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**BIOLOGICAL METHODS OF DEEP CARIES TREATMENT IN  
PERMANENT TEETH**

**323.01 STOMATOLOGY**

**Summary of the Doctoral Thesis in Medical Sciences**

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The thesis was developed at the Department of dental propaedeutics „Pavel Godoroja”, State University of Medicine and Pharmacy „Nicolae Testemițanu”

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## INTRODUCTION

**Relevance and importance of the topic.** Dental caries remains the most widespread pathology of the dental system worldwide and is the main cause of tooth loss. According to World Health Organization (WHO) statistics from 2022, the European region had the highest prevalence of caries in permanent teeth among all 6 WHO regions - 33.6% of the population, which constitutes almost 335 million cases. According to WHO data, 8 out of 10 Moldovan adults suffer from tooth decay [14]. A study carried out in 2018 in the Republic of Moldova, among rural and urban schoolchildren found a caries prevalence index of 87.4% at the age of 6 years, 77.53% % at the age of 12 years, 86.2% at the age of 15 years [1]. According to Cartaleanu A. (2006), the share of deep dental caries constitutes 23% of all caries, and the complications arising from the evolution of deep caries are determined by the extension of the inflammatory process in the pulp. This inflammatory process can be reversible with appropriate conservative treatment, or it can be irreversible and will require endodontic treatment. Therefore, tooth decay continues to represent a complex problem both from a medical and social point of view.

Vital pulp therapy (VPT) is a group of conservative methods of dental treatment, the main purpose is to preserve and maintain the pulp tissue, which has been compromised, but not destroyed by caries, trauma or restorative procedures. Depending on the level of pulp tissue preservation, vital pulp therapy includes the following techniques: indirect pulp capping, direct pulp capping, mini-pulpotomy, partial pulpotomy (Cvek) and full coronary pulpotomy. The positive prognosis of these techniques is influenced by the age and general condition of the patient, the elimination of etiological factors, the complete removal of contaminated tissues and the hermetic restoration of the tooth [12]. The American Association of Endodontists in 2021 updated the protocols regarding vital pulp therapy and highlighted the following key points: determination of pulp status - is based on patient history, pain characteristics and evaluation of pulp sensitivity (cold testing and electric pulp testing); complete removal of carious tissue (demineralized enamel and infected dentin) and preservation of dentin that can be remineralized; disinfection of the carious cavity and hemostasis with the use of high-concentration sodium hypochlorite; the use of calcium silicate cements as the curative materials of choice; application in the same visit of the permanent restorative material [5].

Pulp capping is a biological method of deep caries treatment, a complex therapeutic act that consists in the application of a curative material directly or indirectly on the dental pulp, which ensures the disinfection of the dentine wound; sealing of the dentinal canals; protecting and isolating the dental pulp against physical, chemical and bacterial irritants; stimulating neodentinogenetic processes, thus preserving the vitality and functionality of the dental pulp. Indirect pulp capping is defined as a biological treatment method, which consists of applying a bioactive material over a thin layer of remaining dentin after excavating carious tissues, without pulp exposure. Direct pulp capping is defined as a method of conservative treatment of a traumatic, mechanical or carious exposure of the vital pulp, which consists in sealing the pulp wound with a biomaterial applied directly to the exposed pulp [6].

To reduce the risk of infection and further damage to the pulp, pulp capping should be performed with a biocompatible material with curative properties and which ensures adequate sealing of the dentinal or pulpal wound. Numerous biomaterials have been used over the years for pulp capping, calcium hydroxide being considered the gold standard for many decades. Nowadays, calcium silicate

cements, which possess multiple advantages, are increasingly used in vital pulp therapy and other dental treatments.

**The purpose of the research:** Comparative study of the effectiveness of 3 curative dental materials used in the conservative treatment of deep caries in permanent teeth, to optimize the therapy of this pathology.

**Research objectives:**

1. Determination of EPT values obtained at the diagnostic stage and post-treatment (3 and 6 months), depending on the material used.
2. Evaluation of the results of the cold tests obtained initially and after treatment (3 and 6 months), depending on the applied material.
3. Comparative analysis of the paraclinical parameter - the presence or absence of the dentine bridge on the radiograph, after 6 months, depending on the remedy used.
4. Comparison of the clinical efficiency of the dental materials used as a pulp capping agent – calcium hydroxide, mineral trioxide aggregate, tricalcium silicate.
5. Development of a clinical algorithm for the diagnosis and treatment of deep dental caries in permanent teeth.

**Scientific research methodology.** National and international bibliographic sources were comparatively analyzed, which facilitated the development of the algorithm for the diagnosis and treatment of deep caries in permanent teeth. In order to achieve the purpose and objectives of the research, an analytical, clinically controlled study was carried out. It was compared the efficiency of 3 materials used in the treatment of deep dental caries: calcium hydroxide, mineral trioxide aggregate and tricalcium silicate.

The study was conducted on a sample of 159 patients aged between 10 and 35 years. The patients were divided into 3 equal groups of 53 patients with deep carious lesions in permanent teeth with fully built apex, who were treated by direct or indirect pulp capping method in a single session, by applying a thin layer of a curative material. For group 1 (control) - calcium hydroxide was used, group 2 (research) - mineral trioxide aggregate was applied, group 3 (research) - tricalcium silicate was used.

The approval of the thesis topic was obtained during the meeting of the Scientific Council of SUMPh "Nicolae Testemițanu", documented in minutes no. 1/5.2 dated 14.12.2020. The positive approval of the Research Ethics Committee for the study was obtained at the meeting of 06.19.2017, minutes no. 94 and at the meeting of 01.02.2024, minutes no. 2.

**Scientific novelty and originality:**

1. A clinical trial was conducted comparing the effectiveness of calcium hydroxide versus cements based on mineral trioxide aggregate and tricalcium silicate for pulp capping in permanent teeth affected by deep caries.
2. For the first time, the dynamics of EPT values were analyzed after 3 and 6 months after the treatment (pulp capping) in case of deep caries, depending on the applied material.
3. For the first time, the radiological presence/absence of the newly created dentine bridge was determined after 6 months from the applied treatment, especially for mineral trioxide aggregate and tricalcium silicate.

4. Recommendations were issued regarding the diagnosis of pulpal status in case of deep caries and the delimitation of the criteria according to which carious permanent teeth can be treated by conservative methods.
5. A clinical algorithm for the diagnosis and conservative treatment of deep caries in permanent teeth with calcium silicate cements was developed.

**Practical significance:**

1. The limits of the EPT values according to which the permanent teeth can be subjected to conservative treatment in case of deep caries have been defined.
2. It was established that the tertiary dentin bridge is visible on the radiograph after 6 months of treatment with any of the 3 materials studied.
3. Conservative treatment with the application of materials from all groups allowed to preserve the vitality and functionality of the majority of treated teeth.
4. Following the algorithm of diagnostic and treatment developed during the research decreased the risk of endodontic complications and the number of treatment visits.

**The scientific findings** have been implemented into the research, methodological, and clinical activities within the Department of dental propaedeutics "Pavel Godoroja" at SUMPh "Nicolae Testemițanu" and are implemented in the practical activity of the university dental clinic no. 2, dental clinic "Dia Dents", dental clinic "Dicri Med".

**Approval of scientific results.** The results of the study were presented and discussed in the following national and international scientific forums, including: Days of the State University of Medicine and Pharmacy "Nicolae Testemițanu" (2017, Chisinau, Republic of Moldova; 2022, Chisinau, Republic of Moldova; 2023, Chisinau, Republic of Moldova); MedEspera International Medical Congress (2018, Chisinau, Republic of Moldova); The national scientific-practical conference with international participation In memoriam Gheorghe Țibîrnă (2023, Chisinau, Republic of Moldova); The conference devoted to the Saint Apollonia Dentist's Day and the Commemoration of the Academician, University Professor, Doctor habilitate, Ion Lupan. (2018 Chisinau, Republic of Moldova); Scientific conference "Digital Technologies in Multidisciplinary Dentistry", 2023; Chisinau, Republic of Moldova); VIII International Congress of the Romanian Dental Association for Education "Theory versus Practice in Medical Sciences" (2016, Iași, Romania); XII International Scientific and Practical Conference «Priorities of pharmacy and dentistry: from theory to practice», (2023, Almaty, Kazakhstan).

**The volume and structure of the thesis.** The text of the thesis is presented on 105 text pages, processed on the computer, consisting of: list of abbreviations, introduction, 4 chapters, synthesis of the obtained results, general conclusions, practical recommendations, bibliography from 120 sources and 1 appendix. Illustrative material includes 16 tables and 54 figures.

**Key words:** deep dental caries, pulp-dentine complex, pulp capping, calcium hydroxide, mineral trioxide aggregate, tricalcium silicate, dentine bridge.

# 1. MATERIALS AND RESEARCH METHODS

## 1.1. General characteristic of the research

The thesis was developed at the Department of dental propaedeutics "Pavel Godoroja" of the Faculty of Dentistry of IP SUMPh "Nicolae Testemițanu". The research was carried out within the University dental clinic and the dental clinics "Dia Dents" and "DicriMed", during 2017-2023.

In order to achieve the purpose and objectives of the research, we carried out an analytical, experimental, clinically controlled study to compare three materials used in the conservative treatment of deep dental caries.

The applied biological method of treatment will consist of performing direct or indirect pulp capping in one visit. After local anesthesia and isolation of the operating field with rubber dam, the carious cavity will be prepared and disinfected using optical magnification. Afterwards, a layer of 1-1.5 mm of curative material will be applied on the deepest point of the cavity or directly on the exposed pulp. The the isolating and permanent filling will be applied in the same visit. Recall visits for clinical and paraclinical examination will be made after 3 and 6 months, and every 6 months during 2 years from the treatment.

The study included 159 patients aged between 10 and 35 years, with deep carious lesions in permanent teeth with fully built apex. It was divided into 3 equal groups of 53 patients each treated by the method of pulp capping with a curative material according to the distribution in groups: control group 1 - calcium hydroxide, research group 2 - mineral trioxide aggregate, research group 3 - tricalcium silicate.

The inclusion criteria were:

1. Presence of informed consent.
2. Clinical diagnosis of deep dental caries in permanent teeth with fully formed apex.
3. Age between 10 and 35 years.
4. The absence of periodontal diseases of the treated teeth.
5. Lack of periapical radiological changes of the teeth in question.
6. Availability of patients to participate in periodic examinations during the study.

The exclusion criteria included:

1. Refusal to participate in the study or unavailability of supervision during the study.
2. Clinical diagnosis of medium caries, irreversible pulpitis or chronic apical periodontitis.
3. The presence of periodontal diseases in the teeth in question.
4. The presence of periapical radiological changes of teeth to be treated.
5. Acute dental trauma with exposure of the dental pulp, accidental exposure of the dental pulp.
6. Patients with general illnesses (diabetes, cardiovascular diseases, treatment with cytostatics or chemotherapy).
7. Patients with cardiac pacemaker - due to the specificity of the electroodontometry method.
8. Pregnancy or lactation - due to the limitation of indications for radiography and anesthesia.

## 1.2. Clinical investigation methods

All patients included in the study were examined according to the same algorithm, by the following methods: subjective examination, objective clinical examination, complementary examinations and paraclinical (radiological) examinations. These methods were applied both at the initial diagnostic and treatment visit and at the follow-up visits after 3 and 6 months.

The subjective examination elucidated the personal data, anamnesis vitae, anamnesis morbi and the specific complaints for deep caries: sensitivity or pain to cold and sweet stimuli that disappears once the stimulus is removed, the presence of a cavity in the tooth with/without food retention.

Following the clinical extraoral objective examination, no pathological changes were detected in the case of deep dental caries. The clinical intraoral objective examination included the clinical-instrumental examination by the following methods: inspection, probing, palpation and percussion. No pathological changes of the mucosa were detected.

The inspection of the teeth revealed: the lack of dental hard substance (carious cavity), the presence of an old, defective filling, color changes of some dental surfaces, from chalky white to gray-brown. The probing revealed: the carious cavity with or without the external enamel wall, the presence of altered dentin inside the cavity, its consistency depending on the type of evolution of the carious process, acute or chronic, sensitivity to probing, the loss of dental hard substance, , no communication with the pulp chamber was detected. Palpation revealed no pathological changes. The vertical percussion of the examined tooth was not painful.

The complementary examination of the patients was carried out by the following methods: cold thermal test and electrical pulp testing (EPT).

For the cold testing of the pulpal sensitivity of the tooth, a cotton pellet pre-soaked with ROEKO Endo-Frost cooling spray (Coltene) was used, which was placed on the middle of the vestibular surface (figure 1). The patient is instructed to raise their hand when they first feel the sensation from the thermal stimulus, to hold their hand up as long as this sensation persists, and to lower their hand when the sensation disappears. If the patient does not perceive any sensation in the tooth after 5-6 seconds, then the stimulus should be removed. Because some pulpally affected teeth may not be immediately stimulated by the heat test, the examiner should wait a few seconds after the stimulus is removed from the tooth to see if any pain occurs before placing the stimulus on the next tooth. Some teeth with irreversible pulpitis require repeated stimuli to reach the threshold that causes pain. It is important to note that once a tooth has been cold tested, there is a refractory period of several minutes before a second thermal stimulation can be accurately performed on the same tooth.



**Figure 1. Cold thermal testing with Endo-Frost refrigerant spray**

Pulp sensitivity testing to electrical stimuli or electroodontodiagnostics was performed using the DigiTest digital pulp tester, by placing the tip of the device on the middle of the vestibular surface, in the presence of a conductive medium (figure 2).





Figure 2. **Performing EPT using the DigiTest digital pulp tester**

The radiological method of examination and diagnosis consisted from a digital orthopantomography using the Planmeca ProMax 3D device. The evaluation of the presence/absence of the newly created dentin bridge after 6 months was performed on digital retroalveolar radiographs where new dentine bridges of variable thicknesses were detected, as illustrated in figure 3.

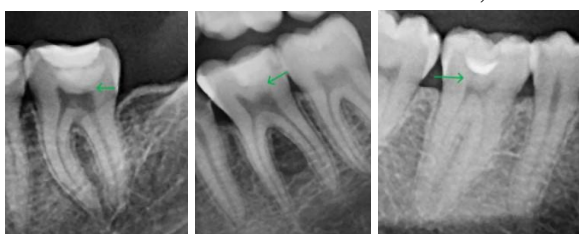


Figure 3. **Radiological determination of the presence of the newly created dentin bridge 6 months after the pulp capping**

### 1.3. Statistical analysis of the results

The research database includes qualitative and quantitative data obtained from the examination of 159 patients, divided into 3 equal groups, according to an identical algorithm at the initial visit, 3 and 6 months after treatment. The primary materials of the study were entered into an electronic database and processed using the functions and modules of the program SPSS version 16.0 for Windows (SPSS Inc., Belmont, CA, USA, 2008) and Microsoft Office Excel 2019 on the personal computer through procedures descriptive and inferential statistics. We calculated absolute and relative frequencies for nominal or categorical variables, mean values, standard errors and standard deviations of the mean for quantitative or continuous (interval or ratio) variables. We used the  $\chi^2$  method after Pearson,  $\chi^2$  with Yates' correction or Fisher's exact method for comparing discrete variables; the Kolmogorov-Smirnov test for checking the normality of interval-scaled variables; "t" test or non-parametric statistical tests to determine the statistical difference of mean values between groups; univariate analysis of variance with the application of post hoc analysis tests and the non-parametric Kruskal-Wallis test for testing multiple differences between mean values in the study groups; correlation analysis (r Pearson,  $\rho$  Spearman,  $\tau$  Kendall) to assess the degree of intensity and direction of statistical links. We considered statistically significant differences with a bilateral value of  $p < 0.05$ .

## 2. ASSESSMENT OF BIOLOGICAL METHODS OF DEEP CARIES TREATMENT

### 2.1 General characteristics of the studied groups

The distribution of patients by gender was: male - 47 patients (29.6%; CI 95% [22.0-37.1]), female - 112 patients (70.4%; CI 95% [62.9- 78.0]). The distribution according to the place of residence of the patients was: urban environment - 94 patients (59.1%; CI 95% [51.6-66.7]), rural

environment - 65 patients (40.9%; CI 95% [ 33,3-48,4]). The distribution according to these 2 criteria is illustrated in figure 4.

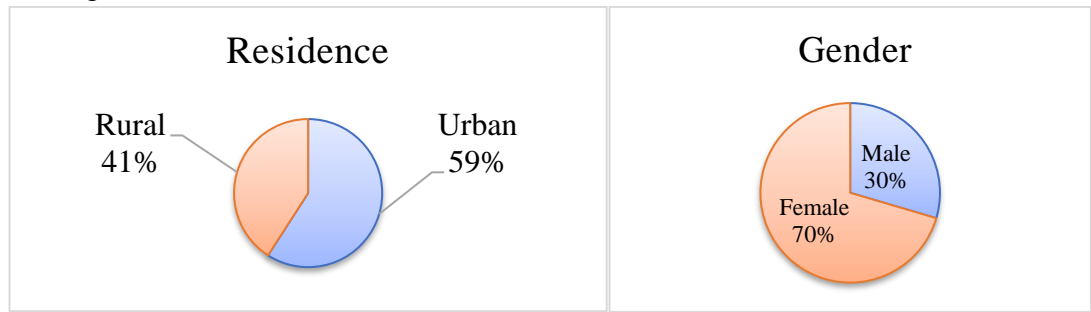


Figure 4. Patients` distribution by place of residence and gender (%)

The age distribution of the patients was: 10-18 years: 76 (47.8%; 95% CI [40.3-56.0]) patients; 18-25 years: 60 (37.7%; 95% CI [29.6-45.9]) patients; 25-35 years: 23 (14.5%; 95% CI [9.4-20.1]) patients. Out of the total number of treatments performed, 73 cases (45.9%; 95% CI [38.4-54.1]) were treated by the direct pulp capping method, and 86 cases (54.1%; 95% CI [45 ,9-61,6]) were treated by the method of indirect pulpal capping. The distribution according to these 2 criteria is illustrated in figure 5.

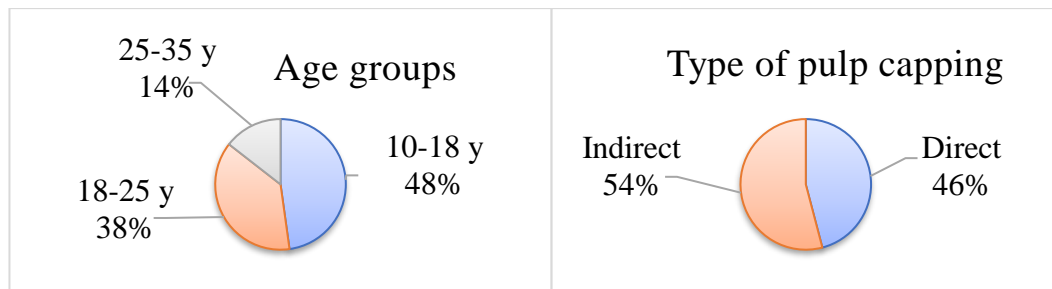


Figure 5. Distribution according to age groups and type of pulp capping performed (%)

From the total of 159 treated teeth, 137 teeth were primarily affected by caries (86.2%; 95% CI [80.5-91.8]), and 22 teeth were previously subjected to dental treatment, presenting secondary or recurrent caries (13.8%; 95% CI [8.2-19.5]). According to the type of clinical evolution, the share of chronic deep caries constituted 127 cases (79.9%; CI 95% [73.6-86.2]), and of acute deep caries - 32 cases (20.1%; CI 95% [13.8-26.4]). These parameters are represented in figure 6.

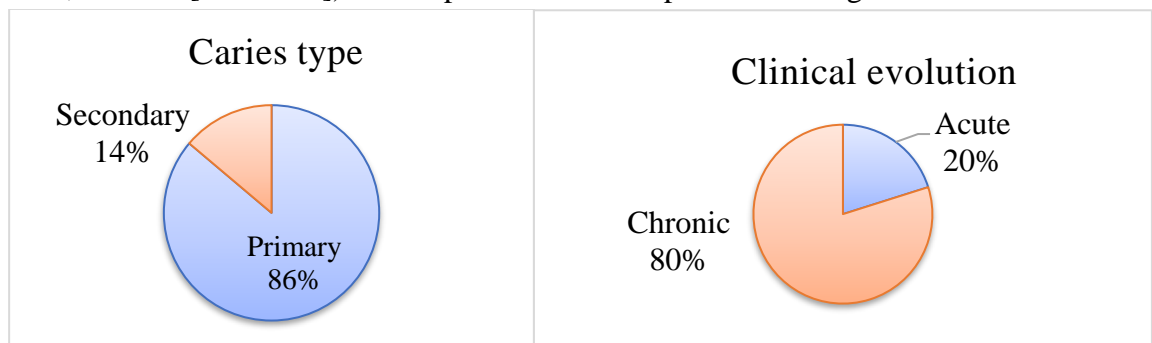


Figure 6. Distribution of dental caries according to type and clinical evolution (%)

From the total of 159 treated teeth, 106 teeth were molars (66.3%; 95% CI [59.3-74.0]), 45 - premolars (28.3%; 95% CI [21.3- 53.5]), 8 - anterior teeth (5.0%; CI 95% [1.6-8.4]). According to the classification by Black, class I cavities were treated in 38 teeth (23.9%; 95% CI [17.6-30.8]), class

II – 113 teeth (71.1%; 95% CI) [64.2-77.4]), class III – 8 teeth (5%; CI 95% [1.9-8.8]), class IV, V, IV – 0 teeth (0%). These distributions are shown in figure 7.

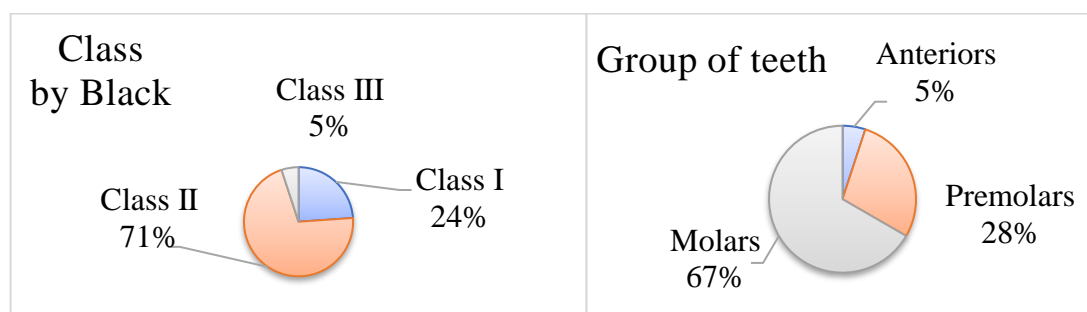


Figure 7. Distribution of carious cavities according to group and Black classification (%)

## 2.2. Analysis of EPT values obtained at the initial stage and after treatment

The results of the EPT performed at the initial visit served as a diagnostic criterion for deep dental caries and were within the reference limits, according to the group of the studied tooth. In group 1 the mean values were:  $18.88 \pm 3.050 \mu\text{A}$  (Median 20.00, IQR = 4), in group 2 the mean values were:  $19.08 \pm 1.948 \mu\text{A}$  (Median 19.00, IQR = 4), in group 3 the mean values were:  $18.92 \pm 2.213 \mu\text{A}$  (Median 20.00, IQR = 5). No statistically significant differences were detected between groups ( $F=0.173$ ;  $p=0.841$ ). The average values obtained are shown in table 1.

At the 1st control visit, after 3 months, the EPT values decreased but did not reach the reference limits considered the norm, according to the group of the studied tooth. In group 1 the mean values were:  $17.15 \pm 2.917 \mu\text{A}$  (Median 18.00, IQR = 4), in group 2 the mean values were:  $16.24 \pm 1.818 \mu\text{A}$  (Median 17.00, IQR = 2), in group 3 the average values were:  $16.56 \pm 2.578 \mu\text{A}$  (Median 18.00, IQR = 6). No statistically significant difference was detected between the values obtained between groups 1 and 2 ( $p=0.087$ ), between groups 1 and 3 ( $p=0.087$ ), nor between groups 2 and 3 ( $p=1.000$ ).

Table 1. Mean EPT values at baseline and after 3 and 6 months of treatment, ( $\mu\text{A}$ )

EPT ( $\mu\text{A}$ )		Initial	After 3 months	After 6 months
Group 1	Media	18,88	17,15	14.94
	DS	3,050	2,917	2,613
	Mediana	20,00	18,00	16,00
	IQR	4	4	4
Group 2	Media	19,08	16,24	13.57
	DS	1,948	1,818	1,616
	Mediana	19,00	17,00	14,00
	IQR	4	2	3
Group 3	Media	18,92	16,56	13,75
	DS	2,213	2,578	2,104
	Mediana	20,00	18,00	15,00
	IQR	5	6	4

At the 2nd control visit, after 6 months, the EPT values decreased, and reached the reference limits considered the norm, according to the group of the studied tooth. In group 1 the average values were:  $14.94 \pm 2.613 \mu\text{A}$  (Median 16.00, IQR = 4), in group 2 the average values were:  $13.57 \pm 1.616 \mu\text{A}$  (Median 14.00, IQR = 3), in group 3 the values averages were:  $13.75 \pm 2.104 \mu\text{A}$  (Median 15.00,

IQR = 4). A statistically significant difference was identified between all 3 groups ( $F=5.931$ ;  $p=0.003$ ). A significant statistical difference was detected between the values obtained between groups 1 and 2 ( $p=0.005$ ), as well as between groups 1 and 3 ( $P=0.019$ ). No statistically significant difference was detected between groups 2 and 3 ( $p=1.000$ ).

### 2.3. Comparison of the results of cold testing at diagnostic stage and after treatment

The results of the cold test performed at the initial visit were distributed as follows: 18 teeth had a similar response to the reference healthy tooth (11.3%; 95% CI [6.3-16.4]), 141 teeth had short-lasting pain that disappeared with the removal of the stimulant (88.7%; CI 95% [83.6-93.7]).

At the 1st control visit, a response similar to the healthy tooth chosen as a reference was presented by: group 1 - 48 teeth (90.6%; CI 95% [82.4-98.1]), group 2 - 51 teeth (96, 2%; 95% CI [90.0-100.0]), group 3 - 52 teeth (98.1%; 95% CI [92.7-100.0]). Long-lasting pain after removal of the thermal agent was presented in: group 1 – 5 teeth (9.4%; 95% CI [1.9-17.6]), group 2 – 2 teeth (3.8%; 95% CI [0.0-10.0]), group 3 – 1 tooth (1.9%; 95% CI [0.0-7.3]). Statistically significant difference between groups was not determined ( $\chi^2=3.422$ ;  $gl=2$ ;  $p=0.181$ ). At the 2nd control visit, all 151 teeth remaining in the study were evaluated and all had a response similar to the healthy tooth chosen as a reference to the cold thermal test (95.0%; CI 95% [91.2-98.1]). This distribution is reflected in table 2.

Table 2. **Distribution of cold test results performed at the initial visit, at 1st and 2nd control visits, (abs., %)**

	Similar response	Short-lasting pain	Long-lasting pain
Initial visit	18 (11, 3%; $\hat{\Pi}$ 95% [6,3-16,4])	141 (88,7%; $\hat{\Pi}$ 95% [83,6-93,7])	-
1st control visit	151 (95,0%; $\hat{\Pi}$ 95% [91,2-98,1])	-	8 (5,0%; $\hat{\Pi}$ 95% [1,9-8,8])
2nd control visit	151 (95,0%; $\hat{\Pi}$ 95% [91,2-98,1])	-	-

### 2.4. Comparative analysis of the results of paraclinical investigations after 6 months

The presence of the newly created dentine bridge with variable thickness on the radiograph taken after 6 months is the final criterion for determining the success of the treatment,. The presence of the bridge was observed according to the distribution by groups, as follows: group 1 - 48 cases (90.6%; CI 95% [82.4-98.1]), group 2 - 51 cases (96.2%; CI 95% [90.0-100.0]), group 3 - 52 cases (98.1%; 95% CI [92.7-100.0]).

The lack of the dentine bridge together with the other clinical parameters (thermal test and EPT) served as an indicator of treatment failure. The bridge was not detected on radiography, as follows: group 1 – 5 cases (9.4%; CI 95% [1.9-17.6]), group 2 – 2 cases (3.8%; 95% CI [0.0-10.0]), group 3 – 1 case (1.9%; 95% CI [0.0-7.3]).

Statistically significant difference between groups regarding the presence/absence of the dentine bridge was not determined ( $\chi^2=3.422$ ;  $gl=2$ ;  $p=0.181$ ). These values are shown in table 3.

Table 3. Frequency of the presence/absence of the dental bridge on radiograph after 6 months, depending on the study group, (abs., %)

Presence of the dental bridge		Research group	Absence of the dental bridge	
Abs.	%		Abs.	%
48	90,6	Group 1	5	9,4
$\chi^2=3,422; gl=2; p=0,181$		Statistical significance <i>P</i>	$\chi^2=3,422; gl=2; p=0,181$	
51	96,2	Group 2	2	3,8
$\chi^2=3,422; gl=2; p=0,181$		Statistical significance <i>P</i>	$\chi^2=3,422; gl=2; p=0,181$	
52	98,1	Group 3	1	1,9
$\chi^2=3,422; gl=2; p=0,181$		Statistical significance <i>P</i>	$\chi^2=3,422; gl=2; p=0,181$	

#### 2.4. Comparison of the clinical effectiveness of pulp capping materials

The criteria for determining the success of the treatment after 3 months were the absence of spontaneous or nocturnal pain, the response of the studied tooth to the cold thermal test similar to a healthy tooth chosen as a reference and decreasing EPT values. The final success criterion was the presence of the dentine bridge on the radiograph after 6 months of treatment. The criteria for determining the failure of the treatment were the presence of spontaneous or nocturnal pain, when performing the cold thermal test – the pain was long-lasting after the removal of the stimulant and increasing EPT values.

All failures occurred within the first three months after treatment and required endodontic treatment. In order to determine the effectiveness of the treatment carried out, with the help of the MedCalc program, the following parameters were calculated: relative risk (RR), confidence interval (CI), statistical value *z*, statistical significance *P* and the number needed to treat (NNT), the results are presented in table 4.

Treatment success rates after both control visits were: group 1 - 48 cases (90.6%; CI 95% [82.4-98.1]), group 2 - 51 cases (96.2%; CI 95% [90.0-100.0]), group 3 - 52 cases (98.1%; 95% CI [92.7-100.0]). From a statistical point of view, no significant difference was obtained between the groups ( $\chi^2=3.422; gl=2; p=0.181$ ).

Table 4. Comparison of treatment results, presence of dental bridge on radiograph after 6 months in study groups

Group 2 - research	Group 1 - control	RR	95% $\hat{I}\hat{I}$ , <i>z</i> statistic, <i>P</i>	NNT
51	48	1.0625	0.9595 - 1.1765, <i>z</i> = 1.166, <i>P</i> = 0.2438	18
Group 3 - research	Group 1 - control	RR	95% $\hat{I}\hat{I}$ , <i>z</i> statistic, <i>P</i>	NNT
52	48	1.0833	0.9856 - 1.1908, <i>z</i> = 1.659, <i>P</i> = 0.0971	13
Group 2 - research	Group 3 - research	RR	95% $\hat{I}\hat{I}$ , <i>z</i> statistic, <i>P</i>	NNT
51	52	1.0000	0.9486 - 1.0542, <i>z</i> = 0.000, <i>P</i> = 1.000	-

Treatment failure rates were for group 1 – 5 cases (9.4%; CI 95% [1.9-17.6]), group 2 – 2 cases (3.8%; CI 95% [0, 0-10.0]), group 3 – 1 case (1.9%; 95% CI [0.0-7.3]). These values also did not reach the threshold of statistical significance ( $\chi^2=3.422; gl=2; p=0.181$ ). At follow-up visit 2, after 6 months, there were no further failures. It is important to note that all failures occurred in patients between 25 and 35 years of age.

The type of clinical evolution of dental caries (acute or chronic) and the type of carious involvement, primary or secondary, did not influence the overall treatment success rates ( $p > 0.05$ ). Between the success rates of the treatment applied according to the location of the carious cavity according to the Black classification, no statistically significant differences were found ( $p > 0.05$ ). They constituted: class I – 36 (94.7%; CI 95% [86.7-100.0]), class II – 107 (94.7%; CI 95% [90.2-99.0] ), class III – 8 (100.0%). The type of treatment performed, direct or indirect pulp capping, also did not influence the percentage of success ( $p > 0.05$ ).

The only parameter that significantly influenced the success rate of the treatment was the age of the patients involved in the research, they were divided into 3 groups: 10-18 years, 18-25 years and 25-35 years. No failures were recorded in the 10-18 years and 18-25 years group, and 8 failures were recorded in the 25-35 years group ( $p = 0.000$ ).

### **3. ANALYSIS OF THE OBTAINED RESULTS**

In our study, we obtained an average success rate of 95%, and the 5% of failures were detected by clinical and paraclinical examinations in the first 3 months, and were subsequently subjected to endodontic treatment. In control group 1 where Ca(OH)<sub>2</sub> was applied, we obtained a success rate of 90.6%, in research group 2 where MTA was applied, we obtained a success rate of 96.2% and in research group 3 where tricalcium silicate was applied, we obtained a success rate of 98.1%.

No statistically significant difference was found between the success and failure rates between the 3 groups. Therefore, the initial hypothesis of the research was not confirmed, that the effectiveness of tricalcium silicate and mineral trioxide aggregate is greater than that of calcium hydroxide, in the conservative treatment of deep dental caries. This proves that all 3 materials are effective and can be successfully used in the treatment of the studied pathology.

The type of clinical evolution of dental caries (acute or chronic) and the type of carious involvement, primary or secondary, did not influence the overall treatment success rates ( $p > 0.05$ ). The type of treatment performed, direct or indirect capping, also did not influence the percentage of success ( $p > 0.05$ ).

In the research carried out, no statistically significant differences ( $p = 1.000$ ) were found between the success rates of the treatment applied according to the location of the carious cavity according to Black classification. Therefore, we can state that the location on the occlusal or approximal surface of the carious cavity does not influence the result of the pulp capping treatment.

In the past, the location of the carious cavity on the proximal surfaces of the tooth served as a contraindication to performing pulp crowns. With the widespread implementation of the absolute isolation of the operating field with a rubber dam, this fact has lost its relevance.

The only parameter that significantly influenced the treatment success rate was the age of the patients involved in the research, they were divided into 3 groups: 10-18 years, 18-25 years and 25-35 years. The 10-18 and 18-25 years groups had no failures and in the 25-35 group were depicted 8 failures.

The high success rate obtained from our study could be explained by the following key points:

- the accuracy of diagnosing the initial pulp status,
- exact compliance with the improved treatment protocol,
- the use of optical magnification to control the removal of carious tissue,
- the use of isolating the tooth treated with a rubber dam from the oral environment,

- respecting the sterility of instruments at all stages of cavity preparation, especially near the pulp chamber,
- the effectiveness of the curative materials used,
- the tightness of the permanent filling,
- young age of the patients (mean age  $19.25 \pm 0.8$  years).

In a 2012 study, the authors found that the clinical success rates of indirect pulp capping with MTA and calcium hydroxide were 93% and 73%, respectively, after 3 months of application, and after 6 months, the success rate of was 89.6% in the group treated with MTA and 73% in the group treated with calcium hydroxide. They also measured the newly formed dentin bridge on radiographs taken 3 and 6 months after treatment. The mean dentine bridge thickness after 3 months was 0.121 mm in the MTA-treated group and 0.136 mm in the calcium hydroxide-treated group. After 6 months, an average thickness of 0.235 mm was recorded in the group treated with MTA and 0.221 mm in the group treated with calcium hydroxide [8].

A 2017 study of 50 patients aged 7 to 9 years demonstrated a 100% success rate at 1-year follow-up using both Biodentine and MTA in direct pulp capping of young permanent molars affected by caries [7].

The authors of a 2017 study, which compared the effectiveness of 3 dental materials in the case of direct pulp capping of young permanent teeth affected by caries, found the following success rates: 86.36% for the group treated with calcium hydroxide, 86.36% for the group treated with MTA, 100% for the group treated with Biodentine [2].

A systematic review and meta-analysis conducted in 2020 by Cushley et al. showed that the success rate of calcium hydroxide pulp capping was 74% at six months, 65% at one year. This success rate is lower than that of direct pulp capping with MTA (91% - 6 months, 86% - 12 months) and Biodentine (96% - 6 months, 86% - 12 months). No statistically significant difference was observed between the success rates of MTA and Biodentine [4].

In a 2021 randomized clinical trial on the efficacy of different calcium silicate materials as pulp capping agents, the following conclusions were reached: the clinical and radiographic success of the direct pulp capping procedure was: 86% for the group treated with MTA+, 80% for Biodentine, 72.5% for Theracal LC, 70% for the Dycal group; after 6 months of treatment, newly formed dentine bridges were detected in all studied groups, but no statistically significant differences were detected ( $p=0.576$ ); regarding the thickness of the newly created dentin bridge after 36 months, statistically significant differences were found between the groups treated with calcium silicates and those with calcium hydroxide ( $p=0.004$ ), and the resulting dentin bridge after the treatment with calcium silicates was thicker and more uniform [9].

A retrospective study from 2023 evaluated the long-term outcome of direct pulp capping with calcium hydroxide of mature teeth with deep caries. The success rate of the maneuver was 100%, 95%, 95%, 86%, and 89% at 1, 5, 10, 20, and 35 years, respectively. The main variable that significantly affected treatment outcome at all follow-up periods was the quality/presence of permanent coronary restoration ( $p < 0.001$ ) [11].

The exact diagnosis of the pulp status can only be obtained by histological examination, but because the studied teeth were not predestined for subsequent extraction, a histological analysis could not be applied in our study [3]. However, Ricucci et al. showed that the clinical diagnosis of normal

pulp/reversible pulpitis correlates with the histological diagnosis of normal pulp/reversible pulpitis in 96% of cases, and in the case of irreversible pulpitis, clinical diagnosis and histological diagnosis correlate in 85% of cases [10].

The diagnosis of the preoperative pulp status was performed clinically and paraclinically according to a unified algorithm for all patients: subjective examination (complaints), objective clinical examination (inspection, probing, percussion, palpation), complementary examination (cold thermal test and EPT), paraclinical examination (Digital OPG). For the inclusion of the respective tooth in the study, the parameters of the examinations indicated above must fall within the specific norm values for deep dental caries, according to the group of the tooth. Otherwise, the dental pulp is considered to be compromised, with signs of irreversible inflammation, and requires a different treatment approach.

In our study, after 3 months from the application of the biological method of treatment for all 3 groups, in the cases considered "success" - 95% of cases - the average EPT values decreased compared to the initial ones, but did not fall within the parameters considered the norm for each group of teeth. In the remaining 5% of cases, increased EPT values were recorded, which indicated the irreversible inflammatory damage of the dental pulp, respectively these cases were considered "failure" and were treated by the pulp extirpation method.

After 6 months from the application of the treatment, the cases considered "successful" were re-evaluated from the point of view of the electrical sensitivity of the dental pulp. The average EPT values obtained, from all 3 groups, fell within the reference norm for each group of teeth, which highlights the positive dynamics of pulp regeneration and tertiary dentin deposition.

No statistically significant difference ( $p>0.05$ ) was detected between EPT values after 3 months among the 3 groups. Between the EPT values after 6 months, a statistically significant difference was determined between the values obtained between groups 1 and 2 ( $p=0.005$ ), between groups 1 and 3 ( $p=0.019$ ), and between groups 2 and 3 ( $p=1.000$ ) there was no found statistically significant difference.

Therefore, we can state that MTA and Biodentine caused a faster restoration of pulpal sensitivity than calcium hydroxide after 6 months of treatment application. At the same time, MTA and Biodentine acted similarly, from the point of view of the dynamics of EPT values.

Studies on the normal values of EPT in different groups of teeth have reported different values for both intact and carious teeth. According to Редина et. al (2009), the EPT values for intact teeth differ depending on the group, and include the following average values: from 2 to 6  $\mu\text{A}$  - the incisor group, from 6 to 15  $\mu\text{A}$  - the premolar group, from 8 to 18  $\mu\text{A}$  - molar group. The authors also reported no differences in gender-related values, but pointed out that values differed in different age groups. According to Любомирский (2010), pulp electrosensitivity in molars in case of acute deep caries ( $29.26\pm 2.57$ ) differs insignificantly from EPT values in case of chronic evolution of deep caries ( $22.12\pm 2.62$ ).

Another criterion for establishing pulpal sensitivity was the performance of the cold thermal test, the results of which correlated with the results obtained after EPT. Thus, the results of the test carried out after 3 months indicated the return of pulpal sensitivity to normal parameters - response similar to the healthy tooth chosen as a reference - in 95% of the investigated teeth from all 3 groups. The remaining 5% showed long-lasting pain after the removal of the stimulus, which indicates



irreversible damage to the dental pulp, respectively these cases were considered "failure". The results of the cold thermal test performed after 6 months, confirmed the return of pulpal sensitivity to normal parameters, in the same 95% of teeth considered "success" from all 3 groups.

Statistically significant differences in cold thermal test results between groups and evaluation periods were not found. This fact correlates with the EPT results and further confirms the effectiveness of all 3 materials studied.

It is well known that the long-term success of pulp capping depends on the presence and quality of the newly formed tertiary dentin bridge (>1mm) [8]. The presence of the newly created dentin bridge served as the final criterion for establishing the success of the treatment performed, as it demonstrates the continuity of pulp regeneration, the deposition of tertiary dentin and, therefore, the preservation of the vitality and functionality of the dental pulp.

Studies have shown that there is little evidence of reparative dentin formation before 30 days after pulp capping. The rate of its formation is the highest in the initial interval of 27 to 48 days - 3.5  $\mu$ /day, reducing considerably in the interval of 49-71 days to 0.74  $\mu$ /day, and in the range of 72-132 days - 0.23  $\mu$ /day [8].

In our study, we evaluated the presence or absence of the newly created dentin bridge using digital radiographs 6 months after treatment. In all 95% of cases considered "success" from all 3 batches, this bridge was radiographically detectable after 6 months, but showed variable thicknesses. This dentinal bridge is visualized as a thin radiopaque layer immediately below the curative filling and demarcates the radiolucency of the pulp chamber from the radiopacity of the curative, isolation or permanent filling.

No significant statistical difference between the groups regarding the presence of the dentine bridge was determined ( $p=0.181$ ). This fact confirms the efficiency of all 3 materials studied in the research, from the point of view of the formation of the tertiary dentin bridge.

The high rate of success of MTA and Biodentine in the case of pulp capping is attributed to their ability to stimulate the formation of the dentin bridge, their antibacterial properties and their ability to seal damaged pulp tissue. Both materials produced a similar pulpal response, this phenomenon may be due to their similar chemical composition (tricalcium silicate is one of the components of MTA), and the by-products released during the setting reaction being also similar. Unlike MTA, Biodentine has a shorter setting time and some studies reported that the thickness of the dentin bridge formed beneath Biodentine was thicker and denser.

### **3.1. Diagnostic algorithm of pulpal status in deep caries lesions**

Characteristics of the pain: pain that is always caused by a stimulus, thermal (cold) or chemical (sweet, sour, salty) that disappears with the removal of the stimulus. If the pain is spontaneous, nocturnal, has a prolonged duration after the removal of the stimulus, the pulp is irreversibly inflamed. If the pain is pulsating or present during mastication and is absent at cold stimuli, we can state the necrosis of the dental pulp. The characteristics of dental pain felt by the patient are presented in figure 8.

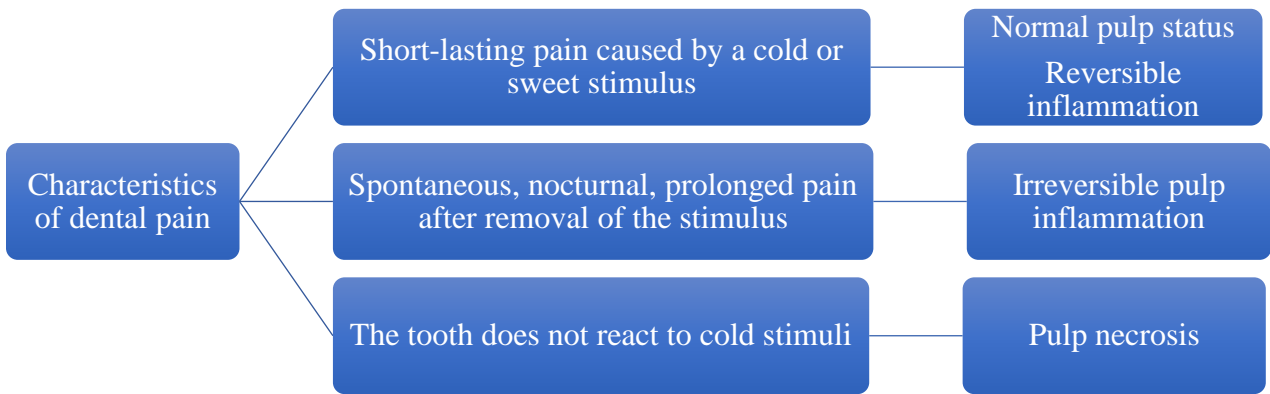


Figure 8. **Characteristics of dental pain felt by the patient**

Clinical examination: The lack of hard dental substance (carious cavity), the change in the color of a tooth surface with the preservation of the integrity of the overlying enamel layer or the presence of an old, defective filling, as seen in figure 9, can be highlighted by inspection. When examining the floor of the cavity, by probing, painful sensitivity is determined and a communication with the pulp chamber should not be detected. Percussion in the axis of the tooth in question should not be painful, and palpation should not reveal pathological changes.



Figure 9. **Clinical aspects of deep carious cavities**

Complementary examination: Pulp sensitivity testing to electrical stimuli (EPT) is performed using a digital pulp-tester first on a healthy tooth, then on the researched tooth. The values should fall within the reference norms for deep caries according to group membership, illustrated in figure 10.

EPT reference values in healthy teeth		
Anteriors 2 - 6 $\mu\text{A}$	Premolars 6 - 15 $\mu\text{A}$	Molars 8 - 18 $\mu\text{A}$

↓

EPT reference values in teeth affected by deep caries		
Anteriors 9 - 13 $\mu\text{A}$	Premolars 15 - 20 $\mu\text{A}$	Molars 22 - 29 $\mu\text{A}$

↓

EPT reference values in teeth affected by irreversible pulpitis		
Anteriors > 14 $\mu\text{A}$	Premolars > 21 $\mu\text{A}$	Molars > 30 $\mu\text{A}$

Figure 10. **EPT reference values according to tooth group membership**

The cold thermal test is performed using the cooling spray, first on a healthy tooth, then on the examined tooth. The tooth affected by deep caries may have a response similar to the healthy tooth

chosen as a reference or may present short-term pain, which disappears with the removal of the stimulus. If the pain is long-lasting, does not disappear with the removal of the stimulus or does not show any response, we assume an irreversible pulpitis or non-vital tooth. The interpretation of the results at cold thermal testing are shown in figure 11.

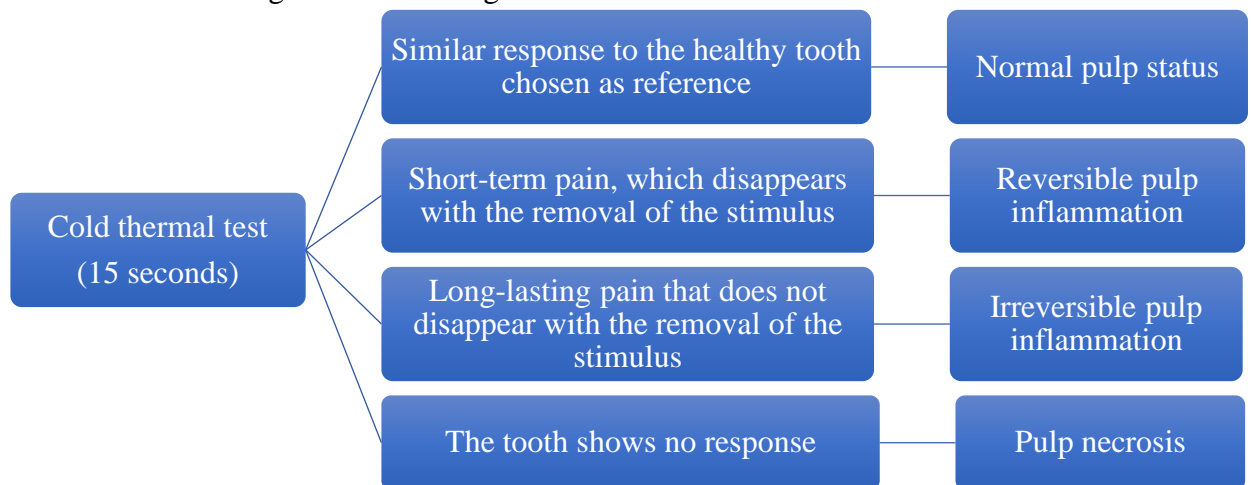


Figure 11. **Interpretation of the results at cold thermal testing**

Radiological examination: An extensive radiolucency (carious cavity) that does not communicate with the pulp chamber or a defective pre-existing filling with radiological signs of secondary or recurrent caries is detected on the radiograph, the periodontal gap is uniform. A series of examples are illustrated in figure 12. In case of a radiolucency that already communicates with the pulp chamber, as illustrated in figure 13, the tooth must undergo endodontic treatment.

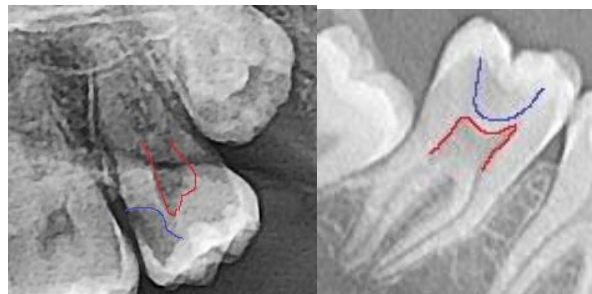
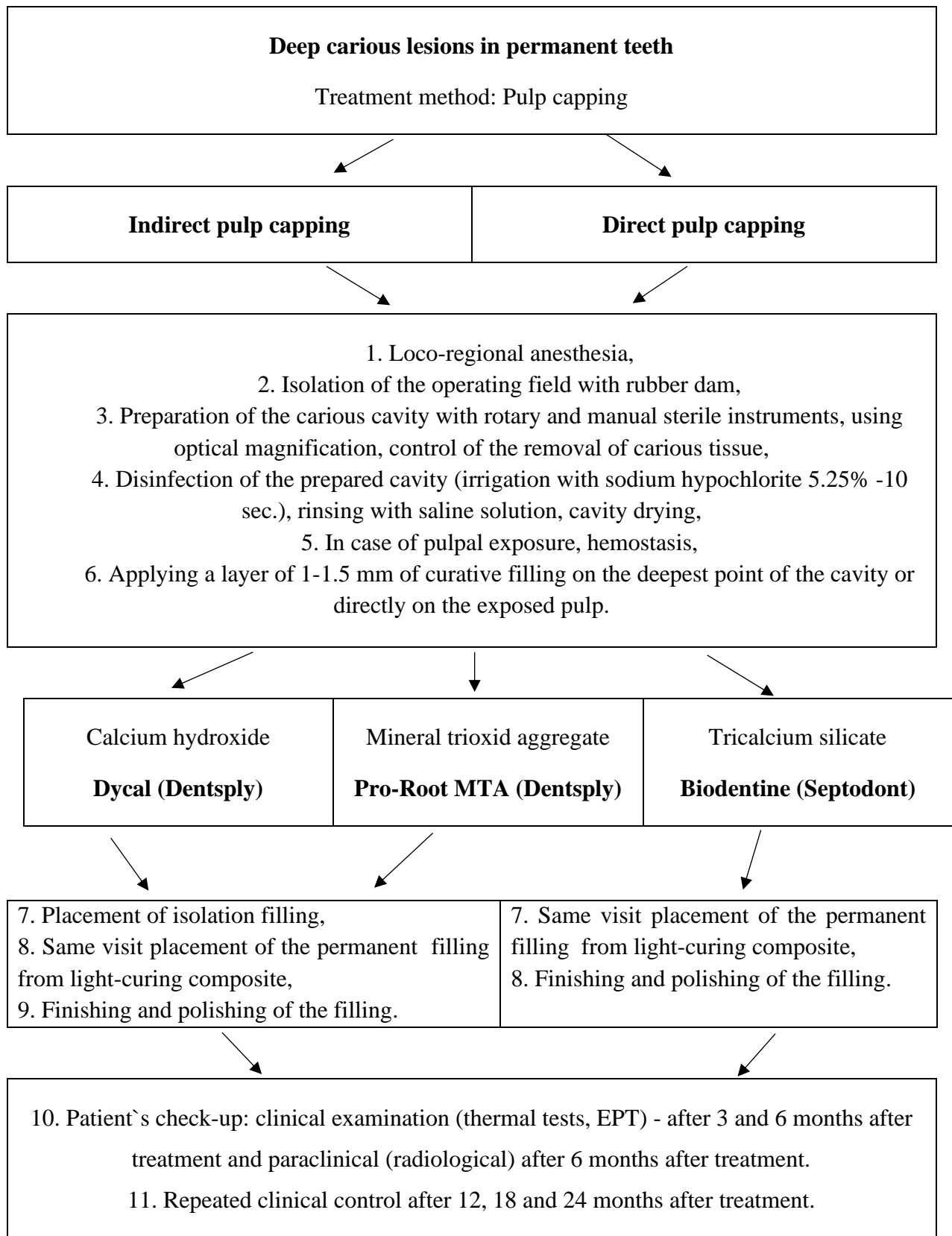


Figure 12. **Radiological aspects of deep carious cavities – radiolucency that does not communicate with the pulp chamber**



Figure 13. **Radiological aspects of deep carious cavities – radiolucency communicating with the pulp chamber**

### 3.2. Treatment protocol of deep carious lesions in permanent teeth



## GENERAL CONCLUSIONS

1. No statistically significant differences between the EPT values after 3 months, was detected between groups 1 and 2 ( $p=0.087$ ), between groups 1 and 3 ( $p=0.087$ ), nor between groups 2 and 3 ( $p=1.000$ ). After 6 months, a statistically significant difference was detected between EPT values both between groups 1 and 2 ( $p=0.005$ ) and between groups 1 and 3 ( $P=0.019$ ), no statistically significant difference was detected between groups 2 and 3 ( $p=1.000$ ). Therefore, we can state that MTA and Biodentine caused a faster restoration of pulpal sensitivity than calcium hydroxide, while MTA and tricalcium silicate acted similarly.
2. No statistically significant differences were established in the cold thermal test results between the 3 groups and both evaluation periods after 3 and 6 months ( $p=0.181$ ). Thus, we can conclude that all 3 pulp capping agents used in the study had a similar influence on the restoration of pulpal sensitivity to cold thermal stimuli.
3. The comparative analysis of the paraclinical parameter evaluated by the presence or absence of the dentine bridge on the radiograph after 6 months demonstrated the absence of a statistically significant differences between the studied groups ( $p=0.181$ ). This demonstrates that the studied remedies contribute equally to the edification of the tertiary dentin bridge.
4. The clinical success rates of the curative dental materials used in pulp capping were similar, without significant statistical differences between the studied groups: calcium hydroxide ( $p=0.181$ ), MTA ( $p=0.181$ ), tricalcium silicate ( $p=0.181$ ). This fact correlates with the clinical and paraclinical results obtained and further confirms the similar efficiency of the 3 materials studied.
5. The developed clinical algorithm increased the accuracy of the diagnosis of the preoperative pulp status and improved the results of the treatment of deep dental caries, contributing to the preservation of the vitality of 95% of the treated teeth.

## PRACTICAL RECOMMENDATIONS

1. The dentist should establish the clinical diagnosis of the initial pulp status according to the proposed algorithm, because it directly influences the result of the applied treatment.
2. For the success of the biological treatment methods of deep dental caries, it is very important to strictly follow the diagnostic algorithm and the work protocol proposed in the research.
3. It is recommended that the clinicians use pulp capping methods to preserve the dental pulp in case of deep caries, as this method has demonstrated high rates of long-term effectiveness, as long as they are performed correctly.
4. The curative materials studied in the research have demonstrated similar clinical effectiveness, therefore the practitioner is entitled to use any of them, taking into account the advantages and disadvantages of each.

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