Clinic and laboratory testing of some antiseptic products on aerobic endodontic microflora isolated from gangrenous pulp tissue

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Summary

The ecological medium in the root canal is extremely complex. Adequate nutrition is essential for bacterial growth, and diverse bacteria species have different nutritional requirements.[18] The elimination of bacteria present in the root canal space is the fundamental objective of endodontic treatment, because the bacteria plays an important role in development and maintenance of periapical lesions. The high percentages of endodontic failure of the teeth with periapical lesions have been related to implications of microbial origin.[7,19]

The endodontic treatment using different irrigating solutions for the canal space is becoming more efficient using nontoxic intracanalar medication.[21,1]

***Objectivs* .The aim of these study was the identification of bacterial aerobic species in teeth with simple and complicated gangrene and to investigate the bactericidal effect of three antiseptics. The three antiseptics tested were: Rockle's solution, chlorhexidine gluconate solution and povidone-iodine solution.**

Material and method. **The bacterial stains collected from gangrenous pulp tissue were cultivate in the microbiology laboratory on different culture-medium: Muller-Hinton medium, agar-blood medium and colony medium. For bacterial species identification in the laboratory was used the Microscan-Walkaway 40 bacteriological analyzer. The antiseptics efficiency was tested by placing small filter paper with antiseptic on the Muller-Hinton medium.**

***Conclusion.*.** **The largest inhibition area presented the Rockle's solution, followed by the povidone-iodine solution and the smaller inhibition area presented chlorhexidine gluconate solution.**

Key words: **endodontic treatment, antiseptics, gangrene**

Introduction

Different clinic situations as: cavity preparation, dental trauma and another tipes of dental injuries determine loss of hard tissue, so that the dental pulp is exposed to direct iritation and bacterial colonization.[16] These has a distructive action even on unexposed pulp. Once the enamel was afected, it can not offer protection to the external injuries and the bacteria can invadate dentinal tubules.[9] The size of dentinal tubules is big enought for the majority of bacteria which can colonize the radicular space. [4,11] In the healthy dentine the bacteria has reduced mobility and rarely can fi found in the healthy pulp tissue.[17]

Once the bacteria multiply and decompose, it results different substances which can maintaine inflamatory reaction.[15] In

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case of masive bacterial invasion, the pulp tissue is strongly affected, these beeing the premise of bacterial growth in the radicular canal space.[20]

Because of the improvement in bacteriological techniques, it is possible to cultivate most microorganisms associated with endodontic infections.[12,6]

Objective

The aim of these study was the identification of bacterial aerobic species in teeth with simple and complicated gangrene and to investigate the bactericidal effect of three antiseptics: Rockle‘s solution, chlorhexidine gluconate solution and iodo-iodine solution.

Material and method

The materials used for canal dezinfection in clinical cases of simple and complicate gangrene were: Rockles solution, Septomixine paste, chlorhexidine gluconate solution, calcium hydroxide paste.[13,8,10]

The study was performed in two cases of simple gangrene and five cases of complicated gangrene.

In the microbiology laboratory the bacterial samples were plated onto plates containing the following culture medium: agar-blood, colony medium, Muller Hinton medium. In our study we used the bacteriological analyzer Microscan-Walkaway 40. The bacteriological analyzer Microscan-Walkway 40 has a soft for bacterial species identification. The soft reveals the result (the bacterial species identified) in 24 hours (fig.1). The bacterial strains were placed in individual panels for every pacient and the panel was inserted in the Microscan-Walkaway 40 (fig.2).

Clinic protocol consists of creating a pathway to the apex by inserting a #15 K-file, then a sterile absorbant paper point was

Fig. 1

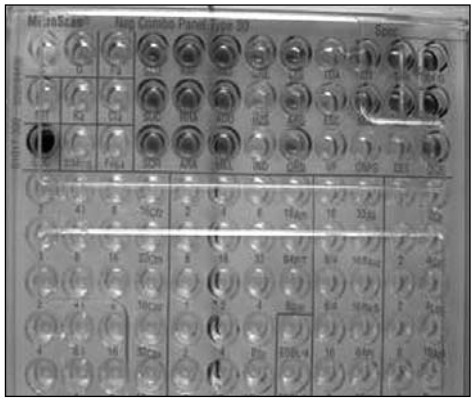

Fig. 2

inserted in the canal to reach the apical portion and allowed to remain in place to absorb as much exudates as possible. If the paper point was dry upon withdrawal from the canal, then another paper point was moistened with transport medium and inserted into to the canal to obtain a sample. Paper points were removed using sterilized cotton forceps and immediately placed in a transport tube. For multi-radicular teeth, all the paper points were placed in the same tube. Sample tubes were identified with the patient’s name, date, involved tooth, and then sent to laboratory to culture and perform microbial tests.

In the microbiology laboratory: with paper points the bacterial stains were plated on cultures medium: agar-blood (solid medium) and colony medium (liquid medium); the culture plates were left in the incubator at 37 ºC for 24 hours; on culture plates were observed the bacterial colony growth and than these were examinee with dark field microscopy; the bacterial colony formated were introduced in Microscan-Walkaway 40 for bacterial species identification.

The Microscan-Walkway 40 has a soft for bacterial species identification and reveal the results after a 24 hours period. On Muller-Hinton medium were plated the bacterial colony obtained on agar- blood medium; on these medium, the three antiseptic solutions were tested: Rockles solution, chlorhexidine gluconate solution and povidone-iodine solution, by evaluating the inhibating area produced. For these there were used small pieces of filter paper with the antiseptic solutions placed on the plates containing Muller-Hinton medium.

In the dental office was performed the endodontic treatment for gangrene using the biomechanical preparation of root canal space; the canals were irrigated using antiseptic solutions: NaOCl 2,5%, chlorhexidine gluconate solution and povidone-iodine solution; sterilization of root canal space was performed with antiseptic solution: Rockles solution. When the canal was steril, evaluated clinic with clean, dry and non smelling tents, a second sample was taken from the canal space with a sterile paper point. [5] Paper points were placed in transport tube and sent to the laboratory. *In the microbiology laboratory* a second culture were made on liquid medium in order to appreciate the sterility of the samples after the endodontic treatment. When the result from the laboratory arrived and the sterility of the radicular canal space was confirmed, in the dental office we performed the radicular sealing using sealer and gutapercha. The final coronal sealing was performed with glassionomer cement.

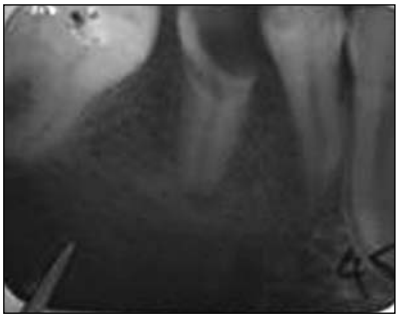

Fig. 3

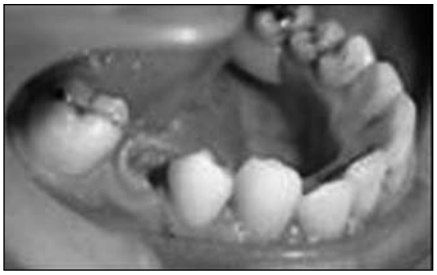

Fig. 4

Fig. 5

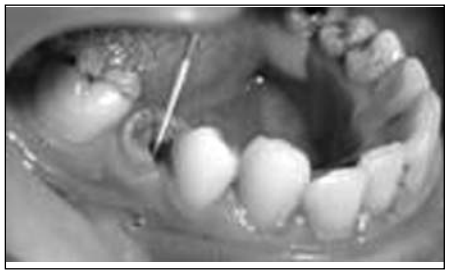

Fig. 6

Results

Clinic case 1

Pacient B.C. 29 years old, masculine gender, diagnosis: 45 complicate gangrene. The diagnosis was based on X-ray image (fig. 3) and clinic evaluation (fig. 4). Steps

of endodontic treatment: a pathway to the apex was created by inserting a #25 K-file (fig.5), with a sterile paper point the bacterial exudates was absorbed from the radicular canal space (fig.6), the transport tube was labeled (fig.7), the culture was made on colony medium (fig.8) and agar-blood medium (fig.9).

Fig. 7

Fig. 8

Fig. 9

The panel with bacterial colony was inserted in the bacterial analyzer – Microscan Walkaway 40. From the cultures were isolated Gram positive aerobic coccus (fig.10) and cocobacilus. (fig.11). On Mulller-Hilton medium were tested the three antiseptic solutions: Rockle‘s solution, chlorhexidine gluconate solution and povidone-iodine solution.

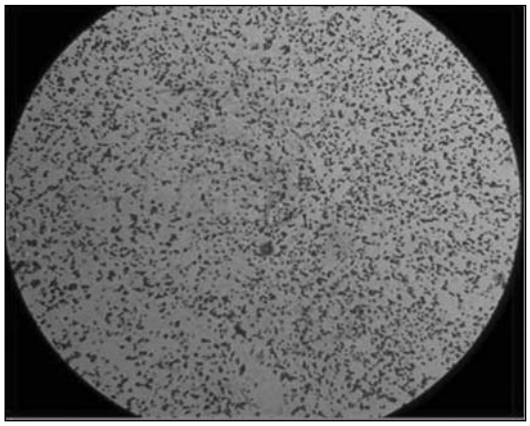

Fig. 10

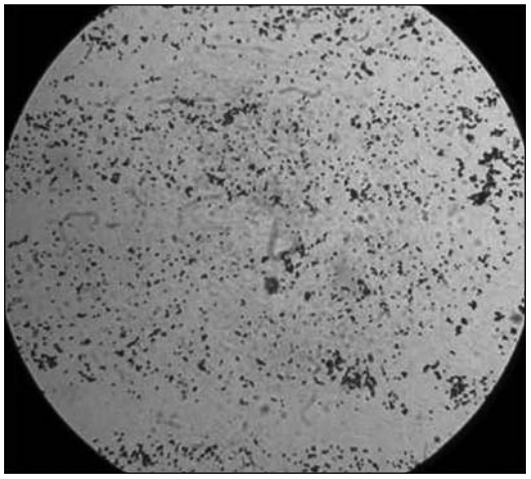

Fig. 11

The largest inhibition area was exhibit by the Rockle‘s solution, followed by povidone-iodine solution and then chlorhexidine gluconate solution. In the dental office using Rockle‘s solution we obtained the sterilization of the radicular canal space, proved by the second culture that was made on colony medium. (fig.12). The final X-ray reveals no inflammatory reaction in the periapical tissues and the and the sealing of the radicular canal space. (fig.13)

Fig. 12

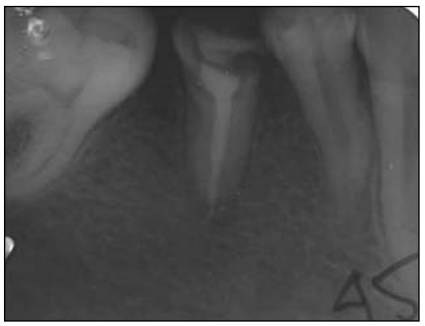

Fig. 13

Clinic case 2

Pacient C.L.; 45 years old; masculin gender, diagnosis: 36 complicated gangrene. Folowing the same clinic steps we treated the complicated gangrene at 36 (fig.14); in these case in the laboratory were identified chains of streptococus (fig.15) and small

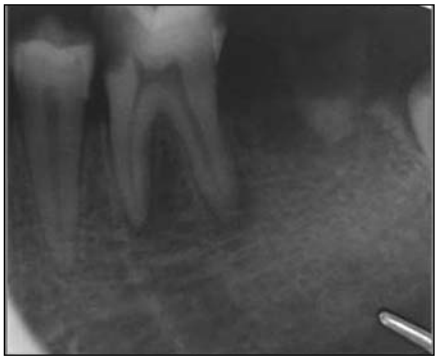

Fig. 14

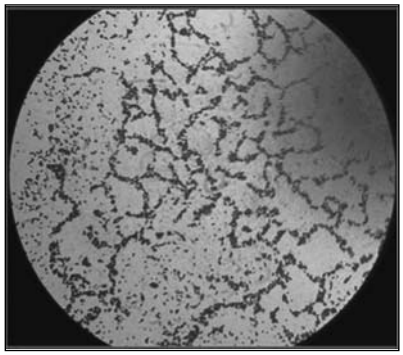

Fig. 15

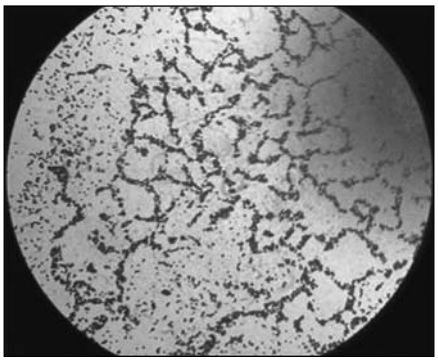

Fig. 16

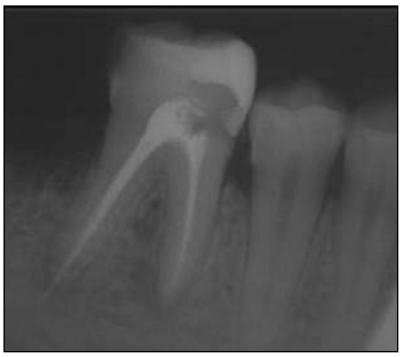

Fig. 17

bipolar cocobacilus (fig.16). The final X-ray shows complete apical healing of the bone (fig.17).

Discutions

At 24 hours from the introduction of bacterial colony in the bacterial analyzer Microscan-Walkaway 40, it reveals the bacterial species identified.

From the seven clinical cases that we treated in the laboratory were identified : big coccus, staphylococcus colonies, streptococcus chains, bipolar cocobacilus, big Gram positive coccus and Candida albicans colonies. Candida albicans is a yeast that usually lives in the oral cavity, and which can penetrate the pulp tissue through the dentinal tubules, dentinal caries, trauma, or through root canal during canal treatment. Clinically Candida species present a high resistance to calcium hydroxide.[14,2] Its resistance to calcium hydroxide and its ability to penetrate into the lateral canals and dentinal tubules are responsible for its presence in persistent apical periodontitis. [19]

Testing the three antiseptic solution on Muller- Hinton mediums showed that the largest inhibition area was exhibit by the Rockle‘s solution , followed by povidone-iodine solution and then chlorhexidine gluconate solution.

In the second sample that were plated on colony medium showed the sterilization of the radicular canal space in five cases from seven. These is according with dates from the dental literature that the use of antiseptics promotes the reduction of endodontic micro biota, however a considerable number of microorganisms were still observed.[3]

Conclusions

In all seven cases was identified aerobic polymorphic micro flora with the majority of Gram positive coccus and Candida Albicans.

Sterilization of the radicular canal space can not be completely obtained, in some cases there remain bacterial species which can not be inhibit by the antiseptic solutions.

All three antiseptic solution were proven to be active on micro biota isolated from the infected radicular canal space.

However, in our study the Rockle´s solution was proven to be the most active, both in the clinic treatment and at the laboratory test.

Chlorhexidine and povidone-iodine proved to have antibacterial and antifungal effects when used in the treatment of pulp gangrene and in cases failing to respond to conventional therapy.

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