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ORIGINAL RESEARCH

Clinical and Cytotoxic Comparison of Two Periodontal Dressings after Periodontal Flap Surgery

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ABSTRACT

Aim: Postoperative coverage of periodontal surgery sites can help protect the treated area, facilitate wound healing and decrease postperiodontal surgery pain. The aim of this study was to compare the cytotoxic and clinical efficacy of two periodontal dressings after periodontal flap surgery.

Materials and methods: In this study 23 patients requiring modified Widman flap in at least two quadrants in the same arch were selected; one quadrant was dressed with Reso-pac, and the other was dressed with Coe-pak. The clinical efficacy of these two dressings was evaluated by comparing plaque, granulation tissue formation, pain, bleeding on probing, and color of gingiva. To compare their cytotoxicity, human gingival fibroblast were exposed to 1- and 3-day extracts of the dressings and MTT test was used to measure cell viability after 24 and 48 hours. Cell apoptosis and necrosis were evaluated by flow cytometric analysis. Data were analyzed by Chi-square and independent t-test and SPSS 20 Software.

Results: Plaque and granulation tissue formation rates were significantly lower in Reso-pac covered sites compared to coe-pak (p value < 0.001). Other variables including pain, bleeding and gingival color did not show any significant differences (p value 0.05). Viable fibroblast cells were higher for Reso-pac compared to Coe-pak (p < 0.05). A higher percentage of necrotic cells in the day one Coe-pak extract group after 24 and 48 hours were observed compared to Reso-pac (6.23 and 4.97 *vs.* 2.71 and 2.76%).

Conclusion: According to our results, Reso-pac is as effective as Coe-pack. It also has further positive effects of less plaque accumulation and granulation tissue formation and is more biocompatible for HGF cells with less cytotoxic effects on cells in the first days after surgery.

Clinical significance: Reso-pac may be considered as a dressing of choice in periodontal surgeries with less plaque accumulation and granulation tissue formation plus better biocompatibility and ease of application compared to Coe-pak.

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INTRODUCTION

Periodontal disease is one of the most common diseases of the oral cavity, with inflammatory properties which cause destruction of tooth-supporting structures, including alveolar bone and periodontal ligament, and results in crestal bone resorption.[1] Periodontal surgery is a common oral surgical method which is used to access the root surface for removing all local predisposing factors.[2] Postoperatively the surgical site can be covered by periodontal dressings. These materials usually have no therapeutic properties but they can decrease infection and bleed to a minimum level. In addition, they can help reduce postoperative pain by avoiding food and tongue irritations to the surgical wound and be used as a template in the wound healing process to prevent granulation tissue formation.[3,4] Most importantly; they can prevent damage of tissue and exposed bone, leading to less pain during wound healing.[5]

Periodontal dressings were used for the first time in 1923 by Ward in order to protect surgical sites from mechanical trauma and splint soft tissue and mobile teeth.[6] From 1923 until today many different periodontal dressings have been produced and many studies regarding their properties have been conducted. However, controversies still remain about the need for their use and the most appropriate dressing.

Coe-pak (GC, USA) is one of the most widely used types of dressings. It is based on a metallic oxide and fatty acid reaction; however, it has some disadvantages including inappropriate setting time and weakness of appearance and poor flowability. Besides that, Coe-pak causes bacteria and plaque accumulation at the site of surgery, which can delay post-surgical wound healing.[7] Reso-pac (Hager and Werken GmbH and Co. KG) is a soft, soluble and hydrophilic dressing with the adhesive capability to oral tissues that helps in easy coverage and protection of the

wound. In addition to cellulose which is its main structure, Reso-pac contains myrrh which has disinfective, adhesive and hemostatic properties. This dressing material results in fibrin formation on the wound. It has a pleasant taste and elastic properties which relief the wound from too much tension which might be seen when Coe-pak is used. Reso-pac will gradually be dissolved in the oral cavity, so it is not necessary to remove it and thereby chlorhexidine can reach the surgical site. No allergic reactions have been reported until now.[8]

The use of periodontal dressings is increasing in routine practice since there is still no agreement regarding their positive effects on wound healing after surgery.[6] Greater patient comfort with dressing usage seems to be an important factor in favor of their application. In a study by Soheilifar et al., there was no significant difference between quadrants covered with a dressing and without dressing in terms of inflammation, bleeding, gingival consistency, and granulation tissue formation, the color of gingiva and patient's convenience. However, mean pain perception among patients was significantly reduced in quadrants with dressing.[9] The search for the best periodontal dressing has been going on, and many different studies have been designed and performed to compare various periodontal materials.[10-12]

Some newly designed dressings have been compared with Coe-pak in order to find the best dressing for clinical application such as a new collagen-based dressing (colla cote) which was found to have a significant difference on palatal wound healing in terms of collagen formation and reconstruction compare to traditional non-eugenol dressing (Coe-pak).[11] Reso-pac seems to have promising characteristics for clinical application, however, up to our knowledge it has never been clinically investigated previously.

In Petelin et al.'s study, Reso-pac and Barricaid were recognized as the most suitable dressings considering cell cytotoxicity.[13] Kadkhodazadeh et al. studied Reso-pac cytotoxicity in comparison to Coe-pak and showed slightly less cytotoxicity of Reso-pac than Coe-pak.[8]

Since Reso-pac seems to be a dressing with favorable characteristics, we decided to compare its clinical efficacy and evaluate its cytotoxic characteristics on gingival fibroblasts with the more popular Coe-pack dressing.

MATERIALS AND METHODS

This study was a double-blind randomized clinical and *in vitro* study. We evaluated the clinical effects of two types of periodontal dressings on wound healing after sugary. It was approved by the university ethical committee and conducted in accordance with the Helsinki Declaration. It has also been registered in the Iranian Registry of clinical trials (IRCT ID IRCT2016050718493N2). All patients signed informed consent forms for participating in the study, scaling and root planning was performed on selected patients, then oral hygiene was instructed to them.

Patients were revaluated after two weeks and 23 patients (male: 8, female: 15) with at least two sites of PPD more than and equal to 5 mm on each posterior sextant of a jaw, needing further treatment with modified Widman flap (MWF) surgery in the reevaluation session were enrolled in the study.

Inclusion Criteria

Chronic periodontitis patients needing two similar (MWF) surgeries in two quadrants of a jaw, without intrabony or angular bone loss.

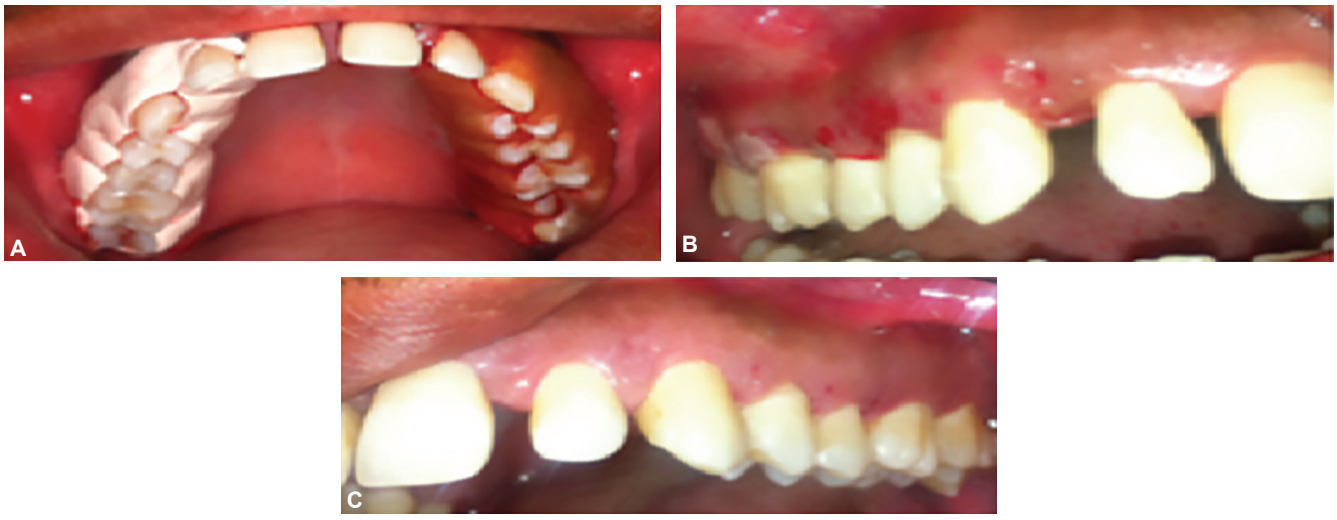
Exclusion Criteria

Use of antibiotics, corticosteroids, and hormonal drugs in the last two months, diabetic patients, and a history of periodontal surgery, surgical treatment of both quadrants was done by an experienced periodontist in a single visit. By simple randomization technique, they were divided into two groups: A and B. The two different dressing was applied to the treated sites on each side of the jaw. After the periodontal flap surgery in group A, Coe-pac was used on the right treated quadrant and in the left side, Reso-pac was used; the application of packs in group B was reverse. Periodontal dressings were prepared based on the manufacturer's instructions.

Postoperatively, 0.2% chlorhexidine (CHX) mouthrinse was prescribed twice a day for two weeks and amoxicillin 500 mg three times a day (for a week) and ibuprofen 400 mg the first two days, was also prescribed to all patients, but if patients needed further use of analgesics they had to let the surgeon know and were excluded from the study. Patients were not informed on the benefits and disadvantages of different periodontal dressings to avoid patient psychological effect, and a blinded periodontist evaluated the outcomes in each quadrant.

On days 7 and 14 after each surgery, the patient were reevaluated clinically (in terms of gingival color, bleeding on probing, granulation tissue formation, and plaque index). The gingival color was determined visually with a comparison of gingival color in healthy sites and the surgically treated region.

Loe and Silness plaque index (0–3) was recorded. Based on this index, code 0 represented no plaque; code 1 thin plaque on the free gingival and adjacent tooth; code 2 moderate plaque deposition; code 3 severe plaque accumulation on marginal gingiva and tooth surface (Fig. 1). To measure bleeding rate, the probe was gently inserted for 1 mm and moved around the tooth. Barnett index has been utilized (from 0 to 3), no bleeding (code 0). Bleeding after



Figs 1A to C: (A) Placement of Coe-pak on the right side sextant and Reso-pac on the left side after periodontal flap surgery; (B) 7 days after surgery and removal of sutures and Coe-pak, plaque formation under the dressing is clearly visible; (C) 7 days after surgery and removal of sutures in the Reso-pac covered side, less bleeding on suture removal and a clean site is observed due to the earlier dissolution of the dressing in the mouth

30 seconds (code 1), immediate bleeding (code 2), spontaneous bleeding (code 3). Granulation tissue formation was assessed visually by examiners. Patients were asked about the history of pain sensation during last week using a visual analog scale (VAS). 0 represents no pain and 10 represents extreme pain sensation.

Cytotoxic Effect Evaluation

Human gingival fibroblasts were obtained from the National cell bank of Iran (Pasteur, Tehran, Iran). The cells were grown in αMEM (Invitrogen LT, Merelbeke, Belgium) and 10% fetal bovine Serum (FBS, Invitrogen LT, Merelbeke, Belgium) and 1% antibiotic–antimitotic (Gibco, Germany) in a 5% CO_2 and 37°C atmosphere. They were passaged after reaching confluency.

Specimen Preparation

Equally sized disks of 5 mm in diameter and 2 mm in thickness were prepared from both periodontal dressings. Coe-pak was allowed to set for half an hour at 37°C and 100% humidity. Then they were placed in 5 mL of MEM to produce extracts. One and three-day extracts were obtained for Reso-pac, Coe-pak. This was decided due to the fact that Reso-pac usually dissolves very quickly and no longer exists in the mouth after three days.

Cytotoxicity Assay with Extracts

5×10^3 cells were seeded into 96 well plates and grown for 24 hours. Then the extracts were added to each well. MTT assay was used to evaluate cell viability. After 24 and 48 hours of contact with dressing using 10% MTT.

At this time Formosan crystals can be seen under an inverted microscope. The supernatant was removed and 100 mL of dimethyl sulfide was pipeted to dissolve the crystals. Then a microplate reader 540–690 nm (Biotech ELx808) was used to evaluate vital cells. The results were reported as percentages.

In addition, flowcytometric analysis was used for evaluating cell apoptosis and necrosis. Propidium iodine (PI) and V annexin were used for this purpose. The annexin V/PI protocol is a commonly used approach for studying apoptotic cells. After cells were cultured with the extracts from the dressing for 24 and 48 hours, they were trypsinated and then centrifuged at 1200 g. This was repeated once again. With 15 mL of phosphate buffer silane. Then 1 mL of the buffer provided in the special kit (eBioscience Cat. No: 88-8005-72) was added and after pipetage 15 m of V annexin was added and incubated in darkness for 15 minutes. Fourteen mL of propidium iodine was added and analyzed with a flowcytometer and necrotic and apoptotic cells were reported as percentages.

Data were analyzed by SPSS version 20 software and Chi-square and independent t-test

RESULTS

Clinical Evaluation

- *Pain:* In the present study the severity of pain in a patient using Reso-pac and Coe-pak was 0.7 and 1.17, respectively. There was no significant statistical difference among patients that use two types of dressing ($p = 0.293$) (Table 1).
- *Gingival color:* 7 and 14 days after surgery color of gingival was the same among patients with Coe-pack and Reso-pack. According to the table, there is no significant difference in gingival color between the

Table 1: Comparison of mean pain scores during the first postoperative week using visual Analysis Scale

Type of dressing materials	*Mean ± SD*	*p-value*
Reso- Pac	0.7 ± 1.36	0.293
Coe-pak	1.17 ± 1.67	

two groups ($p = 0.113$, $p = 0.73$) (Table 2).

- *Bleeding on Probing:* Statistical difference was not significant in term of bleeding status among two groups (Coe-pak vs. Reso-pac) 7 and 14 days after surgery ($p = 0.215$, $p = 0.3$) (Table 3).
- *Granulation tissue:* In terms of the presence of granulation tissue, the statistical difference was significant between the two groups ($p < 0.001$). In patients receiving Reso-pac, granulation tissue formation was less than Coe-pak group. In this study fourteen days after surgery in Reso-pack received sites, no granulation has been formed in 100% of sites but in 56.5% of Coe-pak dressed sites, granulation tissue was visible. This was a statistically significant difference between the two groups (Table 4).
- *Plaque accumulation:* In this study, seven days after surgery, the statistical difference among patients receiving either Reso-pac or Coe-pak was significant about plaque formation ratio ($p < 0.001$). This means in Reso-pac recipient sites, plaque accumulation was low. Also, based on the table, no significant statistical difference has been shown among Reso-pac and Coe-pak recipient patients after 14 days, in term of plaque formation ($p = 0.76$) (Table 5).

Cytotoxicity Evaluation

Comparison of the one and three-day extracts' effect on HGF cells, after 24 and 48 hours of exposure, revealed that HGF cell viability was significantly higher in the Reso-pac group compared to Coe-pak ($p < 0.05$). (Table 6)

Day 1 extract of co-pak showed less viable cells compared to the control but in Reso-pac the percent of viable cells increased by time.

However the day 3 extract of both dressings showed less cytotoxic effects compared to the day1 groups and were not cytotoxic compared to the control group.

According to the flowcytometric evaluation, results the percentage of necrotic cells were higher in the

Table 2: Color of gingiva in the two groups 7 and 14 days after surgery

	Color of gingival/ Type of periodontal dressing	*Pale pink*	*Dark pink*	*Red*	*Total*	
		Count (%)	*Count (%)*	*Count (%)*	*Count (%)*	*p value*
7 days	Rose-pac	5 (21.7)	17 (73.9)	1 (4.3)	23 (100)	0.113
	Coe-pak	3 (13)	14 (60.9)	6 (26.1)	23 (100)	
14 days	Rose-pac	18 (78.3)	5 (21.7)	0 (0)	23 (100)	0.73
	Coe-pak	17 (73.9)	6 (26.1)	0 (0)	23 (100)	

Table 3: Gingival bleeding values 7 and 14 days after surgery (Coe-pak and Reso-pac groups)

	Gingival bleeding/ Type of periodontal dressing	*No*	*Mild*	*Moderate*	*Severe*	
		Count (%)	*Count (%)*	*Count (%)*	*Count (%)*	*p value*
7 days	Rose-pac	2 (8.7)	18 (78.3)	3 (13)	0 (0)	0.215
	Coe-pak	2 (8.7)	12 (52.2)	8 (34.8)	1 (4.3)	
14 days	Rose-pac	19 (82.6)	4 (17.4)	0 (0)	0 (0)	0.3
	Coe-pak	16 (69.6)	7 (30.4)	0 (0)	0 (0)	

Table 4: Granulation tissue formation values 7 and 14 days after surgery (Coe-pak and Reso-pac groups)

	Granulation tissue formation/ Type of dressing materials	+		–		
		Count (%)	*Count (%)*	*Count (%)*	*Count (%)*	*p value*
7 days	Rose-pac	2	8.7	21	91.3	<0.001
	Coe-pak	14	60.9	9	31.9	
14 days	Rose-pac	0	0	23	100	<0.001
	Coe-pak	13	56.5	10	43.5	

Table 5: Plaque formation values 7 and 14 days after surgery (Coe-pak and Reso-pac groups)

	Plaque/ Type of dressing materials	*No*		*Mile*		*Moderate*		*Severe*		
		Count	*(%)*	*Count*	*(%)*	*Count*	*(%)*	*Count*	*(%)*	*p value*
7 days	Rose-pac	12	52.2	11	47.8	0	0	0	0	<0.001
	Coe-pak	0	0	1	4.3	15	65.2	7	30.4	
14 days	Rose-pac	12	52.2	11	47.8	0	0	0	0	0.76
	Coe-pak	11	47.8	12	52.2	0	0	0	0	

Table 6: Percentage of viable gingival fibroblast cells after exposure to 1- and 3-day extracts of Coe-pak and Reso pac for 24 and 48 hours

		Coe-pak			*Reso-pac*				
Time	*Control*	*24 hours*	*48 hours*	*p value*	*24 hours*	*48 hours*	*p value*	*p value*	*p value*
Day 1	100 ± 6.23	66.49 ± 7.23	74.03 ± 8.33	0.012	84.95 ± 8.93	97.97 ± 9.13	0.041	0.017	0.000
Day 3	100 ± 7.26	121.75 ± 1.35	97.63 ± 10.35	0.032	138.5 ± 1.41	109.5 ± 9.32	0.022	0.000	0.000

Table 7: Apoptotic and necrotic fibroblast cells after exposure to 24 and 48 hours extracts of Coe-pak and Reso pac periodontal dressings

Exposure times	*Extracts*	*Vital cells (%)*	*Necrosis (%)*	*Apoptosis (%)*	*Late apoptosis (%)*
Control		97.02	2.85	0.08	0.05
Coe-pak 24 hours	Day 1 extract	92.73	6.23	0.03	0.74
	Day 3 extract	94.93	4.97	0.07	0.04
Reso-pac 24 hours	Day 1 extract	94.90	2.71	1.87	1.33
	Day 3 extract	95.91	2.76	0.98	0.36
Coe-pak 48 hours	Day 1 extract	98.83	0.92	0.16	0.06
	Day 3 extract	99.23	0.71	0.00	0.05
Reso-pac 48 hours	Day 1 extract	99.14	0.47	0.35	0.04
	Day 3 extract	99.90	0.10	0.00	0.00

Coe-pak groups especially in the first 24 hours of exposure in both 1- and 3-day extracts (Table 7)

DISCUSSION

Reso-pac has been preferred because of its plasticity and being ready for use without the need for mixing. Coe-pack is also a widely used conventional periodontal dressing. Since there has been no clinical study comparing these two dressings, we conducted this randomized clinical study and *in vitro* cytotoxic comparison of these two dressings in order to find the most effective and suitable dressing for clinical applications. According to statistical analysis, the results among the two studies groups showed no significant difference in terms of pain, the color of gingiva and bleeding; however, an interesting statistically significant difference was observed regarding the amount of plaque accumulation under the dressings and amount of granulation tissue formation. The cytotoxic effects were also in favor of Reso-pac which showed higher cell viability results that were statistically significant.

The severity of pain was analyzed by VAS analysis within seven days after surgery. There was no statistically significant difference between the two groups ($p < 0.05$) which was similar to the results of studies by Bae,[12] Cheshire,[14] Abed et al.[15] and Sanz et al.[16]

In our study, the periodontal dressing has been placed on both sides and existence of Reso-pack within the first few days after surgery seems to be helpful and enough for patients comfort and to relief pain. In Soheilifar et al.,[9] split-month study comparing two conditions (with/without dressing), also in a similar study by Ghanbari et al.,[10] the degree of pain in the presence of dressing was statistically lower than its absence.

On the other hand, in some studies (such as Shanmugam,[11] and Jorkend's,[17] studies pain was higher in the case of usage of Coe-pack which can be due to the fact that sometimes the mechanical pressure from the coe-pak with becomes very hard after setting may cause difficulty and even a painfulness experience for patients. Furthermore, sometimes this pressure of the dressing after setting can lead to discomfort and damage to surgical sites.[7] However, this characteristic of Coe-pak can be helpful in apically positioned flap procedures to help in replacing the tissue. Overall and with the findings of our study it seems that generally, the presence of dressing in the first few days after surgery can be helpful in relieving of pain.

The degree of bleeding during usage of the periodontal probe in this study on days 7, and 14 after surgery did not show any statistically significant difference ($p > 0.05$)

The color of gingiva also showed no significant difference between the groups, however, red color and severe bleeding was only seen at Coe-pak receiving site. Our results were in accordance with other studies in this respect. Either study comparing with/without the use of Coe-pak dressing,[9-11] or studies which compared Coe-pack with new dressings such as RDs new experimental dressing,[14] or collocate.[11]

In the current study plaque accumulation on Coe-pak sites was more than Reso-pac sites ($p < 0.05$). The reason is that Reso-pac dissolves spontaneously after a few days: this was similar to findings of previous studies.[7,11,16]

However, in Ghanbari's study no statistically significant difference was found about plaque index with/without pack sites;[10] other studies have not compared this index;[9-15] debris and food accumulation under dressings can cause bad breath and delay healing, but in the Reso-pack group, after it is dissolved less plaque is accumulated and also chlorhexidine accessibility to surgery sites may decrease malodor and accelerate wound healing which is in accordance with the results of the current study which

also showed that granulation tissue formation was less in the Reso-pac group and better and faster healing was observed.

In our study, the effect of 1, 3-day extract of the dressings (Reso-pac and Coe-pak) were evaluated on HGF cell culture medium after 24 and 48 hours exposure. At all evaluation times, Reso-pac had fewer cytotoxicity effects than Coe-pak. The results of this study were in accordance with Kadkhodazadeh's study,[8] that showed a difference in cytotoxicity of Reso-pac and Coe-pak groups in their day one extracts. However, in their results, Reso-pac had no-time. Dependent cytotoxicity but cytotoxicity of Coe-pak increased by time and their 3 and 7-day extracts were very cytotoxic compared to the Reso-pak group. Our results, however, showed the most toxic effect of Coe-pak in the day 1 extract which could be because most toxic materials release from the dressing in the first few hours after setting. This is also in accordance with the clinical observations after application of Coe-pack dressing as no significant cytotoxic effect is seen in clinical use. However, Reso-pak seems to have the most favorable results and more biocompatible material. This was also observed in our flow cytometric evaluation as a higher percentage of necrotic cells was observed in the Coe-pak group after exposure of HGF to its day one extract. Our results were also in accordance with Petelin's study,[13] that showed Reso-pac had only small inhibitory effects on fibroblast cell proliferation and found to be the most suitable dressing in comparison to Peri-pac, Barricaid and Fittydent, also same as other articles showed MTT assay was a reliable technique.[18-20]

CONCLUSION

According to the results of the current study, Reso-pac is as effective as Coe-pack and can even be more effective by decreasing plaque and granulation tissue formation postoperatively. It is also more biocompatible for HGF cells than Coe-pak having less cytotoxic effects on cells in the first days after surgery.

CLINICAL SIGNIFICANCE

Reso-pac seems to be considered as a dressing of choice in periodontal surgeries with less plaque accumulation and granulation tissue formation, also better biocompatibility and ease of application compared to Coe-pak.

REFERENCES

1. Heidari Z, Mahmoudzadeh-Sagheb H, Hashemi M, Ansari-moghaddam S, Sheibak N. Estimation of volume density of interdental papilla components in patients with chronic periodontitis and interleukin-6 (-174G/C)gene polymorphisms. Dent Res J 2016 Mar-Apr;13(2):139-144.
2. Moghadam SA, Faghihi S, Amid R, Kadkhodazadeh M, Hosseini SK. Comprehensive oral rehabilitation of a patient with aplastic anemia by periodontal and prosthesis treatments. Compendium of continuing education in dentistry. 2012 Apr;33(4):e62-e66.
3. Sachs HA, Farnosh A, Checchi L, joseph CE. Current status of periodontal dressing. J periodontol 1984;55(12): 689-696.
4. Wennstrom JL, Heijl L, Lindhe J. Periodontal Surgery: Access Therapy. In: Lindhe J, Lang NP. Textbook of clinical periodontology and Implant Dentistry. 5nd ed. New York: Blackwell Publishing Ltd 2; 2008. p. 811.
5. Farnoush A. Techniques for the protection and coverage of the donor sites in free soft tissue grafts. J Periodontol 1978; 49(8):403-405.
6. David K, Shetty Neetha J, Swati P. Periodontal Dressings: An Informed View. Journal of pharmaceutical and biomedical sciences. J Pharm Biomed Sci 2013, January;26(26): 269-272.
7. Newman P, Addy M. A comparison of periodontal dressings and chlorhexidine gluconate mouthwash after the internal bevel flap procedure. J Periodontol 1978;49:576-579.
8. Kadkhodazadeh M, Baghani Z, Torshabi M, Basirat B. *In vitro* comparison of biological effects of Coe-Pak and Reso-Pac periodontal dressings. J Oral Maxillofac Res 2017;8(1):e3.
9. Soheilifar S, Bidgoli M, Soheilifar S, Jafari M. Evaluating the Effect of Periodontal Dressing on Wound Healing after Periodontal Flap Surgery. Sci J of Hamadan Univ Med Sci 2014;20(4): 267-272.
10. Ghanbari H, Forouzanfar A, Fatemi K, Mokhtari M, Abrishami M, Ebrahiminik Z, and Farazi F. Modified Widman flap procedure: With or without periodontal dressing? Open Journal of Stomatology, 2012;2:170-172.
11. Shanmugan M, Kumar T, Arun KV, Arun R, Kathik SJ. Clinical and histological evaluation of two dressing material in the healing of palatal wounds. J Indian Soc periodontal 2010;14:241-244.
12. Bae SB, Lim SB, Chung CH. A Comparative study of clinical effects following periodontal surgery with and without dressing. J Korean Acad Periodontal 1999 Sep;29(3): 693-701.
13. Petelin M, Pavlica Z, Batista U, Stiblar- Martincic D, Skaleric U. Effects of periodontal dressings on fibroblasts and gingival wound healing in dogs. Acta Veterinaria Hungarica. 2004;52:33-46.
14. Cheshire PD, Griffiths GS, Griffiths BM, Newman HN. Evaluation of the healing response following placement of Coe-pak and an experimental pack after periodontal flap surgery. J Clin Periodontol. 1996 Mar;23(1):188-193.
15. Moghare Abed A, Yaghini J, Tavakoli M, Amjadi MR, Najafian E. Evaluation of the effect of post-operative packing of gingiva on clinical signs and symptoms: a pilot study. J Isfahan Dental School 2011;6(6):705-711.
16. Sanz M, Newman MG, Anderson L, Matoska W, Otomo-Corgol J, Saltini C. Clinical enhancement of post periodontal surgical therapy by a 0.12% chlorhexidine gluconate mouth rinse. J Periodontol 1989;60(10):570-576.
17. Jorkend L, Skoglund LA.Effect of non-eugenol and eugenol containing periodontal dressings on the incidence and severity of pain after periodontal soft tissue surgery. J Clin Period 1990;17:341-344.
18. Augustine D, Rao RS, Anbu J, Chidambara Murthy K N. *In vitro* antiproliferative effect of earthworm coelomic fluid of Eudriluseugeniae, Eiseniafoetida, and Perionyxexcavatus

on squamous cell carcinoma-9 cell line: A pilot study. Phcog Res 2017; 9(Suppl S1):61-66.

19. Augustine D, Rao RS, Anbu J, Chidambara Murthy KN. Anti-cancer prospects of earthworm extracts: A systematic review of in vitro and in vivo studies. Phcog Rev 2018;12:46-55.
20. Augustine D, Rao RS, Jayaraman A, Chidambara Murthy K N. Anti-proliferative activity of earthworm coelomic fluid using oral squamous carcinoma KB 3-1 cells: An *in vitro* study with serine protease analysis. Phcog Mag 2018;14: 528-534.