Majalah Kedokteran Gigi Indonesia
Vol 7 No 2 – August 2021
ISSN 2460-0164 (print), ISSN 2442-2576 (online)
Available online at https://jurnal.ugm.ac.id/mkgi
DOI: http://doi.org/10.22146/majkedgiind.53419

RESEARCH ARTICLE

Effect of advanced-platelet rich-fibrin combined with rosuvastatin application after open flap debridement of infrabony pocket

Tissa Rahadianti*, Dahlia Herawati, Kwartarini Murdiastuti****✉

*Periodontics Specialty Program, Faculty of Dentistry, Universitas Gadjah Mada, Yogyakarta, Indonesia
**Department of Periodontics, Faculty of Dentistry, Universitas Gadjah Mada, Yogyakarta, Indonesia
**Jl Denta No 1 Sekip Utara, Yogyakarta, Indonesia; ✉ correspondence: kmurdiastuti@ugm.ac.id

ABSTRACT

Open flap debridement (OFD) is an invasive therapy for chronic periodontitis with pocket 5 mm or more. However, it is difficult to achieve regeneration and new attachment with this therapy. Periodontitis starts to add growth factors and local drugs delivery as host modulation therapy. Advanced-PRF (A-PRF) contains more growth factor than PRF which plays a role in promoting fibroblast proliferation, reepithelization, extracellular matrix production, and endothelial cell migration. 1.2% rosuvastatin gel (RSV) is a local delivery drug with a pleiotropic effect that can modify host response to promoting BMSCs, BMP-2, OPG, ALP, RANKL, and osteoblasts. This study aimed to examine the effect of the application of A-PRF+RSV in OFD therapy of which the parameters were probing depth (PD), relative attachment loss (RAL), and alveolar bone height. The study samples consisted of 24 periodontal pockets which were divided into 2 groups of 12 pockets each, namely A-PRF+RSV for group 1 and PRF+RSV for group 2. Clinical evaluations were carried out on baseline, day-30, and day-90 for PD and RAL, and on baseline and day -90 for alveolar bone height. Data of PD and RAL reduction were analyzed with non-parametric test Mann-Withney, while data of reduction of alveolar bone height were analyzed with parametric Independent-T test. Group 1 obtained a statistically more significant result in reducing PD, RAL, and alveolar bone height compared to group 2 ($p<0.05$) To conclude, the application of A-PRF and 1.2% rosuvastatin gel in OFD procedure promotes a higher PD and RAL reduction and alveolar bone height increase than the application of PRF coupled with 1.2% rosuvastatin gel.

Keywords: advanced platelet rich-fibrin; chronic periodontitis; open flap debridement; periodontal pocket; rosuvastatin

INTRODUCTION

Periodontitis is the second most common dental and oral diseases in Indonesia with a prevalence of 60% of Indonesia's population.[1] The main etiology of periodontitis is plaque and calculus bacteria that infect the supporting tissues of the teeth like gingiva, periodontal ligament, cementum, and alveolar bone.[2] Based on the severity, periodontitis is divided into 3 phases: mild, moderate, and severe. These three phases are determined based on clinical examinations of probing depth, bleeding on probing, clinical attachment loss, and radiographic bone loss.[3] Treatment of periodontal disease based on severity are scaling root planning (SRP), curettage, or open flap debridement (OFD).[4]

Treatment of periodontitis has now shifted from simply eliminating the etiological factors and controlling the predisposing factors to obtaining new periodontal attachment.[4,5,6] The principle of a periodontal tissue regeneration treatment is a continuous complex process of cell adhesion, migration, proliferation, and differentiation.[7] Open flap debridement only eliminates the etiologic factors but could not restore the structure of periodontal tissues which are damaged and lost due to bacterial infections,[6,8] so research on a variety of growth factors and host modulation drugs has started to be conducted in OFD treatment to enhance tissue regeneration and periodontal treatment success.[5,9]

Previous research by Suwondo et al (2018) showed that Advanced-Platelet Rich Fibrin (A-PRF) was able to regenerate the periodontal tissues better than the conventional Platelet Rich Fibrin (PRF) based on parameters of probing depth (PD) and relative attachment loss. However, there was no difference in the increase in alveolar bone height because PRF only had an effect on the first initial stage, i.e., osteogenesis phase by optimizing osteoblast diffierentiation.[10]

Pradeep et al (2016) showed that OFD + PRF + 1.2% rosuvastatin (RSV) gel was able to decrease the value of clinical attachment level (CAL) and PD and significantly repair the infrabony defect better than OFD + PRF and OFD treatment.[4] The 1.2% RSV gel acts as host modulation therapy which aims to reduce tissue damage and regenerate the inflamed tissue by modifying the response of host factors.[4,11]

Rosuvastatin belongs to third generation statins that can reduce the level of fat or cholesterol in blood flow.[12] The effect of statin use in periodontal treatment has been reported both *in vitro* and *in vivo*. Statins control periodontal inflammation by inhibiting the production of pro-inflammatory factors such as cytokines and increasing the production of anti-inflammatory factors such as interleukin 10.[13] Statins are able to modulate the host response to prevent bone resorption due to inflammation and increase bone regeneration. In addition, statins are able to kill bacteria by destroying the bacterial membrane which can prevent future recurrence.[14,15] Petit et al (2019) concluded that a local statin therapy is recommended to be combined with other regenerative materials to boost the response of periodontal tissue healing and regeneration.[14]

MATERIALS AND METHODS

This research has been approved by the Ethics Committee of Faculty of Dentistry Universitas Gadjah Mada, Indonesia with registration number 00118/ KKEP/FKG UGM/EC/2019. An explanation of the procedure to be carried out in this research was given to the selected patients. The patients were asked to sign an informed consent form if they agreed to be the research subjects. Then, an initial phase therapy and the preparation of acrylic stent, A-PRF+ 1.2% RVS gel preparation, also PRF were done.

The 1.2% rosuvastatin gel was made from mixture of 10 ml of water and 0.2 grams of sodium carboxy-methyl cellulose which was pharmaceutical grade. The solution was stirred until gel consistency was obtained. A total of 120 mg rosuvastatin was continuously added and slowly stirred until homogeneous. The homogeneous rosuvastatin gel was then transferred into a 2 ml syringe applicator.[9]

The preparation of A-PRF started by taking blood intravenously from the median cubital vein using a 10 ml syringe then the blood was placed into a glass tube and centrifuged at a speed of 1,500 rpm for 14 minutes. The centrifugation promoted the formation of three layers, namely cellular plasma in the upper layer, A-PRF in the middle layer, and red blood cells in the bottom layer. The A-PRF was taken from the glass tube using a tweezer and separated from the red blood cells using scissors to obtain A-PRF clot only. Ten-millimeter A-PRF clot was cut into small pieces to be immediately applied for covering all the defect area.

The preparation of PRF was performed by taking 10 ml blood intravenously from the median cubital vein using a syringe then the blood was placed into a glass tube and centrifuged at a speed of 2,700 rpm for 12 minutes. The centrifugation resulted in the formation of three layers, namely cellular plasma in the upper layer, PRF in the middle layer, and red blood cells in the bottom layer. The PRF was taken from the glass tube using tweezers and separated from the red blood cells using scissors to obtain PRF clot only. Ten-millimeter PRF clot was then cut into small pieces to be immediately applied to cover all the defect area.

The baseline data were taken before the operation and O'Leary plaque index ≤ 10% was required after the initial phase therapy. PD and relative attachment loss (RAL) were measured before the dental treatment, followed by taking a Cone Beam Computed Tomography (CBCT) image. The alveolar bone height was measured on the basis of the distance between the cementoenamel junction (CEJ) to the alveolar bone defect, before and after treatment OFD.

The treatment started by an aseptic surgery procedure using extraoral iodine glycerin and intraoral use of 0.12% chlorhexidine gluconate. Local anesthesia using lidocaine HCL and epinephrine were delivered by an infiltration technique to the muco-buccal fold of the operating

Majalah Kedokteran Gigi Indonesia. August 2021; 7(2): 110 – 117
ISSN 2460-0164 (print)
ISSN 2442-2576 (online)

area. Interdental and lingual full-thickness incision was followed by a vertical and sulcular flap elevation. The granulation tissue and subgingival calculus were removed. The root surface of the tooth was smoothed. A total of 75 mg/ml tetracycline solution HCl was applied to the root surfaces of the tooth for 3 minutes, then rinsed with distilled water. After that, 0.1 ml of 1.2% rosuvastatin gel was added into the small cuts of PRF and A-PRF clot, then applied to the defect area. The flaps were returned, stitched, and covered with zinc oxide paste.

The patients were instructed to treat and clean the teeth postoperatively. Periodontal pack removal and debridement were performed on the 7th day after the surgery. Control on the 14th day was done to remove the stitches. Oral hygiene and healing control was performed once a week for 4-weeks post-surgery. Clinical parameters including PD and RAL were evaluated during the control on days 30 and 90 after the surgery. The CBCT procedure was repeated during the control on day 90.

RESULTS

The probing depth was measured at baseline, day 30, and day 90. The mean PD for each measurement time and treatment group is presented in Table 1. Mean PD in both groups decreased over time.

Table 2 shows data on the mean PD reduction between observation time. The data showed that the PD reduction in the A-PRF + RSV group was higher than that in the PRF + RSV group, both at baseline – 30 day or baseline – 90 day. However, the PD reduction between day 30 - day 90 in both groups was almost the same, namely 1.45 ± 0.52 mm in the A-PRF + RSV group and 1.45 ± 0.82 mm in the PRF + RSV group.

The difference of PD reduction between the groups was tested using the Mann Whitney U Test (Table 3). The data analysis showed a significant difference in the PD reduction on day 30 ($p = 0.029$) and on day 90 ($p = 0.033$) from baseline. Meanwhile, the reduction between day 90 and 30 was not significantly different ($p = 0.971$) between the groups. In conclusion, A-PRF + RSV reduced PD better than PRF + RSV with a significant difference as shown by the p value less than 0.05.

The RAL values at baseline, day 30, and day 90 are listed in Table 4. The mean RAL in both groups decreased on day 30 and day 90. The mean RAL reduction is listed in Table 5. The data showed that A-PRF + RSV resulted in better RAL reduction than PRF + RSV from baseline to day 30 and 90. The RAL reduction from day 30 to 90 was 1.54 ± 052 in the A-PRF + RSV group and 1.36 ± 0.67 in the PRF + RSV group. The difference of RAL reduction between the groups was tested using the Mann Whitney U Test because almost all the RAL reduction data were not normally distributed (Table 6).

The test results on the RAL reduction difference between the A-PRF + RSV and PRF + RSV groups showed a significant difference on day 30 ($p = 0.002$) and day 90 ($p = 0.026$) from baseline. The RAL reduction from 30 to 90 day was significantly different between the two groups ($p = 0.553$). Based on this test, the indicator of significant difference was $p < 0.05$. This showed that A-PRF + RSV reduced RAL better than PRF + RSV.

The alveolar bone height was measured from the distance between the cemento-enamel junction (CEJ) to the alveolar bone crest on CBCT examinations. The examinations were performed at baseline and day 90. The mean alveolar bone height is presented in Table 7.

The data in Table 7 showed a higher increase in alveolar bone height in the A-PRF + RSV group than the PRF + RSV group. The difference in the alveolar bone height reduction between the groups was tested using Independent T-test because the data had normal distribution. Based on the test results, the bone height reduction between the A-PRF + RSV and PRF + RSV groups was significantly different ($p = 0.000$). Thus, it can be concluded that A-PRF + RSV promoted better alveolar bone height increase than PRF + RSV because the p value was less than 0.05.

Table 1. Mean and standard deviation of probing depth (PD) according to observation time and treatment group (mm)

	Group					
	A-PRF + RSV			PRF + RSV		
	Mean	±	standard deviation	Mean	±	standard deviation
Baseline	6.00	±	0.89	5.82	±	0.75
day 30	2.91	±	0.83	3.36	±	1.03
day 90	1.45	±	0.52	1.91	±	0.54

Table 2. Mean and standard deviation of PD reduction by observation time and treatment group (mm)

	Group					
	A-PRF + RSV			PRF + SRV		
	Mean	±	standard deviation	Mean	±	standard deviation
Day 30 - Baseline	3.09	±	0.54	2.45	±	0.69
Day 90 - Baseline	4.55	±	0.52	3.91	±	0.70
Day 90 - day 30	1.45	±	0.52	1.45	±	0.82

Table 3. Test of difference in PD reduction between groups with Mann Whitney Test

	P	I			II		
		(30-bsl)	(90-30)	(90-bsl)	(30-bsl)	(90-30)	(90-bsl)
	Day 30 – bsl	–	.000*	.000*	.029*	.000*	.007*
I	Day 90 – 30		–	.000*	.003*	.971	.000*
	Day 90 – bsl			–	.000*	.000*	.033*
	Day 30 – bsl				–	.009*	.000*
II	Day 90 – 30					–	.000*
	Day 90 – bsl						–

*) : p<0.005
Bsl : Baseline
I : OFD + A-PRF + RSV
II : OFD + PRF + RSV

Table 4. Mean and Standard Deviation of Relative Attachment Loss (RAL) by observation time and treatment group (mm)

	Group					
	A-PRF + RSV			PRF + RSV		
	Mean	±	standard deviation	Mean	±	standard deviation
baseline	13.18	±	1.66	12.81	±	1.16
Day 30	10.09	±	1.51	10.72	±	1.27
Day 90	8.54	±	1.36	9.36	±	0.92

Majalah Kedokteran Gigi Indonesia. August 2021; 7(2): 110 – 117
ISSN 2460-0164 (print)
ISSN 2442-2576 (online)

Table 5. Mean and Standard Deviation of RAL reduction by observation time and treatment group (mm)

	Group					
	A-PRF + RSV			PRF + RSV		
	Mean	±	standard deviation	Mean	±	standard deviation
Day 30 - Baseline	3.09	±	0.53	2.09	±	0.70
Day 90 - Baseline	4.63	±	0.50	3.45	±	0.82
Day 90 - day 30	1.54	±	0.52	1.36	±	0.67

Table 6. Test of differences in RAL reduction between groups with Mann Whitney U Test

	P	I			II		
		(30-bsl)	(90-30)	(90-bsl)	(30-bsl)	(90-30)	(90-bsl)
	Day 30 - bsl	–	.000*	.000*	.002*	.000*	.048*
I	Day 90 - 30		–	.000*	.062	.553	.000*
	Day 90 - bsl			–	.000*	.000*	.026*
	Day 30 - bsl				–	.029*	.000*
II	Day 90 - 30					–	.000*
	Day 90 - bsl						–

*) : p<0,005
Bsl : baseline
I : OFD + A-PRF + RSV
II : OFD + PRF + RSV

Table 7. Mean and Standard Deviation of high alveolar bone and alveolar bone height reduction according to observation time and treatment group (mm)

		Group					
		A-PRF + RSV			PRF + SRV		
		Mean	±	standard deviation	Mean	±	standard deviation
CEJ – alveolar crest	Baseline	5.68	±	1.98	4.89	±	2.28
	day 90	4.58	±	1.98	4.30	±	2.26
The increase in alveolar bone height		1.10	±	0.19	0.58	±	0.18

Table 8. Test of differences in alveolar bone height reduction between groups with Independent T- test

Reductions	P	Result
Day 90 – baseline	0.000	Significant

DISCUSSION

This study showed that the application of A-PRF + RSV after OFD treatment reduced PD and RAL as well as increased alveolar bone height better than the application of PRF + RSV. The reduction of RAL and PD on day 30 was found on both the A-PRF + RSV and PRF + RSV groups. However, the reduction was significantly higher in the A-PRF + RSV group. A significant difference between these two was due to their growth factor

contents. Ghanaati et al. (2014) stated that A-PRF has a higher number of growth factors than PRF, including PDFG, IGF-1, VEGF, EGF and TGF-β.[16,17] A literature review by Park et al (2017) described some of the functions and roles of growth factors produced by A-PRF and PRF, for examples PDFG, IGF-1, and VEGF can increase fibroblast proliferation, extracellular matrix production, re-epithelialization process, angiogenesis, and endothelial cell migration. Meanwhile, EGF and TGF-β help stimulate keratinocyte proliferation and increase granulation tissue.[18] The different number of growth factors between A-PRF and PRF is enough to make significant differences in the value of PD and RAL according to Suwondo et al (2018) who compared OFD + PRF and OFD + A-PRF.[10]

1.2% rosuvastatin gel was added in both groups in this study to maximize healing process and regeneration of soft and hard periodontal tissues. The pleiotropic effects of 1.2% rosuvastatin gel, such as antioxidants, anti-inflammation, osseo-modulation, and immunomodulation play role in the healing process of periodontal tissues by modifying host regeneration.[19] Anti-inflammation and antioxidant effects play an important role in the inflammatory and proliferative phase by inhibiting pro-inflammatory proteins such as IL-1β and reactive oxygen species (ROS) that can make tissue damage and prolong wound healing.[13,20]

The reduction of PD and RAL does not indicate improvement of clinical conditions of periodontal tissue.[21] New attachment occurs when there is a new bond between periodontal ligament fibers into new cementum, while what usually takes place is long junctional epithelium. Long junctional epithelium occurs when gingival epithelium is attached to the tooth surface.[21,22] Unlike previously, the value of PD and RAL reduction on day 30 - day 90 was not significantly different. This condition is because the healing process on the 30th day has entered the maturation and remodeling phase[18,20] Healing process begins 2 or 3 weeks after injury and will continue for one year or more through the reorganization, degradation, and re-synthesis of extracellular matrix.[20]

Alveolar bone height increased on day 90 in both groups, but the A-PRF + RSV group showed a significantly higher increase in the alveolar bone height compared to the PRF + RSV group. This result indicated that 1.2% rosuvastatin gel application might have a role in the alveolar bone height differences between the two groups because research by Suwondo et al (2018) showed that there was no difference in the alveolar bone height between the A-PRF and PRF application on OFD procedures.[10]

1.2% rosuvastatin gel is a lipid mediator that acts as Host Modulation Therapy (HMT).[11] This gel can reduce periodontal tissue damage and promote tissue regeneration by modifying response factor of host.[23] Petit et al (2019) stated in their research report that 1.2% rosuvastatin gel can increase the number of vascular endothelial growth factor (VEGF) and bone morphogenetic protein-2 (BMP-2). Both of these proteins then regulate osteoblast differentiation, increasing new bone formation during regeneration and wound healing.[14] Millan et al (2019) mentioned that local application of 1.2% rosuvastatin gel improved some important proteins that play a role in bone formation, such as bone marrow-derived mesenchymal stem cells (BMSCs), BMP-2, expression of osteoprotegerin (OPG), alkaline, phosphatase enzyme (ALP), receptor activator for nuclear factor K B ligand (RANKL), and osteoblats.[19] 1.2% rosuvastatin gel also increases IL-10 which affects regeneration process by increasing osteoblasts and inhibiting osteoclasts.[24]

The different numbers of the growth factor contents in A-PRF and PRF also contributed to the significantly different mean alveolar bone height between the two groups. Both A-PRF and PRF contain TGF, IGF, and VEGF, but A-PRF contains more growth factors than PRF.[17,25] A study by Sumida et al (2019) showed that A-PRF and PRF increased the expression of (OPG) and RANKL.[26] Besides, Toit et al (2015) stated that there was a disadvantage of PRF compared to A-PRF, namely the absence of Bone Morphogenetic Protein-2 (BMP-2) content in PRF. Meanwhile, A-PRF contains a small amount of BMP-2.[27] BMP-

2 is a compound that can increase osteoblast differentiation in bone formation.[27] THe osteogenic effect of BMP-2 would be more effective if there is a reaction with VEGF.[26]

CONCLUSION

The application A-PRF and 1.2% rosuvastatin gel in OFD procedure promotes higher PD, RAL reduction, and alveolar bone height increase than the application of PRF coupled with 1.2% rosuvastatin gel.

REFERENCES

1. Fadhilah ND, Ulfiana R, Putri FA. Peran periodontitis terhadap patogenesis penyakit alzheimer. BIMKGI. 2016; 4(1): 33-36.
2. Ausenda F, Rasperini G, Acunzo R, Gorbunkova A, Pagni G. New perspectives in the use of biomaterials for periodontal regeneration. Materials (Basel). 2019; 12(13): 2197-2217. doi: 10.3390/ma12132197
3. Geurs N, Lacono V, Krayer J, Mealey B, Paquette D, Pearson B, Rosen P, Schweinebraten MS. American Academy of Periodontology task force report on the update to 1999 classification of periodontal disease and conditions. J Periodontal. 2015; 86(7): 835-838. doi: 10.1902/jop.2015.157001
4. Pradeep AR, Garg V, Kanoriya D, Singhal S. Platelet-rich fibrin with 1.2% rosuvastatin for the treatment of intrabony defects in chronic periodontitis: a randomized controlled clinical trial. J Periodontol. 2016; 87(12): 1468-1473. doi: 10.1902/jop.2016.160015
5. Borie E, Olivi DG, Orsi LA, Garlet K, Weber B, Beltran V, Fuentes R. Platelet-Rich fibrin application in dentistry: a literature review. Int J Clin Exp Med. 2015; 8(5): 7922-7929.
6. Rathod S, Wanikar I, Paralikar A. Soft and hard tissue changes after the placement of smart blood platelet-rich fibrin derivative into intrabony defects: a systematic review and meta-analysis with at least. J Int Clin Dent Res Organ. 2018; 10(2): 49-58. doi: 10.4103/jicdro.jicdro_10_18
7. Chandran P, Sivadas A. Platelet-rich fibrin: its role in periodontal regeneration. The Saudi Journal for Dental Research. 2014; 5(2): 117-122. doi: 10.1016/j.ksujds.2013.09.001
8. Reynolds MA, Kao RT, Nares S, Camargo PM, Caton JG, Clem DS, Fiorellini JP, Geisinger ML, Mills MP, Nevins ML, Rosen PS. Periodontal Regeneration - intrabony defects: practical applications workshop from the aap regeneration. Clinical Advances in Periodontics. 2015; 5(1): 21-29. doi: 10.1902/cap.2015.140062
9. Agrawal M, Agrawal V. Platelet rich fibrin and its applications in dentistry- a review article. Natl J Med Dent Res. 2014; 2(3): 51-58.
10. Suwondo CI, Herawati D, Sudibyo. Effect of platelet-rich fibrin advanced applications on periodontal regeneration in infrabony pocket treatments. Majalah Kedokteran Gigi Indonesia. 2018; 4(3): 155-159. doi: 10.22146/majkedgiind.37414
11. Sulijaya BN, Takahashi, Yamazaki K. Host modulation therapy using anti-inflammatory and antioxidant agents in periodontitis: a review to a clinical translation. Arch Oral Biology. 2019; 105: 72-80. doi: 10.1016/j.archoralbio.2019.07.002
12. Elavarasu S, Suthanthiran TK, Naveen D. Statins: A New era in local drug delivery. J Pharm Bioallied Sci. 2012; 4(2): 248-251. doi: 10.4103/0975-7406.100225
13. Kirzioglu FY, Bulut MT, Dogan B, Fentoglu O, Ozmen O, Carsancak SA, Ergun Ag, Ozdem M, Orhan H. Anti-inflammatory effect of rosuvastatin decreases alveolar bone loss in experimental periodontitis. J Oral Sci. 2017; 59(2): 247-255. doi: 10.2334/josnusd.16-0398
14. Petit C, Batool F, Bugueno IM, Schwinte P, Jessel NB, Huck O. Contribution of statin towards periodontal treatment: a review. Mediators Inflamm. 2019; 2019: 6367402. doi: 10.1155/2019/6367402
15. Preshaw PM. Host modulation therapy with anti-inlammatory agents. Singapore: Periodontology 2000; 2017. 1-19.

16. Ghanaati S, Booms P, Orlowska A, Kubesch A, Lorenz J, Rutkowski J, Landes C, Sader R, Kirkpatrick CJ, Choukroun J. Advanced platelet-rich fibrin: a new concept for cell-based tissue engineering by means of inflammatory cells. J Oral Implantol. 2014; 40(6): 679-689. doi: 10.1563/aaid-joi-D-14-00138
17. Kobayashi E, Fluckiger L, Kobayashi MF, Sawada K, Sculean A, Schaller B, Miron RJ. Comparative releas of Growth Factors from PRP, and Advanced-PRF PRF. Clin Oral Investig. 2016; 20(9): 2353 -2360. doi: 10.1007/s00784-016-1719-1
18. Park JW, Hwang, Yoon SRIS. Advanced Growth Factor Delivery Systems in Wound Management and Skin Regeneration. Molecules. 2017; 22(8): 1259-1279. doi: 10.3390/molecules22081259
19. Millan ER, Navarro BG, Gomez KI, Roig AM, Salas EJ, Lopez JL, Ortega EV. The application of statins in the regeneration of bone defects. Systematic review and meta-analyis. Materials. 2019; 12(18): 2992. doi: 10.3390/ma12182992
20. Gonzalez ACO, Andrade ZA, Costa TF, Medrado ARAP. Wound Healing - a literature review. An Bras Dermatol. 2016; 91(5): 614-620. doi: 10.1590/abd1806-4841.20164741
21. Borges CDA, Ricoldi MS, Messora MR, Palioto DB, Souza SLS. Clinical attachment loss and molecular profile in inflamed sites before treatment. J Appl Oral Sci. 2019; 27: 1-10. doi: 10.1590/1678-7757-2018-0671
22. Newman MG, Takei HH, Klokkevold PR, Carranza, FA. Carranza's Clinical Periodontology, 12th Edition. Missouri: Elsevier Saunders; 2015. 50-51.
23. Shinwari MS, Tanwir F, Hyder PR, Saeed HMB. Host modulation therapeutics in Periodontics: Role as an adjunctive periodontal therapy. J Coll Physicians Surg Pak. 2014; 24(9): 676-684.
24. Zhang Q, Chen B, Guo J, Zhu X, Ma S, Yang W. Interleukin-10 inhibits bone resorption: a potential therapeutic strategy in periodontitis loss and other bone diseases. Biomed Research International. 2014; 2014: 284836. doi: 10.1155/2014/284836
25. Caymaz MG, Uyanik LO. Comparison of the effects of advanced platelet rich fibrin and leukocyte- and platelet-rich fibrin on outcome after removal of impacted mandibular third molar: A randomized split-mouth study. Niger J Clin Pract. 2019; 22(4): 546-552. doi: 10.4103/njcp.njcp_473_18
26. Sumida R, Maeda T, Kawahara I, Yusa Y. Platelet-rich fibrin increases the osteoprogentaor/receptor activator of nuclear factor-kB ligand ration in osteoblasts. Exp Ther Med. 2019; 18(1): 358-365. doi: 10.3892/etm.2019.7560
27. Toit JD, Gluckman H, Salama M. Platelet-Rich fibrin (PRF): a growth factor-rich biomaterials. part 1 - the platelet concentrates milieu and review of the literature. International Dentistry - African Edition. 2015; 5(5): 62-70.