Recent advances in root canal disinfection

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Abstract
Contemporary endodontics has seen an unprecedented advance in technology and materials. This review is aimed to bring some light over the advances in root canal disinfection. Jointly, these advances are aimed at improving the state of the art and science of root canal treatment.

Keywords: Disinfection, root canal, advancement

Introduction
Complex root anatomy makes endodontic treatment challenging, but innovative technologies have simplified canal cleaning and disinfection, improved efficiency, and increased clinical success. The modalities described here are designed to help clinicians effectively irrigate and chemically debride vital and necrotic pulp tissue, microorganisms and biofilm from the entire root canal system [1].

The past couple of decades have witnessed one of the most rapid and extensive technological evolutions in dentistry. This period has presented some remarkable developments of endodontic technologies.

Disinfection Devices and Techniques
Syringe Delivery
Application of an irritant into a canal by means of a syringe and needle allows exact placement, replenishing of existing fluid, rinsing out of larger debris particles, as well as allowing direct contact to microorganisms in areas close to the needle tip.

In passive syringe irrigation, the actual exchange of irrigant is restricted to 1 to 1.5 mm apical to the needle tip, with fluid dynamics taking place near the needle outlet. Volume and speed of fluid flow are proportional to the cleansing efficiency inside a root canal. Therefore, both the diameter and position of the needle outlet determine successful chemo mechanical debridement; placement close to working length is required to guarantee fluid exchange at the apical portion of the canal, but close control is required to avoid extrusion. Therefore, the choice of an appropriate irrigating needle is important. Although larger-gauge needles allows a quicker and larger amount of fluid exchange, the wider diameter does not allow cleaning of the apical and narrower areas of the root canal system. Excess pressure or binding of needles into canals during irrigation with no possibility of backflow of the irrigant should be avoided under all circumstances to prevent extrusion into periapical spaces. In immature teeth with wide apical foramina or when the apical constriction no longer exists, special care must be taken to prevent irrigation extrusion and potential accidents. There are different sizes and types of irrigation needles. The size of the irrigation needle should be chosen depending on the canal size and taper. Most root canals that have not been instrumented are too narrow to be reached effectively by disinfectants, even when fine irrigation needles are used [3-5].

Therefore, effective cleaning of the root canal must include intermittent agitation of the canal content with a small instrument to prevent debris from accumulating at the apical portion of the root canal. Preparation size and taper ultimately determine how close a needle can be placed to the final apical millimeters of a root canal. Open-ended needles are recommended over the end open needles to prevent extrusion of the irrigant. Some needles and suction tips may be attached to the air/water syringe to increase
both the speed of irrigant flow and the volume of irrigant. Examples include the Stropko Irrigator (Vista Dental Products), which is an adapter that connects to the air/water syringe and accepts standard Luer-lock needle tips for irrigant removal and application as well as air-drying. Clinical effect is a rapid relief of pain associated with acute inflammatory conditions of the pulp and periodontium.

Manually Activated Irrigation
Liquid placed inside the root canal more effectively reaches crevices and mechanically untouched areas if it is agitated inside the root canal. Corono-apical movements of the irrigation needle, stirring movements with small endodontic instruments, and manual push-pull movements using a fitted master gutta-percha cone have been recommended.

Sonomatically Activated Irrigation
The EndoActivator System uses safe, noncutting polymer tips in an easy-to-use subsonic hand piece to quickly and vigorously agitate irrigant solutions during endodontic therapy. When comparing sonic with ultrasonic irrigation, the results can be controversial. The majority of the studies benefit ultrasonic irrigation. The difference lies in the oscillating movements: sonic devices range between 1500 Hz and 6000 Hz, and ultrasonic equipment requires vibrations greater than 20,000 Hz. If sonic devices are left in the canal for longer periods of time, better cleaning effects can be found. Sonic or ultrasonic irrigation may be carried out with activated smooth wires or plastic inserts, endodontic instruments, or activated irrigation needles. Examples include EndoSonor (Dentsply Maillefer) and EndoSoft ESI (EMS Electro Medical Systems, Nyon, Switzerland) inserts, IrrisSafe (Acteon Satelec), the EndoActivator System (Dentsply Tulsa Dental Specialties), and the Vibiringe sonic syringe (Vibringe B.V., Amsterdam, Netherlands) [6-10].

Passive Ultrasonic Irrigation
Richman was the first to introduce ultrasonic devices in endodontics. The files are driven to oscillate at ultrasonic frequencies of 25 to 30 kHz to mechanically prepare the root canal walls. It has been shown that ultrasonically driven files are effective to activate the irrigation liquids inside the root canal system by inducing acoustic streaming and cavitation. Two types of ultrasonic irrigation have been described in the literature:

1. Where irrigation is combined with simultaneous ultrasonic instrumentation (UI)
2. Without simultaneous instrumentation, called passive ultrasonic irrigation (PUI).

During UI, the file is intentionally brought into contact with the root canal wall. But because of the complex canal anatomy, the UI will never contact the entire wall and it may result in uncontrolled cutting of the root canal walls without effective disinfection.

Passive ultrasonic irrigation (PUI) was first described by Weller and colleague. The term passive is related to the noncutting action of the ultrasonically activated file. PUI relies on the transmission of acoustic energy from an oscillating file or smooth wire to an irrigant in the root canal. PUI should be introduced in the canal once that the root canal system has a final apical size and taper. A fresh solution of irrigant should be introduced and a small file or smooth wire (for example, size #15) is ultrasonically activated. Because the root canal has already been shaped, the file or wire can move more freely, and the irrigant can penetrate into the apical part of the root canal system, with the cleaning effect being more significant. Using this noncutting methodology, the potential to create aberrant shapes within the root canal are reduced to a minimum. A file larger than a #15 or #20 will require a wide root canal to reduce oscillation dampening by wall contact.

Ultrasonic activation of the irrigant can be intermittent or continue. The ProUltra PiezoFlow (Dentsply Tulsa Dental Specialties) has been introduced to irrigate and activate the liquids at the same time. The device consists mainly of an ultrasonically energized needle connected to a reservoir of sodium hypochlorite (NaOCl). This continuous ultrasonic irrigation (CUI) system allows simultaneous continuous irrigant delivery and ultrasonic activation; unlike PUI. Manually Activated Irrigation Liquid placed inside the root canal more effectively reaches crevices and mechanically untouched areas if it is agitated inside the root canal. Corona-apical movements of the irrigation needle stirring movements with small endodontic instruments, and manual push-pull movements using a fitted master gutta-percha cone have been recommended [6-10]. Other than conventional irrigation, additional techniques for endodontic disinfection have been proposed and tested, including laser systems and gaseous ozone. Several new devices for endodontic irrigation or disinfection have been introduced, among which are -- EndoActivator System (Dentsply Tulsa Dental Specialties),

- Passive ultrasonic irrigation,
- EndoVac
- Safety-Irrigator (Vista Dental Products, Racine, WI),
- Self-adjusting File
- Photoactivation disinfection,
- Ozone.

These new devices use pressure, vacuum, oscillation, or a combination with suction.

Negative Apical Pressure
Another approach to afford better access of irrigation solution to the apical portion of the canal is so-called negative-pressure irrigation. Here, irrigant is delivered into the access chamber, and a very fine needle connected to the dental unit’s suction device is placed into the root canal. Excess irrigant from the access cavity is then transported apically and ultimately removed via suction. First, a macrocannula, equivalent to an ISO size #55, .02 taper instrument, removes coronal debris. Subsequently, a microcannula, equivalent to a size #32, .02 taper, removes particles lodged close to working length. Such a system is commercially available (EndoVac, Discus Dental) and may prove a valuable adjunct in canal disinfection.

One of the main characteristics of the system is the safety. Many studies proved that EndoVac will not extrude irrigation solution through the apex. On the other hand, because the irrigation is deposited in the coronal area, the irrigant flow in the apical last millimeters of the canal is very passive, and some concerns were expressed that flow is laminar and passive in the apical region.

In one study, the apical negative pressure mode of irrigation generated the lowest wall shear stress. Another device that makes use of pressure-suction technology is the Rinscape (Dürr Dental, Bietigheim-Bissingen, Germany). It aspirates the delivered rinsing solution into an irrigation needle that is placed close to working length and at the same
time activates the needle with oscillations of 1.6 Hz amplitude [11].

Safety-Irrigator
The Safety-Irrigator (Vista Dental Products) is an irrigation/evacuation system that apically delivers the irrigant under positive pressure through a thin needle containing a lateral opening and evacuates the solution through a large needle at the root canal orifice. The Safety-Irrigator features a large coronal evacuation tube, enabling the user to safely irrigate and evacuate simultaneously. It fits any standard Luerlock syringe. Designed to limit risk of NaOCl accidents, this “negative-pressure” irrigation device comes fully assembled and fitted with a side-vented irrigating needle for added safety.

Gentle Wave System / No instrumentalation
Sonendo Inc. develops a so-called multisonic cleaning technology (Gentle Wave) that only requires pulp chamber access. This system is no instrumentalation and being tested. The first trial of a method of cleaning without canal preparation was the Non-Instrumentation Technique (NIT) conceived by Lussi et al. This technique did not provide for the enlargement of the root canals because there was no mechanical instrumentation of root canal walls. In fact, root canal cleaning was obtained exclusively with hypochlorite at low concentration, introduced and removed from the canal by a vacuum pump and an electric piston that created fields of alternating pressure inside the canal. This caused the implosion of the produced bubbles and hydrodynamic turbulence that facilitated the penetration of hypochlorite into the root canal ramifications. A method for cleaning the entire root canal system has recently been developed using a broad spectrum of sound waves transmitted within an irrigating solution to quickly remove pulp tissue, debris and microorganisms. One study showed that this technique is able to dissolve the tested tissues at a significantly higher rate compared to conventional irrigation. From a biological point of view, endodontic therapy must be directed to the elimination of microorganisms and prevention of a possible reinfection. Unfortunately, the root canal system with its anatomical complexity is a challenging environment for the effective removal of bacteria and biofilm adhered to the canal walls. The chemo-mechanical preparation involves mechanical instrumentation and antibacterial irrigation and it is the most important phase for disinfection of the endodontic space. The technological advances of instruments brought significant improvements in the ability to shape the root canals, with less procedural complications. Various antimicrobial agents have been employed in the management of infected root canal systems. Furthermore, some clinical technical procedures - such as the increase in apical preparation and a more effective system of irrigation delivery and activation of irrigant - can promote and make more predictable the reduction of intracanal bacteria, especially in complex anatomical and not instrumented areas of the root canal system.

Laser-Activated Irrigation
Lasers are widely used in dentistry and include diode, Nd:YAG, erbium, and CO2, which produces radiation in both the near and far-infrared electromagnetic spectrum. Laser devices have been proposed to improve the efficacy of irrigants. Lasers have been studied for their ability to clean and effectively disinfect root canals. The Er:YAG laser wavelength (2940 nm) has the highest absorption in water and a high affinity to hydroxyapatite, which makes it suitable for use in root canal treatment. Laser energy may be used to activate irrigant solutions in different ways-for example, at a molecular level, as in photoactivated disinfection (PAD), or at a bulk flow level, as in laser-activated irrigation (LAI) [13-19].

Laser-activated irrigation (LAI).
Several studies in vivo and ex vivo have indicated that laser activated irrigation is promising for removing smear layer and dentin debris in less time than PUI. The mechanism of action is based on the generation of a secondary cavitation effect with expansion and successive implosion of fluids. These results are in agreement with data related to a new erbium laser technique that used a photon-induced photoacoustic streaming (PIPS) of irrigants. In that technique, the laser tip is placed into the coronal access opening of the pulp chamber only and is kept stationary without advancing into the orifice of the canal. The use of a newly designed tapered and stripped tip with specific minimally ablative laser settings is required, resulting in low energy (20 mJ), a pulse repetition rate of 15 Hz, and a very short pulse duration (50 µs). The difference in laser penetration and bacterial killing is attributed to the difference in the degree of absorption of different wavelengths of light within the dentin. Endodontic pathogens that grow as biofilms, however, are difficult to eradicate even upon direct laser exposure. Photoactivation Disinfection Photodynamic therapy (PDT) or light-activated therapy (LAT) may have endodontic applications because of its antimicrobial effectiveness. In principle, antimicrobial photodynamic therapy (APDT) is a two-step procedure that involves the

STEP 1: Introduction of a photosensitizer (photosensitization of the infected tissue)
STEP 2: Light illumination (Irradiation of the photosensitized tissue) of the sensitized tissue, which would generate a toxic photochemistry on the target cell, leading to cell lysis.

Each of these elements used independently will not have any action, but together they have a synergism effect to produce antibacterial action. Indeed, in vitro experiments showed promising results when used as an adjunct disinfected device.

Antibacterial Nanoparticles
Nanoparticles are microscopic particles with one or more dimensions in the range of 1 to 100 nm. Nanoparticles are recognized to have properties that are unique from their bulk or powder counterparts. Antibacterial nanoparticles have been found to have a broad spectrum of antimicrobial activity and a far lower propensity to induce microbial resistance than antibiotics. Such nanoparticles in endodontics are being studied in different ways, such as mixed with irrigants, photosensitizer, and sealers. Currently, the consensus is that the successful application of nanoparticles in endodontics will depend on both the effectiveness of antimicrobial nanoparticles and the delivery method used to disperse these particles into the anatomic complexities of the root canal system.

Chitosan Nanoparticles
Chitosan, a deacetylated derivative of chitin, is the second most abundant natural biopolymer. Nanoparticles of chitosan could be synthesized or assembled using different methods depending on the end application or physical characteristics.
required in nanoparticles. Chitosan has received significant interest in biomedicine because of its versatility in various forms such as powder, capsules, films, scaffolds, hydrogels, beads and bandages. Nanoparticles of chitosan has been developed for its antibacterial and drug delivery application. They have excellent antibacterial, antiviral and antifungal properties. Gran positive bacteria are more susceptible. Chitosan nanoparticles by virtue of their charge and size are expected to posses enhanced antibacterial properties. Use of chitosan nanoparticles with chlorhexidine can eliminate *E. faecalis* with potential application towards tissue regeneration using membrane barrier in periapical surgery.

**Silver Nanoparticles**

Silver compounds and nanoparticles which are used in biomedicine, mainly because of their antibacterial property. In case of dental application, silver and its nanoparticles have been tested for application as endodontic retrograde filling material, dental restorative material, dental implants, and caries inhibitory solution. Silver is known to produce an antibacterial effect by acting on multiple targets starting from interaction with the sulfhydryl groups of proteins and DNA, alter the hydrogen bonding/respiratory chain, unwind DNA, and interfere with cell wall synthesis/cell division. Ag-NPs are known to further destabilize the bacterial membrane and increase permeability, leading to leakage of cell constituents. Ag-NPs with significant antibacterial activity could be used for root canal disinfection. However, the prolonged interaction time required by Ag-NPs for effective bacterial killing needs to be considered, and its use ideally should be limited to medicament rather than as an irrigant.

**Superoxidized Water /Electrochemically activated water**

Superoxidized water, also called electrochemically activated water or oxidative potential water, is effectively saline that has been electrolyzed to form superoxidized water, hypochlorous acid, and free chlorine radicals. It is commercially available as Sterilox (Sterilox Technologies, Radnor, PA). This solution is nontoxic to biologic tissues yet able to kill microorganisms. The solution is generated by electrolyzing saline solution, a process no different than that used in the commercial production of NaOCl. The difference, however, is that the solution accumulating at the anode is harvested as the anolyte and that at the cathode as the catholyte. These solutions display properties that are dependent on the strength of the initial saline solution, the applied potential difference, and the rate of generation. The technology that allows harvesting of the respective solutions resides in the design of the anode and the cathode and originates either in Russia (electrochemically activated water) or in Japan (oxidative potential water). Although the solutions bear different names, the principles in the manufacturing process appear to be similar. The use of superoxidized water is sparsely described in the endodontic literature but shows early promise. The solutions from both technologies have been tested for their ability to debride root canals, remove smear layer, and kill bacteria and bacterial spores. Results are favorable and show biocompatibility with vital systems.

**Bioactive Glass**

Recently, the bioactive glass or bioactive glass-ceramics have been the object of considerable interest for endodontic disinfection due to their antibacterial properties, but with conflicting results BAGs in micro-and nanoforms have been tested to improve root canal disinfection. *In vitro* root canal disinfection studies showed a significantly less antibacterial effect of BAG compared with calcium hydroxide in preventing residual bacterial growth. They killed planktonic bacteria significantly better compared with biofilm bacteria [4].

**Natural Plant Extracts**

A current trend directed to the use of natural plant extracts takes advantage of the antibacterial activity of polyphenolic molecules generally used for storing food. These compounds have a poor antibacterial efficacy, but a little significant ability to reduce the formation of biofilms, although the mechanism by which this occurs is not clear.

**Apple Vinegar**

The use of different chemical irrigants for smear layer removal during the root canal treatment has also been proposed, with EDTA, citric acid, maleic acid and apple vinegar offering the most interesting results. Apple vinegar has been indicated as an antiseptic agent due to its medicinal properties, with its use as an auxiliary solution in the chemomechanical preparation of root canals showing good results when compared to NaOCl and EDTA.

**Uncaria Tomentosa**

Commonly known as “cat’s claw” because of the small curved spines on the stem at the leaf juncture, it offers an anti-inflammatory, antiviral, antibacterial, antioxidant, and immunomodulating action. Its toxicity is low when used correctly, being an important advantage of medicinal plant treatments over more conventional methods. U. Tomentosa contains triterpenes, vegetal steroids and glycooides, these compounds may be related to its antimicrobial activity.

**Propolis and Zingiber officinale**

Propolis presents anti-inflammatory and antimicrobial actions. Duarte et al. showed its influence in the reduction of acid production by *S. mutans* in the dental biofilm, besides its inhibitory action over the F-ATPase activity of *S. mutans*.

**Castor Oil Detergent (Ricinus communis)**

Castor oil detergent has shown antimicrobial activity and biocompatibility, non-toxic results and detergent properties which are important requirements for an irrigant solution. Endodontic irrigation with castor oil extract is capable of removing debris, showing similar results to 1% NaOCl. Root canal irrigation with castor oil reduces the number of *Escherichia coli* and *E. faecalis* during biomechanical preparation.

**Green Tea Polyphenols (GTP):** GTP are derived from fresh leaves of tea (Camellia sinensis), an important component of traditional Japanese and Chinese cultures. They have shown significant antibacterial activity in *E. faecalis* biofilms grown on dental culture, killing *E. faecalis* completely within 6 min.

**Triphala:** Triphala is a plant blend created by drying and pulverizing the fruit of three plants (termesa bellerica, termesa chebulula, and emblica officinalis) used for medicinal purposes. Triphala kills 100% of *E. faecalis* within 6 min. When used at different rates, its effects can be increased synergistically.

**Conclusion**

Thorough, 3-D disinfection of the entire root canal system
continues to be central to the focus of endodontic improvements, as we remain unsatisfied with "achieving effective reduction" of irritants. However, the physical and biologic obstacles that contribute to decreased successful outcomes have challenged us. Our humbling observation of the infinite anatomical configurations present and the tenacity of the bacteria that contribute to endodontic disease have fueled this continued pursuit of higher levels of disinfection. Many other areas show promise in contributing to safe, active, 3-D disinfection, which illustrate the expansiveness of this focus. Lasers and photoactivated systems have been around for some time, but they have not received wide clinical use. However, these systems may mature and become more popular if they demonstrate clinical relevance in destroying microbes. We know the limitations of current rotary files to directly reach the complex morphology of root canal systems. Some current shaping techniques commonly employed reach less than 50% of canal walls. There has been effort in a new direction of 3-D shaping that strives to increase this percentage. Physically scrubbing more surface area of pulpal walls augments the efficacy of disinfection. Other solutions and techniques may emerge that show greater overall efficacy then those employed today. The hope that we will one day be able to completely eradicate all of the harmful irritants within a diseased root canal system is alive and well. Conquering this next frontier will undoubtedly contribute to even higher successful outcomes of endodontic procedures.

References
1. Zehnder M, Paqué F. Disinfection of the root canal system during root canal re-treatment. Endod Topics, 2008, 19.
2. Zehnder M, Schmidlin PR, Sener B, Waltimo TM: Chelation in root canal therapy reconsidered. J Endod. 2005; 31:817.
3. Zehnder M, Soderling E, Salonen J, Waltimo T. Preliminary evaluation of bioactive glass S53P4 as an endodontic medication in vitro. J Endod. 2004; 30:220.
4. Kakehashi S, Stanley HR, Fitzgerald RJ. The effects of surgical exposures of dental pulps in germ-free and conventional laboratory rats. Oral Surg Oral Med Oral Pathol. 1965; 20:340–349.
5. Grossman LI, Meiman BW. Solution of pulp tissue by chemical agents. J Am Dent Assoc. 1941; 28:223–225.
6. Paqué F, Balmer M, Attin T, Peters OA. Preparation of oval-shaped root canals in mandibular molars using nickel-titanium rotary instruments: A micro-computed tomography study. J Endod. 2010; 36:703–707.
7. Siqueira JF Jr, Alves FR, Versiani MA et al. Correlative bacteriologic and micro-computed tomographic analysis of mandibular molar mesial canals prepared by self-adjusting file, reciproc, and twisted file systems. J Endod. 2013; 39:1044–1050.
8. Richman MJ. The use of ultrasonics in root canal therapy and root resection. Med Dent J. 1957; 12:12–18.
9. Ahmad M, Pitt Ford TR, Crum LA. Ultrasonic debridement of root canals: an insight into the mechanisms involved. J Endod. 1987; 13:93–101.
10. Jensen SA, Walker TL, Hutter JW, Nicoll BK. Comparison of the cleaning efficacy of passive sonic activation and passive ultrasonic activation after hand instrumentation in molar root canals. J Endod. 1999; 25:735–738.
11. Sabins RA, Johnson JD, Hellstein JW. A comparison of the cleaning efficacy of short-term sonic and ultrasonic passive irrigation after hand instrumentation in molar root canals. J Endod. 2003; 29:674–678.
12. Boutsiosiukis C, Verhaagen B, Walmsley AD, Versluis M, van der Sluis LW. Measurement and visualization of file-to-wall contact during ultrasonically activated irrigation in simulated canals. Int Endod J. 2013; 46:1046–1055.
13. Mancini M, Cerroni L, Iorio L, Armellin E, Conte G, Cianconi L. Smear layer removal and canal cleanliness using different irrigation systems (Endo Activator, EndoVac, and passive ultrasonic irrigation): field emission scanning electron microscopic evaluation in an in vitro study. J Endod. 2013; 39:1456–1460.
14. Neuhaus KW, Liebi M, Stauffacher S, Eick S, Lussi A. Antibacterial efficacy of a new sonic irrigation device for root canal disinfection. J Endod. 2016; 42:1799–1803.
15. Alturajii S, Lamphon H, Edrees H, Ahilquist M. Efficacy of three different irrigation systems on removal of calcium hydroxide from the root canal: a scanning electron microscopic study. J Endod. 2015; 41:97–101.
16. Sabins RA, Johnson JD, Hellstein JW. A comparison of the cleaning efficacy of short-term sonic and ultrasonic passive irrigation after hand instrumentation in molar root canals. J Endod. 2003; 29:674–678.
17. Johnson M, Sidow SJ, Looney SW, Lindsey K, Niu LN, Tay FR. Canal and isthmus debridement efficacy using a sonic irrigation technique in a closed-canal system. J Endod. 2012; 38:1265–1268.
18. Nielseni BA, Baumgartner JC. Comparison of the EndoVac System to needle irrigation of root canals. J Endod. 2007; 33:611–615.
19. Munoz HR, Camacho-Cuadra K. In vivo efficacy of three different endodontic irrigation systems for irrigant delivery to working length of mesial canals of mandibular molars. J Endod. 2012; 38:445–448.
20. Tay FR, Gu LS, Schoeff G, et al. Effect of vapor lock on root canal debridement by using a side-vented needle for positive-pressure irrigant delivery. J Endod. 2010; 36:745–750.
21. Pawar R, Alqaied A, Safavi K, Boyko J, Kaufman B. Influence of an apical negative pressure irrigation system on bacterial elimination during endodontic therapy: a prospective randomized clinical study. J Endod. 2012; 38:1177–1181.
22. Dua A, Dua D. Comparative evaluation of efficacy of EndoVac irrigation system to Max-I probe in removing smear layer in apical 1 mm and 3 mm of root canal: An in vitro scanning electron microscope study. Dent Res J. 2015; 12:38–43.
23. Suman S, Verma P, Prakash-Tikka A, Bains R, Kumaar-Shakya V. A comparative evaluation of smear layer removal using apical negative pressure (EndoVac), sonic irrigation (Endo Activator) and Er: YAG laser - An In vitro SEM Study. J Clin Exp Dent. 2017; 9:e981–e987.
24. Versiani MA, Alves FR, Andrade-Junior CV et al. Micro-CT evaluation of the efficacy of hard-tissue removal from the root canal and isthmus area by positive and negative pressure irrigation systems. Int Endod J. 2016; 49:1079–1087.
25. Celaletin Topbas1, Ozkan Adiguzel. Endodontic Irrigation Solutions: A Review. IDR. 2017; 7(3):54-61.