“VICTOR BABEȘ” UNIVERSITY OF MEDICINE AND PHARMACY FROM TIMISOARA

FACULTY OF DENTAL MEDICINE

Department I

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PHD THESIS

EFFICACY OF ANTIBACTERIAL SOLUTIONS ON INFECTED DENTIN IN CARIOUS LESIONS

A B S T R A C T

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T i m i ș o a r a

2023

A B S T R A C T

Over time, dentists and dental medicine researchers have been concerned about the occurrence of carious lesions, their causes, determinants, risk factors, but especially, about their treatment. Lately, the definition of a "caries lesion" has gained interest, being considered more of a "caries disease," as it describes a very complex pathology and its treatment often become great challenge for practitioners.

The greatest challenges arise, especially in clinical cases with deep carious lesions, with a thin layer of infected/affected dentin in close proximity to the pulp chamber, the exposure risk being imminent.

The motivation behind the current research themes is the desire to deepen and complement existing studies on the treatment of deep carious lesions, especially when it comes to the situation of performing an indirect pulp capping. Thus, the study of the effects of well-known and approved antibacterial substances on infected and affected dentin, located in close proximity to the pulp chamber, can have significant clinical implications. Specifically, the aim is to determine whether these products/substances have a clear antimicrobial effect on infected dentin, in order to increase the success rate of indirect pulp cappings and, consequently, preserve pulp vitality.

Therefore, **the first study** evaluated the effectiveness of remineralization using two solutions with potential antibacterial roles, namely a natural paste created from natural products and a bonding agent enriched with silver nanoparticles.

With the help of **the second study**, we aimed to comparatively evaluate the effectiveness of chlorhexidine and the solution with silver nanoparticles, Nanocare Plus, in inhibiting bacterial activity. The efficacy of the products in limiting local bacterial activity was measured using a medical device called the Diagnodent pen.

The third study followed with an in vitro analysis of morphological aspects in the remineralization of teeth using the Advantage Arrest Silver Diamine Fluoride solution, produced by Elevate Oral Care. The results were interpreted using an optical microscope, digitally equipped with Snapshot Software and Hardware; this

microscope was created by Dental Technician Paul Popovici, within the Discipline of Propaedeutics and Dental Materials.

The recent interface studies evaluated the ex vivo adaptation of composite acrylic resins to the walls of the dental cavity using confocal microscopy and micro-CT. This aims to draw attention to the importance of early detection of marginal infiltration, with the potential for secondary caries, in order to maintain tooth vitality.

The doctoral thesis is divided into **two sections**: the first part includes significant aspects from the relevant literature that are pertinent to the thesis topic, while the second part focuses on personal contributions, concluding with final remarks, bibliographic references, and annexes.

The **general part** comprises two chapters, providing an overview of the anatomy of hard dental tissues and carious pathology, as well as general notions related to the impact of nanotechnology on dental medicine.

The first chapter extensively defines the composition of enamel and dentin, as well as the demineralization process and carious pathology. It then presents the clinical characteristics of carious lesions, explaining the stages leading to the progression of a carious lesion, from incipient to deep carious lesions.

The second chapter reviews the mechanisms of action of silver nanoparticles in dental medicine, as well as the areas in which these nanoparticles can be utilized.

The section dedicated to **personal research**, the **special part**, is divided into 5 chapters, presenting detailed accounts of the conducted studies. These chapters include an introduction, clarification of the purpose and proposed objectives, a description of the materials and methods used in the research, presentation of the obtained results, discussions, conclusions, and general conclusions. Special emphasis is placed on highlighting personal contributions, and at the end of the section, bibliographic references are included.

For the implementation of the studies described in this thesis, after presenting the research protocol, approval was sought and obtained from the Research Ethics Committee (CECS) within the "Victor Babeș" University of Medicine and Pharmacy in Timișoara (Annex 1 - Approval No. 30/2016).

In the case of clinical studies, following the review of their written protocols, the involved patients or their legal representatives signed the written agreement to participate in the study - "Patient Consent for Involvement in Medical Research."

Additionally, each patient or their legal representative signed the informed consent, providing agreement on the procedures to be performed.

A part of the utilized materials and experimental activities were financially supported through the doctoral grant won in the competition organized by UMFVBT, with grant number 3722/03.10.2016.

Chapter 3 presents the evaluation of new approaches to combating dental demineralization through unconventional treatments. The aim of this study was to introduce new methods of tooth remineralization. Currently, various products such as ICON (Infiltration Concept - DMG America) or products containing fluoride are used to remineralize enamel. The study observed an improvement in the remineralization of white spots on the smooth surfaces of teeth using a natural paste. The OCT device was able to detect different levels of remineralization in all cases studied. Demineralized areas can be seen as spots with a reduced refractive index in OCT images; after remineralization, the surfaces appear more compact. The specific non-invasive features of these products, along with their low costs and environmental safety, are strong positive aspects of this method for remineralizing dental white spots. However, the natural remineralization process is a lengthy one, and refining key substances to expedite the process, in addition to further in vivo studies, should be pursued. The non-invasive OCT investigation method has proven effective in detecting different degrees of enamel demineralization and remineralization in all cases studied.

Chapter 4 moves on to the clinical evaluation of other solutions, namely Nanocare Plus, to assess its effectiveness on dental biofilm. A report of two clinical cases with existing carious lesions was thus conducted. This study succeeded in comparing the antibacterial effect of the two solutions, collecting valuable information potentially useful for further research in this field. The decrease in bacterial fluorescence was higher with chlorhexidine than with Nanocare Plus. Both cases had intermediate carious lesions where infected dentin could be completely removed, and the DIAGNOdent values were below the diagnostic threshold of 20.

Chapter 5 comes as a supplement to Chapter 4, studying another product containing silver nanoparticles, namely Advantage Arrest. The studies have demonstrated that silver nanoparticles (AgNP) can inhibit the replication of bacterial deoxyribonucleic acid (DNA) when in contact with Ag+ ions and induce significant structural changes in the bacterial membrane.

A possible improvement in the remineralization of white spots on flat and occlusal surfaces was observed, with the optical microscope detecting demineralization and the penetration of Advantage Arrest through enamel. The main component of Advantage Arrest is SDF, with the following chemical formula and concentrations: Ag(NH3)2F, content: 25–27% Ag, 5–6% fluoride (F), 9–10% ammonia (NH3) (w/v) (107). This product, incorporating both an antibacterial and remineralizing substance, enhances tooth resistance to acid action by forming Ag–protein conjugates on the surface of the caries-affected tooth. It also increases mineral density by promoting the growth of hydroxyapatite (HA) and fluorapatite. Ag+ and F– ions penetrate approximately 25 μm into enamel and 50–200 μm into dentin; F promotes remineralization, while Ag promotes antimicrobial action.

Chapter 6 demonstrates the efficiency of MICRO-CT in assessing the marginal and internal adaptation of dental composites. Several aspects were concluded, such as the tight connection between polymerization shrinkage and the internal and marginal adaptation of resin composite restorations. Significant discrepancies were detected between different polymerization times, and notable variations were identified between bulk-fill dental composite and hybrid resin, concluding that bulk fill might be the better option. The proposed method for evaluating internal adaptation using a micro-CT system was found to provide a new measurement tool for assessing internal adaptation of restorations with a non-destructive approach.

Chapter 7 employs laser scanning confocal microscopy to evaluate the tooth-composite resin interfaces and surface texture. Marginal integrity is a crucial aspect to ensure the durability of a restoration. This integrity can be affected by margin separation, which occurs due to polymerization shrinkage. Polymerization shrinkage is typically the main cause of failure in direct composite restorations in the posterior area of dental cavities. This shrinkage depends on various factors, including cavity configuration, material composition, and the amount of material used. It was concluded that laser scanning confocal microscopy (CLSM) provides detailed and

reliable information on penetration, distribution, marginal discrepancy, and the surface of composite resin, making it a valuable technique for evaluating adhesion at the interface of composite restorations compared to conventional methods.

The results obtained in the conducted studies pave the way for future directions and research topics, providing perspectives for further investigations.

The conclusions, original aspects, and new perspectives developed through the personal contributions in this doctoral thesis present direct clinical applicability. This brings benefits to practitioners and patients, offering the possibility of improving the quality of treatments that patients receive.