactive components. *Open Med* (*Wars*) 2016;**11**:242–7
13 Anitua E, Prado R, Padilla S, Orive G. Platelet-rich plasma therapy: another appealing technology for regenerative medicine? *Regen Med* 2016;**11**:355–7
14 Cobden SB, Ozturk K, Duman S, Esen H, Aktan TM, Avunduk MC *et al.* Treatment of acute vocal fold injury with platelet-rich plasma. *J Voice* 2016;**30**:731–5
15 Woo SH, Jeong HS, Kim JP, Koh EH, Lee SU, Jin SM *et al.* Favorable vocal fold wound healing induced by platelet-rich plasma injection. *Clin Exp Otorhinolaryngol* 2014;**7**:47–52
16 Mobarsa V, Samdani SK, Gurjar VS. Outcome analysis of microlaryngeal surgery for benign lesions of vocal cord using videostroboscopy and voice handicap index. *Indian J Otolaryngol Head Neck Surg* 2019;**71**:327–32
17 Shiotani A, Fukuda H, Kawaida M, Kanzaki J. Vocal fold vibration in simulated head voice phonation in excised canine larynges. *Eur Arch Otorhinolaryngol* 1996;**253**:356–63
18 Gray SD, Pignatari SS, Harding P. Morphologic ultrastructure of anchoring fibers in normal vocal fold basement membrane zone. *J Voice* 1994;**8**:48–52
19 Kim T, Kim S, Park J, No S, Jin S, Lee S, eds. Efficacy and safety of vocal cord platelet-rich plasma (PRP) injection in patient with intractable vocal cord disease. *Proceedings of the KSLP Conference*. Seoul: The Korean Society of Logopedics and Phoniatrics, 2017

https://doi.org/10.1017/S0022215121004564 Published online by Cambridge University Press