RESEARCH

Comparison of Er:YAG Modalities (PIPS-SWEEPS) on Eliminating of Enterococcus Faecalis Populations

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ABSTRACT

Comparison of Er:YAG Modalities (PIPS-SWEEPS) on Eliminating of Enterococcus Faecalis Populations

Background: To determine the effectiveness of novel two Er:YAG lasers modalities, photon-initiated photoacoustic streaming (PIPS\SSP) and shock wave-enhanced emission photoacoustic streaming (SWEEPS\AutoSWEEPS) in terms of removal Enterococcus faecalis (E. faecalis) with or without antimicrobial agents [Sodium hypochlorite (NaOCl) and chlorhexidine gluconate (CHX)].

Methods: Forty-two extracted single-rooted mandibular premolars were prepared, sterilized, and then inoculated with E. faecalis for 4 weeks, and randomly divided into to two main and three subgroups (n= 7). Group Ia (PIPS + distilled water), Group Ib (PIPS + 5% NaOCl), Group Ic (PIPS + CHX), and Group IIa (SWEEPS + distilled water), Group IIb (SWEEPS + 5% NaOCl), and Group IIc (SWEEPS + CHX). Following incubation for 48 hours at 37°C, the colony forming units (CFU) were counted.

Results: NaOCl and CHX activated with PIPS\SSP or SWEEPS\AutoSWEEPS modalities showed significantly higher reduction rates than distilled water activated with both laser-activated methods ($p<0.05$), but no significant differences were detected between the NaOCl and CHX groups. Besides, in all groups, no significant difference was detected between PIPS\SSP and SWEEPS\AutoSWEEPS performances in the reduction of CFU counts ($p>0.05$).

Conclusion: Even novel laser activation methods or modalities are not sufficient alone to adequately reduce bacterial load and using antimicrobial agents with laser activation is necessary for the best reduction for microbial elimination. Novel SWEEPS\AutoSWEEPS modalıty with the conical 600µm tip showed no increased efficacy compared with PIPS\SSP mode. Besides, smaller fiber tip may increase the success of applications for minimally invasive access cavities and preparation.

KEYWORDS

Disinfection, Endodontics, Er:YAG laser, PIPS SWEEPS

ÖZ

Enterococcus Faecalis Biyofilmlerinin Ortadan Kaldırılmasında Er: YAG Yöntemlerinin (PIPS-SWEEPS) Karşılaştırılması

Amaç: İki yeni Er: YAG lazer modalitesi olan foton-indüklü fotoakustik dalgalanma (PIPS \ SSP) ve şok dalgası ile geliştirilmiş emisyon fotoakustik akımının (SWEEPS \ AutoSWEEP) antimikrobiyal ajanlar (NaOCl ve klorheksidin glukonat) kullanılarak veya kullanılmadan E. faecalis uzaklaştırmadaki etkinliğinin belirlenmesidir.

Gereç ve Yöntemler: Kırk iki ekstrakte tek köklü mandibular premolar hazırlandı, sterilize edildi ve daha sonra 4 hafta boyunca E. faecalis ekimi yapıldı ve rastgele iki ana ve üç alt gruba (n = 7) ayrıldı. Grup Ia (PIPS + distile su), Grup Ib (PIPS +% 5 NaOCl), Grup Ic (PIPS + CHX) ve Grup IIa (SWEEPS +distile su), Grup IIb (SWEEPS +% 5 NaOCl) ve Grup IIc (SWEEPS + CHX). 37 ° C'de 48 saat süreyle inkübasyonun ardından, oluşan koloniler (CFU) sayıldı.

Bulgular: PIPS \ SSP veya SWEEPS \ AutoSWEEPS yöntemleri ile aktive edilen NaOCl ve CHX, her iki lazer aktivasyon metodu ile aktive edilen distile sudan anlamlı şekilde daha yüksek indirgeme oranları gösterdi ($p <0.05$), ancak NaOCl ve CHX grupları arasında anlamlı bir fark bulunmadı. Ayrıca, tüm gruplarda, CFU sayılarının azaltılmasında PIPS \ SSP ve SWEEPS \ AutoSWEEPS performansları arasında anlamlı bir fark saptanmamıştır ($p> 0.05$).

Sonuç: Yeni lazer aktivasyon metod veya modülleri bile bakteri yükünü yeterince azaltmak için tek başına yeterli değildir. En etkili mikrobiyal eliminasyon için lazer aktivasyonun antimikrobiyal ajanlar ile birlikte kullanılması gerekmektedir. Konik 600µm uçla uygulanan yeni SWEEPS \ AutoSWEEPS modalitesi, PIPS \ SSP'e kıyasla artan bir etkinlik göstermedi. Daha küçük fiber uç kullanımı, minimal invaziv giriş kavitesi ve preperasyonlarda uygulamaların başarısını artırabilir.

ANAHTAR KELİMELER

Dezenfeksiyon, Endodonti, Er: YAG lazer, PIPS, SWEEPS

The reduction of bacteria and toxins from the root canal is a key factor in achieving long-term success in endodontics.[1] Due to the complex morphology of the root canal, it is almost impossible to eliminate the organization of intracanal bacteria with only mechanical preparation.[2] Enterococcus faecalis (E. faecalis) among other bacterial species is being considered as a bacterium that is frequently involved in the etiology of disease after endodontic treatment.[3] It can adhere to dentin and invade many antimicrobial agents and conditions.[4]

Sodium hypochlorite (NaOCl) is the most preferred chemical agent during root canal treatment owing to its ability to kill bacteria, destroy biofilms, and dissolve vital and necrotic tissue.[5,6] However, owing to high surface tension, NaOCl penetration is limited to about 130 micrometers into dentin tubules,

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whereas bacteria can colonize dentin tubules as deeply as 1100 μm from the canal lumen.[7] Chlorhexidine gluconate is an alternative to NaOCl that has antibacterial effects, broad-spectrum substantivity, and less toxicity.[8]

It was shown that conventional chemo-mechanical techniques have a limited ability to effectively reach all regions of root canal architecture.[9] To overcome these limitations, different agitation techniques have been proposed.[10] Laser energy is one of the best irrigation agitation techniques for the disinfection of root canals. It allows for penetration of dentinal tissues and access to unreachable areas in the root canal system. Photon-induced photoacoustic streaming (PIPS) in SSP (Super Short Pulse; 50 μs) mode is a popular Er:YAG laser technology that uses a very low power to rapidly pulse laser light energy using a specially designed radial tip.[11] However, especially in narrow root canals or ramifications, it is very difficult because the friction on the canal walls decelerates the cavitation and the solution cannot move fast enough.[12] Thereby, to overcome this difficulty, an innovative Er:YAG laser called shock wave-enhanced emission photoacoustic streaming (SWEEPS) method has been introduced. It is applied with an Er:YAG laser, using a new tip design and unique mode for higher irrigation activation efficacy. Unlike PIPS, it uses ultra short laser pulse pairs (25 μs) for irrigant activation. The second pulse creates a bubble that causes pressure on the first bubble and accelerates its collapse. Thus, a large number of shock waves occurs that can reach to the irregularities of the root canal and consequently increase the cleaning efficiency of laser agitation. It is claimed that this mechanism leads to a large number of shock waves that reach the irregularities of the root canal and consequently increase the cleaning efficiency of laser agitation compared to SSP modality.[12] The oscillation time of the vapor bubble depends on the same parameters such as laser pulse energy, root canal geometries, and access cavity design. In the SWEEPS modalıty, the optimal time delay of the pulse pair cannot be determined by the clinician. So, to overcome this limitation, a recent AutoSWEEPS modalıty has been developed that automatically created a time delay between 300 and 600 μs. Thus, ıt has been aimed to provide providing optimum time delay during each sweep, providing supreme irrigation efficacy.

To the best of our knowledge, there have been limited studies about SWEEPS modality in simulated root canals. But this cannot imitate the real dentine wall resistance and root canal conditions. However there has been no study investigating the antibacterial efficacy. So, current study aimed to research the effectiveness of two popular Er:YAG laser methods\modalities, PIPS-SSP and SWEEPS-AutoSWEEPS, in the elimination of E. faecalis with or without antimicrobial agents (NaOCl and of chlorhexidine gluconate) in real human root canal. The null hypothesis was that the irrigation activated with SWEEPS-AutoSWEEPS modality would not result in better disinfection than the PIPS-SSP groups.

MATERIALS AND METHOD

Sample collection

The procedures used in this study conformed to the protocols approved by the Ethical Review Committee under the Research Foundation at the Medical Faculty of Recep Tayyip Erdogan University. (No: 2019\116). G Power 3.0.10 (University Kiel, Germany) software was used to calculate the effect size. Effect size was determined based on the results of a previous study 13, an effect size of 0.55 d cohen was found as sufficient for significance. With 80% power, 0.05 type 1 error, at least 42 samples were for required. Fifty- five freshly extracted intact single-rooted mandibular premolars were collected for this study. The samples were stored in 0.1% thymol solution at 4°C until required for use. The external root surfaces were debrided using a hand scaler. The teeth were decoronated to obtain a standard length of 19 mm for each root. A deep of 4 mm reservoir and an access cavity with 3mm of diameter was created with a diamond bur under copious irrigation. Working length was established by subtracting 1 mm from this length. All samples were prepared by the same operator using the crown-down method with ProTaper Ni-Ti rotary files (Dentsply Maillefer, Ballaigues, Switzerland) up to F4 (40 size, 06 taper). During instrumentation, 1 mL 5% NaOCl (ImIcryl, Karatay, Konya) was used as a lubricant. After preparations, the roots were flushed using 1 mL 17% EDTA for 1 min to remove the smear layer, which was followed by needle irrigation with 1 mL 5% NaOCl for 1 min.[14] Before the NaOCl irrigation, the canals were flushed with 2 mL of distilled water. Finally, the canals were flushed with 1 mL of distilled water and dried with paper points. The root ends of the teeth were covered with cyanoacrylate, and embedded in a silicone mold. Then, samples were placed in autoclavable plastic carrier containers and sterilized in an autoclave at 121°C under 1 atm pressure for 15 minutes.[15]

E. faecalis culture and inoculation

An E. faecalis ATCC 29212 vial was removed from stock at -80°C, thawed at room temperature, and seeded into Mueller-Hinton agar (MHA). When the

reproduction occurred, the colony appearance and staining characteristics were evaluated, and one colony that was determined to represent the bacteria in stock was transferred to 2 mL trypticase soy broth (TSB). The turbidity was incubated until McFarland 0.5. One milliliter from each turbidity suspension was transferred to a labeled sterile vial and stored at -80°C.

One day after the quality control of the bacterial stocks, one of the stock vials was removed and dissolved at room temperature. 0.9 mL of sterile phosphate-buffered saline was transferred to 8 sterile 1.5 mL centrifuge tubes. The stock was vortexed by placing 100 µL of the vial into the first tube and pipetting 100 µL to the next to reconstitute the bacterial stock solutions. Of these E. faecalis dilutions, 10 µL of common cultivation was conducted to MHA, and Petri dishes were incubated. After growth, the amount and purity of the bacteria in stock in CFU/mL were evaluated.

Just before the experiment, one of the stock bacterial vials at -80°C was removed and dissolved at room temperature. After vortexing, 1 mL of this solution was pipetted into 2 mL of TSB and incubated until the turbidity was McFarland 0.5.

To infect root canals with E. faecalis, freshly prepared bacterial suspension with turbidity McFarland 0.5 was transferred into the root canals using a 10 µL automated pipette and allowed to reach the apical region via a sterile 30-gauge irrigation needle (Kerr; USA). To prevent the teeth from drying out, moistened cotton was placed in carrier containers and incubated for 48 hours in an oven at 37°C with their lids closed. This procedure was continued every other day for 4 weeks.

After four weeks of planting, in ten samples proliferation was not observed. Therefore, these samples were excluded and totally the sample size was standardized at forty-two. Enough sample the specimens were selected for each group by a blinded examiner and randomly divided two main experimental groups (with three subgroups in each).

Group I: PIPS\SSP

In this group, to activated irrigation, Er:YAG laser device (Fotona LightWalker DT Ljubljana, Slovenia) with a wavelength of 2940 nm was used via H14-N handpiece with a 14-mm long and 300-µm specially designed tapered and stripped fiber tip (PIPS 300\14, Fotona). The power was set to 0.3W, 15 Hz, and 20 mJ at single laser pulse duration mode (SSP) (50 µs) without air or water. A 27-gauge open-ended needle was positioned 1 mm short from the WL and the 2 mm of the fiber tip was positioned in the center of the reservoir and fixed in that position. The root canals were irrigated continuously with the irrigation agents mentioned below for 3 × 20 s with a resting time of 30 s between each cycle. During laser activation, the tip was submerged in irrigant that was continuously applied with the needle. To prevent the microorganism transfer between the samples, the tip was kept in 5.25 % NaOCl for 1 min before each use.8 The total activation time was 60 s for each irrigation solution. And the total volume of irrigation solution each was 6 mL (2mL×3) . After irrigation procedures were completed, the canals were washed using 5 mL 2 mL sodium thiosulfate for 30 s to neutralize the irrigants.

Ia: 2 mL of Distilled Water (DW)

Ib: 2 mL of 5% NaOCl

Ic: 2 mL of 2% CHX

Group II: SWEEPS\AutoSWEEPS

In this experimental group, for laser-activated procedure, the same Er:YAG laser device and handpiece were used with an 8.5-mm long and conical fiber tip with a 600-μm diameter (SWEEPS 600, Fotona). For standardization, the laser device was set at the same parameters as in the PIPS groups (0.3 W, 20 mJ, 15 Hz) and in AutoSWEEPS mode which has two-ultrashort micro pulses (25 µs pulse duration), separated by a continuously varied delay from 300 to 600 µs sec with 10 µs step 16 was chosen. A 27-gauge open-ended needle was positioned 1 mm short of the WL and the 2 mm of the fiber tip was positioned in the center of the reservoir and fixed in that position. The root canals were irrigated continuously as mentioned in the previous experimental group.

IIa: 2 mL of DW

IIb: 2 mL of 5% NaOCl

IIc: 2 mL of 2% CHX

After irrigation procedures were completed, the canals were washed using 5 mL 2M sodium thiosulfate for 30 s to neutralize the irrigants.

Microbial counts

After the irrigation procedure, the root canals were filled with sterile PBS to collect S2 microbial samples. Sterile H type files 15 were used to allow the possible surviving bacteria in the channel walls to pass into the solution. Three consecutive paper points were placed in the root canal and allowed to wait for 60 s. Then the paper samples were transferred to a sterile 1.5 mL Eppendorf tube filled with PBS.

The Eppendorf tubes with paper points were vortexed and the bacteria were transferred to the liquid medium. One hundred microliters of these tubes were pipetted into sterile 1.5 mL centrifuge

tubes containing 0.9 mL PBS (10:1 dilution tube), and the sample group and number of interest was recorded. Ten microliters from the stock sample tube and 10-1 dilution tube were pipetted onto MHA media and spread. After incubation for 48 hours at 37°C, the colonies formed were counted and the number obtained was multiplied by 100 to determine the number of bacteria collected from the root canal.

Statistical analysis

The Jamovi (Version 1.0.4) [ComputerSoftware] (accessed by https://www.jamovi.org) program was used for statistical analysis. The normality of data distribution was checked using the Shapiro-Wilk test. Repeated measures analysis of variance (ANOVA) was used to compare the disinfection rates of each group. Statistical significance was considered as p=0.05.

RESULTS

The initial CFU counts (T1) were significantly higher in all groups than the final CFU counts (T2) (p<0.001) (Table 1).

Table 1.

Presentation of time factor dependent Post Hoc comparisons

Time	Time	Mean Difference	t	p value
T1	T2	123005	17.7	< 0.001*

** indicated statistical significance (p <0.05)*

No significant differences were found between NaOCl and CHX, however, both of them was significantly more effective than DW (p<0.001) (Table 2).

Table 2.

Presentation of Irrigation agents dependent Post Hoc comparisons

Irrigation agents	Irrigation agents	Mean Difference	t	p value
NaOCl	CHX	2793	0.195	0.979
NaOCl	DW	-69839	-4872	< 0.001*
CHX	DW	-72632	-5067	< 0.001*

** indicated statistical significance (p <0.05)*

In addition, in all groups, no significant difference was detected between PIPS\SSP and SWEEPS\AutoSWEEPS performances in the reduction of CFU counts (p>0.05) (Table 3).

Table 3.

Presentation of Techniques dependent Post Hoc comparisons

Techniques	Techniques	Mean Difference	t	p value
PIPS	SWEEPS	7581	0.648	0.521

** indicated statistical significance (p <0.05)*

Among the groups, significantly fewer CFU counts were detected in the samples irrigated with NaOCl and CHX activated with PIPS\SSP or SWEEPS\AutoSWEEPS than in DW-activated PIPS\SSP or SWEEPS\AutoSWEEPS (p<0.05) (Table 4).

Table 4.

Presentation of Irrigation agents * Techniques dependent Post Hoc comparisons

Irrigation agents-Techniques	Irrigation agents-Techniques	Mean Difference	t	p value
NaOCl - PIPS	NaOCl - SWEEPS	12293	0.6064	0.990
NaOCl - PIPS	CHX - PIPS	4607	0.2273	0.999
NaOCl - PIPS	CHX - SWEEPS	13271	0.6547	0.986
NaOCl - PIPS	DW - PIPS	-64586	-0.1861	0.033*
NaOCl - PIPS	DW - SWEEPS	-62800	-0.0980	0.040*
NaOCl - SWEEPS	CHX - PIPS	-7686	-0.3791	0.999
NaOCl - SWEEPS	CHX - SWEEPS	979	0.0483	0.999
NaOCl - SWEEPS	DW - PIPS	76879	-0.7925	0.007*
NaOCl - SWEEPS	DW - SWEEPS	75093	-37044	0.009*
CHX - PIPS	CHX - SWEEPS	8664	0.4274	0.998
CHX - PIPS	DW - PIPS	-69193	-34133	0.018*
CHX - PIPS	DW - SWEEPS	-67407	-33252	0.023
CHX - SWEEPS	DW - PIPS	-77857	-38408	0.006*
CHX - SWEEPS	DW - SWEEPS	-76071	-37527	0.008*
DW - PIPS	DW - SWEEPS	1786	0.0881	0.999

** indicated statistical significance (p <0.05)*

DISCUSSION

E. faecalis is a microorganism commonly used in antimicrobial efficacy studies in endodontics. It can survive in a hard environment and is frequently isolated from root canals after treatment because of its virulence factors and high resistance to many antimicrobial agents.[17] NaOCl is the most preferred irrigant because of its high alkalinity and powerful antimicrobial action. Also, it exerts its antibacterial effect by inducing the irreversible oxidation of the sulfhydryl groups of essential bacterial enzymes, resulting in disulfide linkages, with the consequent disruption of the metabolic functions of bacterial cells.[18] It also has deleterious effects on bacterial DNA, which involve the formation of chlorinated derivatives of nucleotide bases.[19] CHX is another antimicrobial agent that has been recommended for clinical use because of its good antibacterial action and substantivity, as well as its biocompatibility. It has proven bacteriostatic or bactericidal effects owing to the precipitation and/or coagulation of the cytoplasm.[20]

Lasers have been introduced as a powerful activator for root canal irrigation to increase debridement and disinfection. It has been shown that Er:YAG laser-activated irrigations were effective in removing E. faecalis population in an in vitro root canal model.[21] The mechanism is based on the evaporation effect of cellular water during the laser pulse and leads to the disruption of the cell walls of microorganisms.[22] Previously, it was reported Er:YAG laser combined

with NaOCl showed a superior bactericidal effect to NaOCl alone.[23] Er:YAG laser kills bacteria by heating the environment to above lethal values and local heating inside bacteria.[24] When the NaOCl is agitated using laser, vapor bubble expansion and implosion occurs and this creates a large stress on the dentin wall, resulting in it being pushed deeper into the dentin tubules to perform its antimicrobial effect.[25]

PIPS used with Er:YAG aims to activate the irrigation solutions at lower energy levels without thermal damage on the root canal walls. The innovative Er:YAG modality, SWEEPS, was developed to improve the cleaning efficacy of PIPS and to overcome the difficulties in narrow root canals.[26] In both modalities, there is no need to negotiate the tip close to the apex.

In the literature, there are same laboratory study about SWEEPS\AutoSWEEPS modalities, but there no study has been published about the antimicrobial efficacy of nowel modalities in the real root canal conditions. As known, the laboratory set-up, cannot reflect the complexity of a natural root canal anatomy and clinical reality. So, the purpose of this in vitro study was to evaluate two recent Er:YAG laser technologies (SWEEPS\AutoSWEEPS) and (PIPS\SSP) on the effectiveness of E. Fecalis populations with different antimicrobial agents. Thus, it was aimed to determine the best irrigation procedure that destroys the E. faecalis and to increase endodontic treatment success. According to the results of the study, SWEEPS-AutoSWEEPS modality did not show better performance than PIPS\SSP mode, and even these novel laser modalities were not sufficient alone without antimicrobial solutions.

In a study, researchers compared the penetration depths of endodontic irrigants into dentinal tubules using several activation methods including SWEEPS and PIPS. The authors concluded that although PIPS showed deeper penetration, SWEEPS did not increase the penetration depths.[27] Also, they argued that the change in pulse and formation of secondary bubbles could trigger counter-currents that inhibit irrigant flow. However, there are as yet insufficient studies and no consensus regarding SWEEPS. In another study, the cleaning efficacy and pressure measurements of SSP and SWEEPS modalities were evaluated using different tips and geometries in a laboratory setup. They reported that pressure generated and penetration of methylene blue is much larger when using a smaller diameter fiber tip (such as used in the PIPS group) compared to a larger diameter fiber tip (such as used in the SWEEPS group).[28]

A methodological disadvantage in this study is that there is a significant difference between pulse modalities (SSP\Automatic SWEEPS) and fiber tip design. SWEEPS has an 8.5-mm-long and conical tip with a 600-μm diameter. Therefore, the insufficient effect of SWEEPS in this study may be due to both a different laser pulse modality and thick conical fiber end geometries used. And this can demonstrate the inferiority of the conical 600μm tip. Besides this, during the procedure, a large amount of lateral transmission to the outside was observed. This may be the reason that the laser pulse cannot reach into the root canal totally and decreases the efficacy. It may be beneficial to use modified thin and resistant fiber tips for SWEEPS applications.

Regarding the results of this study, the significant difference between DW and NaOCl and CHX confirms the additive importance of antimicrobial agents including chlorine in achieving better disinfection. The use of antimicrobial agents was required for microbial elimination; even novel laser activation methods are not sufficient alone to adequately reduce bacterial load.

The similar efficacy between NaOCl and CHX may be based on the chlorine contents. Chlorine is a strong oxidant that promotes the irreversible oxidation of sulfhydryl groups on bacterial essential enzymes.[29] With laser irradiation, a strong modulation occurs, resulting in a significant increase in the amount of free chlorine and contact with microorganisms.[30] Consistent with these findings, some studies have reported high bactericidal effects when sodium hypochlorite or chlorhexidine were activated by an Er:YAG laser.[31] By contrast, some researchers reported questionable findings. DiVito et al.[32] reported laser-activated irrigation (LAI) using saline improved smear layer removal. Pedulla et al.[33] and Seet et al.[34] reported the antibacterial effect of saline activated with LAI. They attributed this improved performance to the intense streaming and flushing action created within the irrigant. However, the authors impressed that the laser-activated saline had a lower antibacterial effect compared with conventional needle irrigation with NaOCl.

Microbial reduction also depends on the test strains and biofilm model, initial inoculation levels, and subsequent biofilm formation conditions. Also, the difficulty of detecting differences in an infection model might have been related to the insufficient sensitivity of the methodology used for detecting possible viable bacterial cells at lower concentrations; there may be need for a larger sample size to increase confidence.[35]

Numerous methods have been proposed for the collection of microbial samples. Previously, it has been reported that the culture method is sensitive enough to detect E. faecalis in root canals.[36] Paper cone cultures are used more widely compared with the removal of dentin shavings with files or reamers.[37] However, with paper cones, as used in this study, only the microorganisms located on the root canal wall can be removed, microorganisms

that have penetrated dentin tubules cannot be taken.38 To overcome this situation, to ensure that the microorganisms in the tubules fall into the solution, we first tried slight circumferential filing using a sterile No. 15 H-type file on the walls. Then, microbial samples were collected using paper cones in the root canals for one minute. In this study, to ensure sufficient microbial layer, E. faecalis (ATCC 29212) cultivation was performed in root canals for 4 weeks.

On the other hand, one of the limitations of this study is, the culture techniques do not fully reflect clinical conditions, and only show the number of specimens in which no bacteria were recovered; quantification of the surviving bacteria is not possible. The differences in reporting bacterial viability may play a crucial role in the evaluation and comparison of studies. Therefore, the findings should be interpreted with caution. It is required to identify microorganisms that survive after irrigation procedures using advanced technology. Another lack of study is the lasers wavelengths impact on the root walls were not monitored using scanning electron microscopy images (SEM or, even better, ESEM). And the conduction of the thermal measurements on the root surface in the apical region to estimate the thermal impacts on the periapical tissues are not evaluated.

CONCLUSION

The null hypothesis was accepted, irrigation activated with SWEEPS-AutoSWEEPS modality did not result in better disinfection than the PIPS-SSP groups. Within the limitations of the present in vitro study, it can be concluded that the use of antimicrobial agents is required for microbial elimination; even novel laser activation methods or modalities are not sufficient alone to adequately reduce bacterial load. Besides, novel SWEEPS\AutoSWEEPS modality with the conical 600 μm tip showed no increased efficacy compared with PIPS\SSP mode. So, it was supported by this study that the conical 600μm tip is not appropriate for effective debridement and smaller SWEEPS fiber tip may increase the success of applications for minimally invasive access cavities and preparation. Further research is needed to focus on the SWEEPS\AutoSweeps modality and equipment's role in irrigation, disinfection, and other endodontics applications under clinical conditions.

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