



Temperature elevation during root canal treatment with a 445-nm diode laser—an in vitro study

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Abstract

Objective This study investigated the thermal effects of a 445-nm diode laser on tooth roots of endodontic treatment.

Material and methods A total of 15 human single-rooted extracted teeth were included in the survey. Samples were sectioned at the cemento-enamel junction. The root canals were instrumented using a rotary system. Each root was fitted into a resin mold and was fixed in a vertical position using clamps. All samples were placed on a support in a thermal path at 37 °C. The root canals were randomly divided into three groups ($n = 5$ each): samples were irradiated with 0.6 W continuous wave (CW), 0.4 W CW, and 1.2 W gated mode, at a repetition rate of 10 Hz with a pulse duration of 50 ms. The time-dependent temperature elevations of the cervical, middle, and apical thirds of the roots were recorded at a sampling rate of 2 Hz during laser irradiation with a K-type thermocouple measurement system.

Results The mean temperature rise inside the roots in this study was below 10 °C, whereas the maximum temperature rise of 9.83 °C (0.6 W CW) and 9.81 °C (1.2 W gated mode, repetition rate 10 Hz, and pulse duration 50 ms) was registered in lower incisors, and 4.97 °C was detected at 0.4 W CW. The thermal changes seen in the apical thirds of the root were statically significantly ($p < 0.001$) and were greater than those at the coronal and middle thirds.

Conclusion The results of this study suggest that 445-nm diode lasers may provide a safe temperature for endodontic applications.

Keywords Dental laser · Endodontics · Diode laser · Thermal damage · Root canal · Temperature

Introduction

The goal of primary endodontic treatment is to improve root canal disinfection and to prevent re-infection [1–4]. Therefore, there is an agreement that an important aim of the endodontic therapy is to eliminate the microorganisms in the root canal [5].

To date, several studies [6–9] have shown that bio-mechanical, also known as chemo-mechanical, methods play a significant role in the treatment of root canal systems. On the one hand, these methods cannot completely remove the smear layer, preventing the penetration of chemical irrigating solutions into dentinal tubules [6, 10], leading to, as a consequence, less than full elimination of nested bacteria [4, 11, 12]. In fact, the method is mostly limited to a dentin depth between 100 and 300 μm . Deeper dentin tubules house many thousands of remaining bacteria, and these bacteria can migrate into and multiply inside the root canal system [13, 14]. Furthermore, the complexity of the root canal system is well known to be one of the major challenges in root canal preparation [6], lateral canals, ramifications, and accessory canals where conventional treatments cannot reach bacteria [6, 15, 16]. This may result in failure after endodontic treatment and a recurrence of the infection [17, 18].

The advantage of using lasers is due to their high disinfection efficiency, using photo-thermal and photo-mechanical effects [19] to enhance cleaning, the reduction of bacteria [18],

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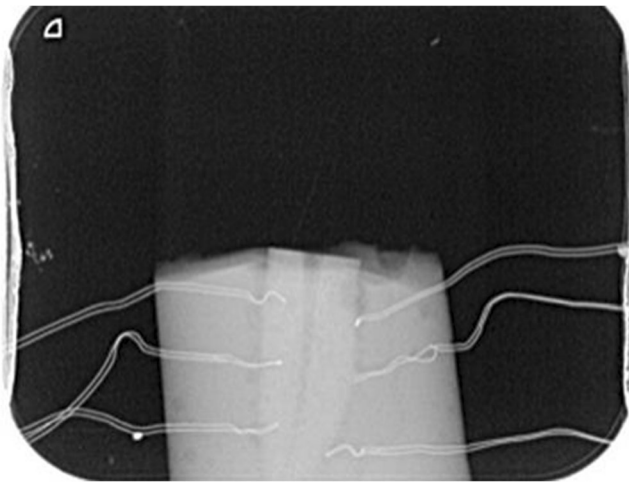


Fig. 1 Radio-graph of a sample of tooth showing the positions of all thermocouples

and removal of the smear layer in root canals [20]. Unlike the conventional method, lasers of suitable wavelengths have an excellent capacity for bacterial reduction at elimination depths exceeding 1000 μm in deep dentin tubules [13, 21, 22]. However, despite the potential benefits of lasers, too high a dose of the absorbed energy may cause thermal damage to periodontal ligaments and lead to bone necrosis [15, 23, 24]. To achieve favorable outcomes and avoid thermal damage to the surrounding tissue, safe parameters must be identified [25, 26].

Goya et al. (2007) considered that successful disinfection of root canals by lasers depends on many factors, such as output power, irradiation time, repetition rate, and pulse duration [27]. According to this study, laser irradiation of one particular wavelength induced different effects on the same tissue under various parameters. The most common wavelengths of diode lasers used in endodontics are 810, 940, and 970–980 nm and Nd:YAG lasers at 1064 nm. Nonetheless, excessive thermal heating of the surrounding tissue must be avoided.

Recently, there have been improvements in diode laser development and success in the expansion of the wavelength range from ultraviolet (UV) to blue-green with high optical output power [28].

Aim of the study

The purpose of the present study was to assess root surface and subsurface temperature elevation using a diode laser emitting at 445 nm in an *in vitro* setup, in order to identify safe irradiation parameters for a later *in vivo* study. These values are yet unknown, and the acceptable safe powers are suspected to deviate from those of near-infrared diode lasers

due to the different absorption characteristics of hydroxyapatite in the visible part of the electromagnetic spectrum at 445 nm. Tested irradiation parameters include 0.6 W continuous wave (CW), 0.4 W CW, and 1.2 W gated mode, with a repetition rate of 10 Hz, pulse duration of 50 ms, and a pulse pause of 50 ms, yielding a 0.5 duty cycle.

Material and methods

Sample preparation

Fifteen single-rooted human teeth were used in this study, which had been extracted for medical reasons, which otherwise would have been disposed of as medical waste. First, remaining calculus and residual tissue were removed from the root surface. Samples were sectioned at the cemento-enamel junction (CEJ), using a diamond saw (Exakt Apparatebau, Hamburg, Germany). All samples were shortened to a length of 14 mm from CEJ, and crowns were discarded. The root canals were enlarged using a rotary system up to an apical size of ISO #40 at 1 mm from the apex. Canals were rinsed with NaOCl (2.5%) between each file, EDTA (17%), and eventually sterile water was applied to flush out any remaining irrigants and to remove any precipitates [29]. All canals were dried with paper points.



Fig. 2 A model was placed on a support in a thermal path of 37.0 °C

Table 1 Descriptive statistics of group I

Group I	T mean [°C]	Tmax [°C]	SD [°C]
Buccal-cervical (<i>n</i> = 5)	38.14	41.06	1.10
Buccal-middle (<i>n</i> = 5)	39.38	44.75	1.82
Buccal-apical (<i>n</i> = 5)	39.88	46.83	2.31
Lingual-cervical (<i>n</i> = 5)	37.59	38.91	0.53
Lingual-middle (<i>n</i> = 5)	37.76	38.86	0.65
Lingual-apical (<i>n</i> = 5)	37.98	39.33	0.78

Temperature measurements and groups

Afterward, three holes were drilled by using a low-speed handpiece bur on the buccal side into each root surface at the cervical third (1 mm depth), the middle third (1 mm depth), and apical third (0.5 mm depth) approximately 0.2 mm away from the apex. This was to measure the temperature in the body of the dentine of the root canals by placing thermocouples into these holes. Three more superficial holes were then drilled on the lingual surface along the root canal surface, i.e., the cervical, middle, and apical thirds, to measure the temperature on the surface of the root. Each root was put into a mold that had been filled with polyurethane resin material mixed in a ratio of 5:1 (ISO-PUR K 760; Elze, Germany); the *hardening process* of polyurethane resin changed at regular intervals, and it took 40 to 45 min to harden completely. This casting resin was chosen for its thermal conductivity of 0.6 W/Km which is similar to that of cortical bone (0.58 W/Km) [23, 30]. Before the resin hardened, the access holes to the root canal and to the drilled holes on both sides of the root canal were exposed. Six K-type thermocouples with an accuracy of ± 0.691 °C and diameter of 0.13 mm were fixed inside the buccal and lingual holes and connected to a USB data acquisition module (5TC-TT-KI-36-1M, Omega Engineering Inc.; Stamford, USA). The positions of all thermocouples have been checked radio-graphically (Fig. 1). Thermal paste (Revoltec Thermal Grease) was placed at the end of the thermocouples to optimize the contact with the root surface to allow for optimal temperature readings, and was fixed to the root surface holes using adhesive wax. Samples

Table 2 Descriptive statistics of group II

Group II	T mean [°C]	Tmax [°C]	SD [°C]
Buccal-cervical (<i>n</i> = 5)	37.52	39.07	0.55
Buccal-middle (<i>n</i> = 5)	38.43	41.65	1.10
Buccal-apical (<i>n</i> = 5)	38.43	41.97	1.23
Lingual-cervical (<i>n</i> = 5)	37.00	37.87	0.30
Lingual-middle (<i>n</i> = 5)	37.29	38.36	0.31
Lingual-apical (<i>n</i> = 5)	37.51	40.53	0.75

Table 3 Descriptive statistics of group III

Group III	T mean [°C]	Tmax [°C]	SD [°C]
Buccal-cervical (<i>n</i> = 5)	38.52	43.63	1.65
Buccal-middle (<i>n</i> = 5)	39.37	44.78	2.09
Buccal-apical (<i>n</i> = 5)	40.19	46.81	2.58
Lingual-cervical (<i>n</i> = 5)	37.55	38.44	0.49
Lingual-middle (<i>n</i> = 5)	37.97	39.22	0.56
Lingual-apical (<i>n</i> = 5)	39.03	43.48	1.71

were fixed in a vertical position by using clamps and an adjustable resin mold. The USB data acquisition module was connected to a PC, and the recorded temperature values inside the root canal could be observed and recorded on the PC in real time for each individual thermocouple.

The temperature elevations were measured simultaneously at the apical, medium, and cervical regions of the root with a sampling rate of 2 Hz. Temperature variation results were submitted for further analysis of variance (ANOVA).

For the irradiation, all models were placed on a support in a thermal path of 37.0 ± 0.5 °C (GFL-Wasserbäder, Type 303, Germany) to simulate the temperature in the mouth (Fig. 2). This method has been successfully applied before, as documented in previous studies [23, 30].

Laser system

In this study, a dental diode laser emitting visible blue light at 445 nm was used (DENTSPLY SIRONA, Sirona Dental Systems GmbH, Bensheim, Germany).

The models, respective the roots, were then randomly distributed into three groups (*n* = 5) with the following settings:

1. Group I: 0.6 W CW (*n* = 5)
2. Group II: 0.4 W CW (*n* = 5)
3. Group III: 1.2 W gated, repetition rate 10 Hz, 50 ms pulse duration (*n* = 5)

For all three groups, a proprietary 200- μ m-diameter endodontic fiber tip (the spot area of the fiber is 0.333 cm²) was used in conjunction with the diode laser. The emitted power at the distal fiber end was confirmed with a power meter (coherent power/energy meter model no. FM/GS) before each

Table 4 Summarized descriptive results of groups I, II, and III

Group	<i>n</i>	T mean [°C]	Tmax [°C]	SD [°C]
I	5	38.45	46.83	1.62
II	5	37.70	41.97	0.96
III	5	38.78	46.81	1.91

Table 5 Statistical analysis among the three groups: I, II, and III

Group	I	II	III
Power value	0.6 W CW	0.4 W CW	1.2 W gated, repetition rate 10 Hz, 50 ms pulse duration
n [-]	5	5	5
ΔT Mean [$^{\circ}$ C]	1.45	0.70	1.78
ΔT Max [$^{\circ}$ C]	9.83	4.97	9.81
SD [$^{\circ}$ C]	1.62	0.96	1.91
<i>p</i> (ANOVA)	$p < 0.001$ (I and II), and (I and III)		
Confidence interval	1.40 to 1.51	0.67 to 0.72	1.72 to 1.83

specimen. The irradiation was repeated four times for each specimen.

This method has been successfully applied in previous studies [24, 31, 32]. The fiber was inserted into the root canal up to the apex; then, the irradiation was started, and the fiber was moved in helicoidal movements along the length of the root canal, up to the cervical part at a vertical speed of approximately 2 mm/s [33], i.e., if the length of a root canal was 14 mm, laser irradiation would be applied for 7 s. Between every single irradiation, an adequate rest period was given for the root canal of around 10 s. This time was also needed to carefully reintroduce the fiber back into the root canal.

Results

To determine the maximum temperature rise in the apical, medium, and cervical regions, the temperature variation

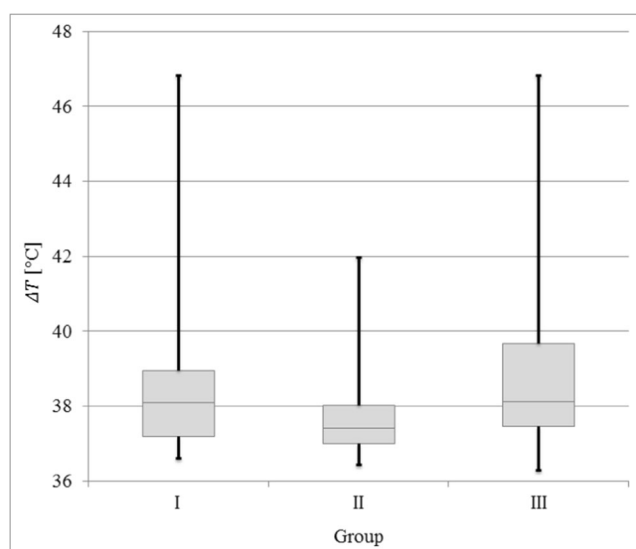


Fig. 3 Boxplot graph displays the statistical variation in the three groups: I, II, and III

(ΔT) of each irradiation cycle was calculated. In general, the maximum temperature change during irradiation in this study was reported as less than 10 $^{\circ}$ C. The difference in the mean values between the groups was greater for the apical region, followed by those of the middle and cervical regions. The mean temperature rise at 0.6 W CW (group I) was 2.88 $^{\circ}$ C while the maximum temperature recorded was 9.83 $^{\circ}$ C. The mean and maximum temperature rises at 0.4 W CW (group II) were 1.43 and 4.97 $^{\circ}$ C, respectively. However, for group III, with a gated output power of 1.2 W, a repetition rate of 10 Hz, and a pulse duration of 50 ms, the recorded mean and maximum temperature values were 3.19 and 9.81 $^{\circ}$ C, respectively.

The descriptive statistics are shown in Tables 1, 2, 3, and 4 and cover the detailed temperature measurements for each group (mean temperature, maximum temperature, and standard deviations). Comparisons of the mean temperature rises and maximum temperature variations among the three groups were statistically carried out and tested by ANOVA and are illustrated in Table 5 and in Fig. 3. The ANOVA indicated a statistically significant difference ($p < 0.001$).

A comparison of each output power in relation to the intra-group thirds (cervical, middle, and apical) is shown in Table 6. All the areas showed significant difference ($p < 0.001$) and show that the highest mean temperature for the apical third is followed by those of middle and cervical thirds. Besides, similar results can be seen for the standard deviation, which is also higher in the apical area.

Discussion

According to Eriksson and Albrektsson [34] (1983), a temperature threshold level of 10 $^{\circ}$ C for 1 min can cause necrosis of the alveolar bone. While this study was done on rabbit teeth, it is still often referred to in temperature-related studies on human bone as well. In the present study, the use of a 445-nm diode laser during root canal therapy at 0.6 W in continuous mode, 0.4 W in continuous mode, and at 1.2 W in gated mode at 10 Hz, with a pulse duration of 50 ms and a pulse pause of

Table 6 The statistical analysis of each group in relation to the intra-group thirds

Root surface area	Group	<i>n</i>	Mean [°C]	SD [°C]	<i>p</i> (ANOVA)	Confidence interval
Cervical	I	5	37.91	0.92	<i>p</i> < 0.001	0.85 to 0.96
	II	5	37.24	0.46		0.22 to 0.27
	III	5	38.04	1.31		0.98 to 1.10
Middle	I	5	38.25	1.39	<i>p</i> < 0.001	1.17 to 1.33
	II	5	37.51	0.60		0.48 to 0.54
	III	5	38.68	1.70		1.60 to 1.76
Apical	I	5	38.92	1.99	<i>p</i> < 0.001	1.81 to 2.03
	II	5	37.97	1.12		0.91 to 1.02
	III	5	39.61	2.27		2.51 to 2.72

50 ms, provided temperature rises below the physiologically critical value of 10 °C.

The mean differences in temperature between the three groups were tested by ANOVA. The results showed significant differences at a significance level of $\alpha = 5\%$ among the tested groups in the cervical, middle, and apical root thirds ($p < 0.001$). In the present study, the temperature reached generally higher values in the apical thirds of the specimens, followed by those of the middle and cervical thirds. The results are in agreement with Strakas et al. (2013), who stated that the difference between the mean values for pulse duration differences is higher for apical points followed by those of mesial and coronal points [35]. In contrast, Falkenstein et al. (2014) found that the highest values for ΔT_{max} were in the middle of the root [30], due to the movement of the fiber tip which was guided parallel to the root surface in a constant horizontally swinging movement (~ 2 mm).

An important factor, which should be taken into consideration, is the thickness of the dentin, where higher temperature rises can be anticipated in teeth with low mass and therefore low heat capacity, which clinically present as teeth with thin dentin. In this experiment, the highest temperature values of ΔT_{max} 9.83 and 9.81 °C were found specifically in the apical area of lower incisors in both groups I and III, respectively. The mean differences in the temperature elevations were statistically significant in the apical third of the root ($p < 0.001$) due to the thickness of the dentinal tubules, where the thinner walls in the apical area are the most susceptible to thermal damage [32].

The rest period between every irradiation cycle in this study was around 10 s. This rest period was proposed by Gutknecht et al. [33] and is critical to avoid temperature increases above the safe limit, with the fiber tip being displaced at a speed of approximately 2 mm/s. In the present study, the maximum temperature value in group I at 0.6 W (CW) was 9.83 °C, while in group III at 1.2 W and 10 Hz, with a pulse duration of 50 ms and 50% duty cycle, the temperature increase was 9.81 °C. There was no significant difference between the mean values of ΔT_{max} of these two groups. The

results of this study did not exceed the critical temperature supported by periodontal tissue.

Conclusion

The results of this study suggest that 445-nm diode lasers may be a safe temperature-wise for endodontic applications at the investigated parameters of 0.6 W CW, 0.4 W CW, 1.2 W, 10 Hz, 50 ms pulse duration, and 50% duty cycle, when using the described protocol. In this study, the use of these parameters did not exceed the 10 °C limit proposed by Eriksson and Albrektsson [34], so the protocol can be considered safe within the limitation of this in vitro study. Therefore, we suggest a follow-up study to assess the bactericidal capabilities of these parameters.

Compliance with Ethical Standards

Conflict of Interest The authors declare that they have no conflict of interest.

References

- Chakraborty P, Chattopadhyay U (2005) A study on the polymicrobial etiology of root canal infections in anterior non-vital teeth in a Government Hospital in Kolkata, India. *Eur Rev Med Pharmacol Sci* 9(2):113–116
- Gutknecht N (2008) Lasers in endodontics. *J Laser Health Acad* 4(1):1–5
- Camargo ASCC (2012) The antibacterial effects of lasers in endodontics. *Infection* 1:32–43
- Vivacqua-Gomes N, Gurgel-Filho ED, Gomes BP, Ferraz CC, Zaia AA, Souza-Filho FJ (2005) Recovery of *Enterococcus faecalis* after single- or multiple-visit root canal treatments carried out in infected teeth ex vivo. *Int Endod J* 38(10):697–704. <https://doi.org/10.1111/j.1365-2591.2005.00992.x>
- Nair P, Henry S, Cano V, Vera J (2005) Microbial status of apical root canal system of human mandibular first molars with primary apical periodontitis after “one-visit” endodontic treatment. *Oral Surg Oral Med Oral Pathol Oral Radiol Endodontology* 99(2):231–252

6. Hülsmann M, Peters OA, Dummer PM (2005) Mechanical preparation of root canals: shaping goals, techniques and means. *Endod Top* 10(1):30–76
7. Berutti E, Marini R, Angeretti A (1997) Penetration ability of different irrigants into dentinal tubules. *J Endod* 23(12):725–727
8. Vianna M, Horz H, Gomes B, Conrads G (2006) In vivo evaluation of microbial reduction after chemo-mechanical preparation of human root canals containing necrotic pulp tissue. *Int Endod J* 39(6):484–492
9. de Gregorio C, Estevez R, Cisneros R, Paranjpe A, Cohenca N (2010) Efficacy of different irrigation and activation systems on the penetration of sodium hypochlorite into simulated lateral canals and up to working length: an in vitro study. *J Endod* 36(7):1216–1221
10. Card SJ, Sigurdsson A, Ørstavik D, Trope M (2002) The effectiveness of increased apical enlargement in reducing intracanal bacteria. *J Endod* 28(11):779–783
11. Gu X-H, Mao C-Y, Kern M (2009) Effect of different irrigation on smear layer removal after post space preparation. *J Endod* 35(4):583–586
12. Love R (2001) *Enterococcus faecalis*—a mechanism for its role in endodontic failure. *Int Endod J* 34(5):399–405
13. Gutknecht N, Franzen R, Schippers M, Lampert F (2004) Bactericidal effect of a 980-nm diode laser in the root canal wall dentin of bovine teeth. *J Clin Laser Med Surg* 22(1):9–13
14. Mancini M, Armellini E, Casaglia A, Cerroni L, Cianconi L (2009) A comparative study of smear layer removal and erosion in apical intraradicular dentine with three irrigating solutions: a scanning electron microscopy evaluation. *J Endod* 35(6):900–903
15. Huggins H (2010) Root Canal Dangers. <http://www.westonaprice.org/holistic-healthcare/root-canal-dangers/>. Accessed June 25 2010
16. Ruddle CJ (2002) Cleaning and shaping the root canal system. In: Cohen S, Burns RC, eds. *Pathways of the pulp*. 8th ed. In: *Pathways of the Pulp*. Mo: Mosby, St Louis, pp 231–291
17. Love R, McMillan M, Jenkinson H (1997) Invasion of dentinal tubules by oral streptococci is associated with collagen recognition mediated by the antigen I/II family of polypeptides. *Infect Immun* 65(12):5157–5164
18. Schäfer E (2007) Irrigation of the root canal. *Endo* 1:11–27
19. Olivi G, Crippa R, Iaria G, Kaitsas V, DiVito E, Benedicti S (2011) Laser in endodontics (part II). *Roots* 2:6–12
20. Asnaashari M, Safavi N (2013) Disinfection of contaminated canals by different laser wavelengths, while performing root canal therapy. *J Lasers Med Sci* 4(1):8–16
21. Franzen R, Gutknecht N, Falken S, Heussen N, Meister J (2011) Bactericidal effect of a Nd: YAG laser on *Enterococcus faecalis* at pulse durations of 15 and 25 ms in dentine depths of 500 and 1,000 μm . *Lasers Med Sci* 26(1):95–101
22. Gutknecht N, Al-Karadaghi TS, Al-Maliky MA, Conrads G, Franzen R (2016) The bactericidal effect of 2780 and 940 nm laser irradiation on *Enterococcus faecalis* in bovine root dentin slices of different thicknesses. *Photomed Laser Surg* 34(1):11–16
23. Al-Karadaghi TS, Franzen R, Jawad HA, Gutknecht N (2015) Investigations of radicular dentin permeability and ultrastructural changes after irradiation with Er, Cr: YSGG laser and dual wavelength (2780 and 940 nm) laser. *Lasers Med Sci* 30(8):2115–2121
24. He H, Yu J, Song Y, Lu S, Liu H, Liu L (2009) Thermal and morphological effects of the pulsed Nd: YAG laser on root canal surfaces. *Photomed Laser Surg* 27(2):235–240
25. Gutknecht N, Franzen R, Lampert F (2002) Finite element study on thermal effects in root canals during treatment with a surface-absorbed laser. *Lasers Med Sci* 17:137–144
26. Todea C, Kerezi C, Balabuc C, Calniceanu M, Filip L (2008) Pulp capping—from conventional to laser-assisted therapy (I). *J Oral Laser Appl* 8:147–155
27. Goya C, da Silveira BL, Aranha ACC, Zzell DM, Matsumoto K, de Paula EC (2007) In vitro evaluation of Er: YAG and Nd: YAG Vaser irradiation on root canal walls—a preliminary study. *J Oral Laser Appl* 7:45–53
28. Kozaki T, Matsumura H, Sugimoto Y, Nagahama S-i, Mukai T High-power and wide wavelength range GaN-based laser diodes. In: *Integrated optoelectronic devices 2006*. International Society for Optics and Photonics, pp 613306–613306–613312
29. Alfredo E, Marchesan M, Sousa-Neto M, Brugnera-Junior A, Silva-Sousa Y (2008) Temperature variation at the external root surface during 980-nm diode laser irradiation in the root canal. *J Dent* 36(7):529–534
30. Falkenstein F, Gutknecht N, Franzen R (2014) Analysis of laser transmission and thermal effects on the inner root surface during periodontal treatment with a 940-nm diode laser in an in vitro pocket model. *J Biomed Opt* 19(12):128002–128002
31. da Costa RA, Nogueira GEC, Antoniazzi JH, Moritz A, Zzell DM (2007) Effects of diode laser (810 nm) irradiation on root canal walls: thermographic and morphological studies. *J Endod* 33(3):252–255
32. Pradhan S, Karnik R (2011) Temperature rise on external root surface during laser endodontic therapy using 940 nm diode laser: an in vitro study. *Int J Laser Dent* 1 (December):29–35. <https://doi.org/10.5005/jp-journals-10022-1004>
33. Gutknecht N, Franzen R, Meister J, Vanweersch L, Mir M (2005) Temperature evolution on human teeth root surface after diode laser assisted endodontic treatment. *Lasers Med Sci* 20(2):99–103
34. Eriksson A, Albrektsson T (1983) Temperature threshold levels for heat-induced bone tissue injury: a vital-microscopic study in the rabbit. *J Prosthet Dent* 50(1):101–107
35. Strakas D, Franzen R, Kallis A, Vanweersch L, Gutknecht N (2013) A comparative study of temperature elevation on human teeth root surfaces during Nd: YAG laser irradiation in root canals. *Lasers Med Sci* 28(6):1441–1444