Evaluation of temperature and healing in treatment of gingival enlargement using different gingivectomy techniques: A randomized controlled clinical study

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Abstract
Aim: Gingival enlargement comprises a common feature of gingival disease in which an increase in size of the gingiva is observed which may result from chronic or acute inflammatory changes. The aim of this study was to compare and investigate epithelization, gingival temperature, inflammation and pain levels in post-operative healing process in 4 different gingivectomy techniques including Er:YAG laser, Nd:YAG laser, electrocautery and conventional gingivectomy in treatment of chronic inflammatory gingival enlargements.

Material and Methods: Our split-mouth designed study was conducted on 37 systemically patients consisting of 19 females and 18 males, who had gingival enlargement areas on maxillar and mandibular anterior regions. Clinical periodontal parameters, gingival crevicular fluid (GCF) levels and gingival temperature levels were recorded before the investigation and during gingivectomy operations with different techniques, gingival temperature was measured. Gingival temperature and epithelization levels in 3, 7, 10 and 15 days; GCF levels in 15, 30 and 90 days and pain levels in 2, 8 hours and between 1-7 days of post-operative healing process were evaluated.

Results: When clinical periodontal parameters were compared, there was no significant difference found between all application within and between groups (p>0.05). When gingival temperature during operations were compared, there was a significant difference between all application groups (p<0.05). All application groups had similar GCF levels at baseline, 30, and 90 days (p>0.05). In 15 days, there was a significant difference between Nd:YAG laser group and the other groups (p<0.05). There were a significant difference in epithelization and pain levels in all applications between groups by time (p<0.05).

Conclusion: It was found that Er:YAG laser assisted gingivectomy technique which has better epithelization rates and rapidity, lower pain levels and no thermal damage effects in tissues, is more advantageous than other techniques.

Keywords: Electrosurgery; gingival enlargement; gingivectomy; laser; temperature

INTRODUCTION
Chronic inflammatory gingival enlargement is one of the most frequently encountered periodontal surgical requirements (1,2). Volumetric increases in the area of inflammation cause clinically significant changes in gingival morphology (3,4).

Excess, over contoured or enlarged gingival tissues should be removed in order to restore the natural anatomical contours (2).

Epithelization of the wound surface is completed within 5-14 days after surgical procedures (2,3). However, certain systemic and local factors such as systemic diseases, malnutrition, and drug use as systemic factors and trauma to wound, thermal damage, infection as local factors might compromise the wound healing process (3). Among these, thermal damage is the most pronounced factor to disrupt wound healing in electrosurgery and laser surgery procedures (3,4). Increase in the temperature in the wound area prevents the epithelization and prolongs the healing period (3,4). On the other hand, laser beam with long wavelength might improve the healing due to the stimulation of the biochemical processes in the wound (2,3). This stimulation effect of lasers occur through reducing toxins at the cellular level, increasing lymphatic fluid flow and blood supply, thereby promoting pain relief,
accelerating repair, and inducing regeneration through collagen and elastic fibers in the early phase of wound healing. Furthermore, antibacterial effect in irradiated tissues was also suggested (2,4).

The type of surgical approach is a significant factor which could affect the growth factors, cytokines, and inflammation in the wound healing process (5).

The present study hypothesized that the temperature alterations in different surgical methods would be different from each other and the epithelization would be affected from the temperature alteration. The aim of this study was to compare and investigate epithelization, gingival temperature, inflammation and pain levels in post-operative healing process in 4 different gingivectomy techniques including Er:YAG laser, Nd:YAG laser, electrocautery and conventional gingivectomy in treatment of chronic inflammatory gingival enlargements.

MATERIAL and METHODS

The present study was designed as a controlled clinical study with a randomized design. The study protocol was approved by the local ethical committee of Clinical Studies of Ondokuz Mayis University (2012/167). The study population consisted of 37 participants (19 women, age 23.10±2.78, 18 men age 21.11±2.63). The inclusion criteria were systemically healthy individuals, the existence of at least 20 functioning teeth, the existence of chronic inflammatory gingival enlargement in anterior quadrants. Exclusion criteria were; pregnancy/lactation, drug use, previous periodontal therapy within 6 months, previous antibiotic use within 6 months, smoking, and the existence of attachment/bone loss. All participants signed informed consent.

Study groups
1. Er: YAG laser (Fotana AT Fidelis III, Ljubljana, Slovenia) 200 mj, 10 Hz, 2 Watt and VLP (long pulse, 1000 μs); 1.3 mm diameter, 8 mm long cylindrical, a sapphire tip was used with air cooling and water irrigation.
2. Nd: YAG laser (Fotana AT Fidelis III, Ljubljana, Slovenia) was applied at 4 watts, 50 Hz, 300 μm microfiber tip and SP (short pulse: 180 μs) settings.
4. Conventional surgical method was applied with hand instruments and gingivectomy knives.

Clinical Parameters
Plaque index (PI) (6), gingival index (GI) (7), bleeding on probing (BOP) (8), probing depth (PD), gingival hyperplasia index (GHI) (4) were recorded and Gingival Crevicular Fluid (GCF) sampling was performed. Clinical periodontal measurements were obtained from 3 regions (mesio-buccal, mid-buccal, disto-buccal) via Williams periodontal probe (Hu-Friedy Co., Chicago, IL, USA) on the vestibule surface of the teeth in the operation area. Measurements were made before and after the operation in 15, 30, and 90 days. A single blinded examiner obtained all clinical measurements and non-surgical periodontal treatment (scaling and root planning) before surgery within two weeks. All patients were instructed with oral hygiene instructions and 4 weeks after non-surgical treatment, patients were evaluated for gingivectomy and gingivoplasty requirements. Patients who required surgical procedures were assigned another appointment and gingivectomy and gingivoplasty operations were performed by another clinician. Measurements of clinical parameters were achieved before to the surgical procedures. All patients underwent non-surgical therapy and oral hygiene motivation and 3 months after non-surgical therapy, after the lack of clinical symptoms of gingivitis was confirmed, surgical procedure was planned. Gingival hyperplasia index is based on vertical and horizontal components of gingival enlargement. All measurements were performed at anterior six teeth from right maxillary canine to the left maxillary canine.

Randomization
The distribution of surgical techniques to regions was decided randomly. In order to determine the randomization, paper drawing method was applied to the patients. The names of all application methods were written on a piece of paper and the papers were folded in a closed box so that the text could not be seen from the outside. Each patient received 4 times the paper from the box and the distribution of the surgical regions according to the quadrants was performed according to the order of arrival. The order of the quadrant to be applied is the maxillary right, maxillary left, mandibular right, and mandibular left quadrants. The randomization was performed by an experienced blinded clinician (I.K.).

Temperature measurements
The initial temperature of the gingival tissue was measured and recorded before surgery in each application site. In order to eliminate temperature variations that may be caused by external factors; individuals were asked not to eat and drink at least 30 minutes before the measurement (9). The temperature changes caused by both the laser or electrosurgery and the inflammation in the operation site were measured. During operation, the papillae between the first and second teeth adjacent to the midline in each quadrant (central and lateral teeth) were touched for 5 seconds. Then, the gingiva between the 2nd and 3rd teeth of the midline was touched (the lateral and canine teeth) for 10 seconds and the following measurement values were recorded:

<table>
<thead>
<tr>
<th>Measurement</th>
<th>Value</th>
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<tr>
<td>a: Highest temperature values reached after 5 seconds</td>
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<tr>
<td>b: Highest temperature values reached after 10 seconds</td>
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<tr>
<td>c: The recovery time of the temperature in the application zone of 5 seconds</td>
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<tr>
<td>d: The recovery time of the temperature in the application zone of 10 seconds</td>
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The areas to be removed with lasers were separated with custom-made protectors which were produced from impression material to avoid collateral laser beam
scattering (10).

The temperature measurement during the process was performed by an infrared thermometer. (Optris GMBH, Manuel LS, Berlin, Germany)

In conventional surgery, hand instruments and gingivectomy knives were used for gingivectomy.

All patients were prescribed with an analgesic (paracetamol, 2x1.5 days) and antiseptic mouthwash (0.12% chlorhexidine, 2x1.7 days).

**Standardized photographs**

Standard intra-oral photographs were taken and archived at a certain angle, distance and light level at baseline and in 3, 7, 10, and 15 days. Photos were taken by the same person. (M.M.T.) The same camera is used with the tripod mechanism at the same angle, distance and light values (10).

**Evaluation of the epithelization degree**

Epithelization was evaluated in postoperative 3, 7, 10 and 15 days. Wound surface epithelization in the healing process was evaluated with a staining solution (Mira-2-tone, GMBH & Co., Duisburg, Germany) (10).

The treatment area was isolated. The solution carrier was applied with cotton pellets. The standardized photographs were taken and transferred to the java based analysis program (Image J 1.31o, National Institutes of Health, Bethesda, MD, USA). The dark stained areas were calculated in the computer environment (10). In the image analysis program, the following method was used in the standardization of consecutive photographs taken during the post-operative follow-up period of the same individual:

The calibration was achieved by measuring the length of the periodontal probe in the consecutive photos. This ensures that the actual length value in the consecutive photos of the same patient is stored by the program. Following this process; painted areas that were drawn and fixed on the photo by the examiner. Each measurement step was carried out by the same examiner each time. In the photographs, the areas where the epithelization was incomplete, the areas of abrasion and the percent of epithelization area during the postoperative recovery were evaluated (10-12).

**Postoperative pain evaluation**

A visual analogue scale (VAS) was used to evaluate pain after surgeries. Patients were asked to mark the pain levels at the levels from 0 (no pain) to 10 (the most severe pain) on the scale in 2, 8 hours and 3, 7, 10, 15 days.

**GCF Sampling**

GCF was collected with paper strips (Periopaper®, Ora Flow Inc., Amityville, NY, USA) and the GCF volume measurement was calculated by automatic volume measurement device. (Periotron® 8000, Pro Flow Inc., Amityville, NY, USA) Paper strips were placed in the sulcus with moderate resistance and allowed to stand for 30 seconds.

**Statistical analysis**

The primary outcome of the present study was the temperature alterations and the secondary outcomes were clinical parameters, epithelization, pain, and GCF volumes. Data were evaluated with digital software (SPSS Inc., Chicago, IL, USA). All data were presented as mean± standard deviation or percentage, which is appropriate. A power analysis was performed before the study and 37 participants provided 90% power (a error of 0.05). The normality was tested with Kolmogorov-Smirnov test. Mann Whitney U, Kruskall Wallis, One Way ANOVA followed by Tukey, and chi-square tests were used. P<0.05 was considered statistically significant.

**RESULTS**

There was no complication and all the patients attended to all control sessions during the post-operative recovery period. The age and gender of the participants were similar in the study groups (p>0.05).

When clinical periodontal parameters were compared, there was no significant difference found between all application within and between groups (p>0.05) (Table 1, 2).

All groups had similar temperature level at baseline (p>0.05). A statistically significant difference was found between 5 seconds and 10 seconds application times in all groups (p<0.05).

For, Er:YAG laser group; there was no significant difference among the baseline temperature, 5, and 10 seconds temperatures (p>0.05).

For Nd:YAG laser group, the temperatures among the baseline, 5, and 10 seconds were statistically significant (p<0.05). Baseline and 5 seconds temperatures were significantly lower than 10 seconds and baseline temperature was also lower than 5 seconds temperature (p<0.05).

Electrosurgery temperature changes exhibited a similar pattern to the Nd:YAG laser temperatures. The significantly lowest temperature was observed at the baseline and the temperature significantly increased from 5 seconds to the 10 seconds (p<0.05).

The recovery time of the temperature were significantly lower in the Er:YAG laser group compared to the Nd:YAG laser and electrosurgery groups in both 5 and 10 seconds (p<0.05). Also, 10 seconds of application in all groups caused longer recovery time compared to the 5 second-application (p<0.05) (Figure 1). The temperature alterations in the tissues in 3, 7, 10, and 15 days in the Er:YAG laser, Nd:YAG laser, electrosurgery, and conventional surgeries were presented in Figure 2. The differences among the groups in 3, 7, and 15 days were statistically significant (p<0.05) (Figure 2).

The unepithelialized areas significantly decreased in each group from day 3 to 15 on each time (p<0.05).
Table 1. Plaque index, gingival index, probing depth, and bleeding on probing values of the study groups

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<tr>
<td><strong>Baseline</strong></td>
<td>1.11±/1.28±/3.71±/33±</td>
<td>1.13±/1.14±/3.75±/35±</td>
<td>1.12±/1.20±/3.85±/35±</td>
<td>1.12±/1.21±/3.82±/34±</td>
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<td><strong>Day 15</strong></td>
<td>2.42±/2.25±/1.49±/75±</td>
<td>2.38±/2.21±/1.51±/76±</td>
<td>2.38±/2.25±/1.50±/76±</td>
<td>2.38±/2.35±/1.52±/75±</td>
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<tr>
<td><strong>Day 30</strong></td>
<td>1.35±/1.25±/1.31±/53±</td>
<td>1.31±/1.27±/1.28±/63±</td>
<td>1.33±/1.28±/1.30±/53±</td>
<td>1.50±/1.28±/1.30±/63±</td>
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<tr>
<td><strong>Day 90</strong></td>
<td>1.17±/1.28±/1.31±/53±</td>
<td>1.33±/1.28±/1.28±/63±</td>
<td>1.33±/1.32±/1.30±/53±</td>
<td>1.28±/1.31±/1.29±/53±</td>
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There was no difference intergroup comparisons while there was a difference in the time comparisons of each group. Superscripts a,b,c indicates comparisons of plaque index (PI), x,y,z indicates comparisons of gingival index (GI), k,m,n indicates comparisons of probing depth (PD), and 1,2,3 indicates comparisons of bleeding on probing (BOP).

There was no difference intergroup comparisons while there was a difference in the time comparisons of each group. Superscripts a,b,c indicates comparisons of plaque index (PI), x,y,z indicates comparisons of gingival index (GI), k,m,n indicates comparisons of probing depth (PD), and 1,2,3 indicates comparisons of bleeding on probing (BOP).

Table 2. Vertical and horizontal gingival hyperplasia index based on time in the study groups

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<tr>
<td><strong>Baseline</strong></td>
<td>1.00 (1.00-2.33)ab</td>
<td>1.00 (1.00-2.33)ab</td>
<td>1.25 (1.00-3.00)ab</td>
<td>1.25 (1.00-3.00)ab</td>
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<td><strong>Day 15</strong></td>
<td>0.00 (0.00-1.10)ab</td>
<td>0.00 (0.00-1.00)ab</td>
<td>0.00 (0.00-1.10)ab</td>
<td>0.00 (0.00-1.10)ab</td>
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<tr>
<td><strong>Day 30</strong></td>
<td>0.00 (0.00-1.10)ab</td>
<td>0.00 (0.00-1.00)ab</td>
<td>0.00 (0.00-1.10)ab</td>
<td>0.00 (0.00-1.10)ab</td>
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<tr>
<td><strong>Day 90</strong></td>
<td>0.00 (0.00-1.10)ab</td>
<td>0.00 (0.00-1.00)ab</td>
<td>0.00 (0.00-1.10)ab</td>
<td>0.00 (0.00-1.10)ab</td>
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<tr>
<td><strong>Baseline</strong></td>
<td>1.00 (1.00-2.11)ab</td>
<td>1.00 (1.00-2.33)ab</td>
<td>1.00 (1.00-2.28)ab</td>
<td>1.00 (1.00-2.14)ab</td>
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<tr>
<td><strong>Day 15</strong></td>
<td>0.00 (0.00-1.20)ab</td>
<td>0.00 (0.00-1.20)ab</td>
<td>0.00 (0.00-1.00)ab</td>
<td>0.00 (0.00-1.10)ab</td>
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<tr>
<td><strong>Day 30</strong></td>
<td>0.00 (0.00-1.20)ab</td>
<td>0.00 (0.00-1.20)ab</td>
<td>0.00 (0.00-1.00)ab</td>
<td>0.00 (0.00-1.10)ab</td>
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<tr>
<td><strong>Day 90</strong></td>
<td>0.00 (0.00-1.20)ab</td>
<td>0.00 (0.00-1.20)ab</td>
<td>0.00 (0.00-1.00)ab</td>
<td>0.00 (0.00-1.10)ab</td>
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The data was presented as median and minimum-maximum. a, b, and c presents statistical difference in the columns. No statistical significance was observed among the groups in the rows.
The Er:YAG laser group exhibited significantly higher epithelization areas compared to the conventional surgery, electrosurgery, and Nd:YAG laser group in 3, 7, 10, and 15 days (p<0.05). On the day 15, electrosurgery provided lower epithelization compared to the Nd:YAG laser surgery which was the opposite in 3, 7, and 10 days (p<0.05) (Figure 3,4).

The Er:YAG laser, Nd:YAG laser, electrosurgery, and the conventional surgery groups had similar GCF levels at baseline, 30, and 90 days (p>0.05). However, in 15 days, Nd:YAG laser surgery group had significantly higher levels compared to the other groups (p<0.05) (Figure 5).

Figure 3. Average epithelialized area ratios (%) between treatment groups in 3, 7, 10 and 15 days

Figure 4. Stained areas during healing process. a: preoperative, b: postoperative, c: 3, d: 7, e: 10 and f: 15 days

Figure 5. Graphical demonstration of preoperative, 15, 30 and 90 day values of GCF values (µl) in treatment groups

Er:YAG laser surgery group had significantly lower pain levels compared to the other groups in 2 hour, 8 hour and 1 day compared to the other groups (p<0.05). In 2, 3, 4, and 5 days, Er:YAG laser and conventional surgery groups caused lower pain levels compared to the Nd:YAG laser and electrosurgery groups (p<0.05). In 6 and 7 days, all groups caused similar pain levels (p>0.05) (Figure 6).

Figure 6. Graphic demonstration of Visual Analogue Scale results

DISCUSSION

The present study was the first investigation which evaluated the alterations in the temperature, epithelization, and pain levels after four different gingivectomy techniques. The results revealed that Er:YAG laser surgery exhibited similar results with conventional surgery compared to the Nd:YAG laser and electrosurgery which also provided similar results in all parameters.

One of the most important factor affecting the wound healing after gingivectomy operations is the changes in the tissue temperature during the operation (13). During the operation, the temperature change from 37°C to 50°C causes bacterial inactivation and sterilization at the surgical site. Coagulation occurs in tissues when the temperature reaches 60°C (14) providing the control of bleeding (15). However, when tissue temperature exceeds 60°C, denaturation in the tertiary structure of amino acids and irreversible deterioration in protein structure
occur (14,16). Temperatures higher than 200°C causes further dehydrated and burnt tissues and carbonization in the tissue (14,17). Therefore; the temperature changes during the laser and electrosurgeries might cause thermal collateral damages (17) and the tissue recovery could be delayed with increased patient discomfort (14). Schwarz et al. reported that Nd:YAG laser may cause thermal damage to the periodontal tissues due to its high tissue penetration (18). Parker et al. also revealed that the thermal damage in the tissues is closely associated with the length of the laser waves which is shorter in the Nd:YAG lasers which can penetrate up to 2-6 mm in the tissues while long-wavelength laser penetration is shorter (19). Cobb also suggested that minimal thermal tissue damage in Er:YAG laser applications are caused by shorter wave penetration (20). An alteration observed in the gingival tissues with inflammation is the increase in the GCF volume and GCF levels are expected to increase depending on the level of post-operative inflammation and tend to return to normal levels after wound healing (2). After the gingivectomy, baseline GCF levels all increased in the study groups. The recovery of the GCF levels was observed in 30 days. In the Nd:YAG laser group, the GCF levels were significantly higher in 15 days.

The increasing of temperature is one of the 5 main symptoms of inflammation with rubor, tumor, dolor and function loss (21). The mean temperature difference between healthy and diseased periodontal tissues was reported to be 0.3°C (22) caused by vasodilatation and increase in blood flow rate accompanying periodontal inflammation (23). Along with the temperature alterations in the tissue, the return to normal temperature should also be considered. Tissues should be allowed to cool immediately after thermal stimulant and tissue contact at the same point for a long time should be avoided (15). Incisions made with scalpel do not cause any thermal damage, but might cause mechanical trauma and dilatation in lymph and blood vessels and consequently a large amount of inflammatory response and increase in the temperature due to the inflammation. The temperature increase in the conventional surgery group also supports this situation (24). Among the lasers and electrosurgery procedures, the initial temperature of the tissues were similar however, in 3 and 7 days, Er: YAG laser caused lower temperatures compared to the Nd:YAG laser and electrosurgery. In days 10 and 15, tissue temperatures were similar. Apart from the increase in the temperature, the recovery of the tissue temperature is also an important factor after laser or electrosurgery procedures (24). The tissues underwent Nd:YAG laser surgery required a longer time to regular tissue temperatures than electrosurgery. The significantly shortest time required for recovery was observed in the Er: YAG laser group. This finding is compatible with Monzavi et al.’s report indicating a fast temperature recovery after Er: YAG laser treatment compared to Nd: YAG laser (25).

In the present study, the effects of various gingivectomy procedures on gingival tissues were evaluated via a split mouth design study protocol which decreases the compromising effects of individual factors like gender, age, systemic factors on wound healing and provides a better comparison among the study groups. Epithelialization begins within a few hours after injury. The epithelial cells move towards the wound area due to the growth factors secreted from the platelets and macrophages (26) and the epithelization process starts within 24-48 hours in uncompromised wound healing (27). Studies reported improved wound healing and epithelization after Er:YAG laser surgeries comparable even better results with conventional surgery procedures (28,29). Sawabe et al. recently showed that Er: YAG laser provided faster healing period with improved epithelization compared to electrosurgery (30). Tao et al. also reported that Er: YAG laser when compared to Nd: YAG laser exhibited better gingival wound healing within a shorter time (31). On the other hand, Goultschin et al. demonstrated increased inflammatory infiltrate and ulcerative epithelial tissue after laser gingivectomy procedures and concluded that conventional surgery provided better wound healing (32). Unlike Er: YAG laser, other lasers such as Nd: YAG laser, CO2, and diode lasers were reported to increase soft tissue damage caused by large ablation areas and thermal damage (33,34).

Several methods have been developed to measure pain, being most common one the VAS analysis which was also utilized in the present study (10,35-38). The early evaluation of pain in 2 and 8 hours and 1 day after gingivectomy procedures showed significantly lower pain in Er: YAG laser surgery group. The patients underwent conventional surgery reported higher pain than Er: YAG laser but lower than electrosurgery and Nd: YAG laser surgeries on early period until 2 days but the VAS scores of 2, 3, and 5 days were similar to Er:YAG laser group. After 6 days, all patients reported similarly lower pain. Similarly, Fekrazad et al. found that Er: YAG laser surgery caused no pain and patient discomfort (39). Chandna et al. compared the pain after diode laser and electrosurgery and found lower patient discomfort and pain after laser surgery (40). As for the present study, Nd: YAG laser and electrosurgery caused more pain compared to the Er: YAG laser and conventional surgeries. A possible reason is high heat damage. Nonetheless, the present results revealed that operative temperature alterations might affect the epithelization and pain levels in post-operative period.

CONCLUSION

The present study evaluated the effects of four different gingivectomy techniques on epithelization, gingival temperature, GCF levels, and pain. The result revealed that Er: YAG laser assisted gingivectomy technique which has better epithelization rates and rapidity, lower pain levels and no thermal damage effects in tissues, is more advantageous than other techniques.
REFERENCES


