Abstract

Introduction. In Ukraine, the prevalence of dental caries in children 12 years old reaches 72.7% -91.4%, in children 15 years old - 81.3% -94.3%, while the intensity of caries in children of these age groups ranges from 2, 23 ± 0.21 to 3.71 ± 0.37 and from 3.91 ± 0.39 to 6.18 ± 1.01, respectively. For the physiological formation of dental hard tissues in children, a sufficient level of calcium intake and assimilation with the participation of vitamin D is of great importance. Children for many reasons need to correct calcium metabolic disorders by using combined calcium and vitamin D preparations and a balanced diet, including foods and drinks, enriched with calcium. Materials and methods. Clinical studies were conducted involving 138 children 7-11 years old. To assess the extent of tooth decay, caries was determined by its prevalence, intensity, growth rate of caries, reduction of the growth rate of caries, a biochemical analysis of oral fluid was performed to determine the content of calcium, phosphorus, acid phosphatase, alkaline phosphatase. Children of the main treatment subgroup were offered the proposed Treatment and Preventive Complex, which included correction of eating behavior and drinking regimen (consumption of naturally-table low-mineralized waters without gas with Ca^{2+} content from 50 to 200 mg / l).

Results. When using Treatment and Preventive Complex in children, a low increase in the intensity of decay of permanent teeth, an increase in the level of Ca, a decrease in the activity of acid phosphatase and alkaline phosphatase in the oral fluid were determined in comparison with the control group. Conclusions. Thus, the use of mineral water with a Ca^{2+} content of 50 to 200 mg / l, as a source of natural Ca intake, can be recommended as an aid to the comprehensive prevention of caries in schoolchildren.

Key words: school-age children, dental caries, oral fluid, treatment-and-prophylactic complex, mineral waters,

Introduction

Almost 100% of the adult population of the planet and 60% -90% of school-age children suffer from caries (1). In Ukraine, depending on the region of residence, the prevalence of dental caries in children 12 years old reaches 72.7% -91.4%, in children 15 years old - 81.3% -94.3%, while the intensity of caries in children of these age groups ranges from 2.23 ± 0.21 to 3.71 ± 0.37 and from 3.91 ± 0.39 to 6.18 ± 1.01, respectively (2, 3, 4). An important factor in the pathogenesis of caries is the resistance of dental hard tissues to the effects of organic acids, which are the product of the vital activity of cariogenic microorganisms. The resistance of hard tissues is due to the structure of the enamel and dentin of the teeth, as well as their level of mineralization. The most intensive processes of maturation and mineralization of hard tooth tissues occur in children (5, 6). For the physiological formation of dental hard tissue in children, a sufficient level of Ca intake and assimilation with the participation of vitamin D is of great importance. Unfortunately, most adolescents, for various reasons, are not provided with sufficient Ca - only 1 out of 10 girls and 1 out of 4 boys in the age of 11-15, they receive the necessary amount of Ca with food, that is, children are at high risk for Ca deficiency (7, 8, 9,10). Therefore, children in particular need to correct violations of calcium metabolism through the use of combined preparations of calcium and vitamin D and a balanced diet, including foods and drinks enriched with calcium (8).
However, the use of medications requires a balanced approach to determining the necessary doses, the duration of administration, taking into account the cumulation of vitamin D, the need for regular monitoring of Ca levels in urine and blood (8, 9,11). These provisions initiate the search for other sources of Ca, in particular, natural, providing the possibility of continuous safe use. Numerous studies have proven the role of drinking water as a source of minerals. With drinking water, a person can receive from 5 to 20% of the daily dose of Ca at a concentration of Ca in water at the level of 25-200 mg/l. (12). The revealed effect of the complex of macromolecules (calcium, magnesium, strontium) and their ratio in drinking water on the prevalence of the pathology of the musculoskeletal system, the positive role of drinking water with high mineralization in the process of supporting bone mineral density and the concentration of calcium and magnesium in the blood, indicate the significance of this source of this element into the body (13, 14).

Interesting, from this point of view, it may be the use of natural table mineral waters (MW) with the high calcium content. Recently, in Ukraine, there has been a growing demand for the use of natural tableware MW, which has a certain chemical composition, which determines their effect on the organism. The biologically active components that make up the MW mutually reinforce (or suppress) each other, especially with prolonged intake, and cause a therapeutic effect, which manifests itself as the sum of many secondary indirect reactions (15). At the same time, the role of individual macronutrients of natural MW in their general use as prophylactic and therapeutic agents in dental diseases remains poorly understood, especially in conditions of their combined effect.

Our preliminary experimental studies showed that the daily use of natural canteen MW with a Ca$^{2+}$ content of 50 to 200 mg / l (natural sources of which are located in Ukraine) when modeling experimental caries in monthly rats reduces the prevalence of caries by 20.0-30.0%, and the intensity of the carious lesion is 1.5 times, and also normalizes calcium-phosphorus metabolism and activates the antioxidant defense system (16, 17).

**The purpose** of this study was to assess the possibility of using MW as an adjunct in the comprehensive prevention of dental caries in schoolchildren.

**Material and methods.** Clinical studies were conducted involving 138 children 7-11 years old. Depending on the activity of the carious process, the children were distributed as follows: group I - children with compensated caries - control group; group II - children with compensated caries - the main group; group III - children with subcompensated caries. Group I (n = 22) underwent oral hygiene (OH) and were given recommendations on food and drink regimen. In each of groups II (n = 60) and III (n = 56), control treatment subgroups (CTS) were created, which included 28 children, respectively, they underwent OH, caries treatment, provided food and drink recommendations, and the main treatment subgroups (MTS), in which there were 32 and 28 children, respectively, who underwent the proposed treatment and prophylactic complex (TPC). Patients were randomized using the blind method.TPC provided for the identification and correction of risk factors for dental caries; professional oral hygiene followed by hygienic training and individual selection of oral care products (Ca-based kinds of toothpaste or fluoride contents of 500-1000 ppm), treatment of dental caries and its complications (if necessary); preventive minimally invasive therapy of caries: professional activities (sealing fissures, deep fluoridation, the use of professional gels and varnishes with fluoride) and the appointment of drugs for home use - topical gels based on calcium and fluorine for a month; the use of a local probiotic containing L. keuteri (DSM 17938 and ATCC PTA 5289) correction of eating behavior (by recommending daily use of foods with the highest calcium content and avoiding tooth decay products); drinking regime (the use of natural table water of weakly mineralized water without gas with a Ca$^{2+}$ content of 50 to 200 mg/l - “Berezovskaya” non-carbonated, “Aqua-vita”, “Morshinskaya” non-carbonated, “Karpatskaya” spring non-carbonated, “Mirgorod” non-carbonated).

Clinical examination of children was carried out according to a standard scheme using generally accepted methods at the beginning of the study and after 12 months. Following the age of children and WHO recommendations, prevalence, intensity (the index of the intensity of carious lesions of permanent teeth + the index of the intensity of carious lesions of temporary teeth, ILPT+ ILTT), and an increase in the intensity of caries were determined to assess the degree of dental caries lesion (18,19). The reduction in caries growth was
determined by Sakharov E. B. (1984) (18). Children underwent biochemical analysis of oral fluid (OF) to determine the content of Ca, phosphorus (P), acid phosphatase (ACPH), alkaline phosphatase (ALPH). OF in children was collected for 7-10 minutes in the morning at rest in sterile disposable containers with a volume of 5-10 ml, which were transported to the laboratory in a thermal container with cooling elements for 3 hours. A biochemical study of OF was performed on a Labline-100 automated biochemical and enzyme immunoassay analyzer, WestMedika (Austria). The study of the concentration of total Ca in OF was carried out using a set of ready-made reagents Calcium-Arsenazo, BioSystems (Spain), concentrations P were performed using the UV photometric method using a set of ready-made reagents Phosphorus Aquapapid, Human (Germany), and ALPH activity was performed using a set of ready-made reagents Alkaline Phosphatase, Human (Germany), ACPH activity - by a set of ready-made reagents Acid Phosphatase, BioSystems (Spain) (20).

The studies were carried out in compliance with the principles of bioethics and the rights of the patient in accordance with the Helsinki Declaration (2000.) and the Fundamentals of Ukrainian legislation on health care (1992). The materials were examined by the bioethics commission of the NMAPE named after P.L. Shupik (Minutes of the meeting of the commission on ethics No. 11 of 11/19/2018).

Statistical analysis of the data included the calculation of mean values, standard deviation, and mean error. To assess the significance of differences between samples subjected to the normal distribution law, Student t-test (p) was used. Statistical calculations were performed in the SPSS 17.0 software environment (IBM SPSS Statistics 17 Free PC Software FullVersion, USA) and MS Excel 2010 (license number K93660931 2016).

### Results

At the beginning of the study, the prevalence of dental caries in children of group I was 59.1% with an intensity (ILPT+ ILTT) of 1.84 ± 0.19. In children from CTS of the II group, the prevalence of caries was determined at the level of 96.4% according to its intensity of 3.14 ± 0.24; in children from MMS in this group the corresponding indicators were 93.8% and 2.93 ± 0.26. The indicators of tooth caries intensity in children from CTS and MMS of group II significantly (p <0.05) differ from such children of group I, while there is no significant (p>0.05) difference between the indicators of children from CTS and MMS. A more detailed analysis showed that the intensity of caries in the first permanent molars in children of these groups was at the same level: group I - 0.45 ± 0.18; II CTS group - 0.54 ± 0.16, II MMS - 0.62 ± 0.19.

The intensity of damage to temporary teeth (ILTT) - 1.38 ± 0.29, 2.57 ± 0.34 and 2.24 ± 0.34, respectively, indicates a more specific weight in these children is the intensity of caries of temporary teeth. At 100% prevalence of dental caries in children of group III, the index of intensity of dental caries (ILPT+ ILTT) in children with CTS and MMS was 6.79 ± 0.26 and 6.64 ± 0.39, respectively. With such a high degree of tooth damage in children of both subgroups of the III group, in contrast to children of the II group, the intensity of caries of temporary teeth (ILTT) is more than 2 times, the intensity of damage to the first permanent molars is almost 3 times (Table 1).

### Table 1. Caries intensity in children 7-11 years before treatment

<table>
<thead>
<tr>
<th>Groups</th>
<th>ILPT+ ILTT</th>
<th>ILTT</th>
<th>ILPT</th>
<th>ILPT The first permanent molars</th>
</tr>
</thead>
<tbody>
<tr>
<td>I, n=22</td>
<td>1.84±0.19</td>
<td>1.38±0.29</td>
<td>0.46±0.18</td>
<td>0.45±0.18</td>
</tr>
<tr>
<td>II CTS, n=28</td>
<td>3.14±0.24</td>
<td>2.57±0.31</td>
<td>0.57±0.17</td>
<td>0.54±0.16</td>
</tr>
<tr>
<td>II MMS, n=32</td>
<td>2.93±0.26</td>
<td>2.24±0.34</td>
<td>0.68±0.20</td>
<td>0.62±0.19</td>
</tr>
<tr>
<td>III CTS, n=28</td>
<td>6.79±0.26</td>
<td>5.00±0.39</td>
<td>1.79±0.35</td>
<td>1.57±0.21</td>
</tr>
<tr>
<td>III MMS, n=28</td>
<td>6.64±0.39</td>
<td>5.18±0.48</td>
<td>1.46±0.34</td>
<td>1.32±0.30</td>
</tr>
</tbody>
</table>

After 12 months, the intensity of decay of permanent teeth (ILPT) in children of the MMS group II, where the proposed TPC was applied, increased only from 0.68 ± 0.20 to 0.86 ± 0.27, in children of group I this indicator increased from 0.46 ± 0.18 to almost the same level - 0.81 ± 0.24. In contrast, children with CTS of group II - from 0.57 ± 0.17 to 1.12 ± 0.24, that is, 2 times (p <0.05). Thus, in children of MMS of the group II, a low increase in the intensity of caries of permanent teeth was determined - 0.18, in contrast to children of Group I and CTS of the group II, in which they found an average level of increase in the intensity of caries, the indices of which were 0.35 and 0, respectively, 55. The reduction of caries in the MMS of the group II in relation to the CTS was 67.3%, to I control group - 48.6%. A positive effect of the proposed TPC was observed in children with acute respiratory infections of the III group, in which dental caries were determined at a high level. The intensity of caries of permanent teeth in children from CTS after 12 months was 2.36 ± 0.22,

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which was significantly ($p < 0.05$) more than in children of MMS, $1.65 \pm 0.23$. The increase in caries of permanent teeth in children with CTS was 0.57, while at the same time as in children with MMS it was three times less - 0.19. The reduction of caries in MMS to CTS was 66.7%, to AI - 45.7% (Table 2).

Table 2. The intensity of caries in children 7-11 years after 12 months after treatment

<table>
<thead>
<tr>
<th>Groups</th>
<th>ILPT± ILTT</th>
<th>ILPT</th>
<th>ILPT the first permanent molars</th>
<th>Caries increase</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>2.38±0.36</td>
<td>1.56±0.43</td>
<td>0.81±0.24</td>
<td>0.81±0.24</td>
</tr>
<tr>
<td>II CTS</td>
<td>3.54±0.39*</td>
<td>2.42±0.36</td>
<td>1.12±0.24</td>
<td>1.12±0.24</td>
</tr>
<tr>
<td>II MMS</td>
<td>2.64±0.26</td>
<td>1.78±0.37</td>
<td>0.86±0.27</td>
<td>0.78±0.27</td>
</tr>
<tr>
<td>III CTS</td>
<td>6.00±0.51*</td>
<td>3.64±0.54</td>
<td>2.36±0.22*</td>
<td>2.08±0.19*</td>
</tr>
<tr>
<td>III MMS</td>
<td>4.94±0.42</td>
<td>3.29±0.51</td>
<td>1.65±0.23</td>
<td>1.39±0.27</td>
</tr>
</tbody>
</table>

Note. 1. * - $p < 0.05$ - significance of differences between children CTS and MMS after 12 months.

The occurrence and development of dental caries in children are accompanied by certain changes in the composition and biochemical properties of the oral fluid. Oral fluid can serve as an alternative diagnostic fluid compared to blood. In turn, by determining changes in the biochemical parameters of the oral fluid, it is possible to assess the condition of the organs and tissues of the oral cavity, as well as assess the impact of the developed treatment and prophylactic methods and approaches. Researchers pay special attention to determining the content of Ca and P, which provides the mineralizing properties of the oral fluid, namely, the formation and maintenance of the mineral composition of tooth tissues, and, above all, enamel (21, 22, 23, 24). The calcium content in the oral fluid in a certain way is affected by the state of phosphorus-calcium metabolism in the child's body, which is due to a sufficient level of Ca intake and assimilation (7).

A biochemical study showed a low initial Ca content in the PP of children of all groups, the P content was fixed within the normal range, which indicates an insufficient mineralizing potential of the oral fluid. 12 months after the use of TPC, we determined an increase in the Ca content in the OF of children of all subgroups and an unreliable ($p > 0.05$) decrease in the content of P. But, unlike group I and CTS of groups II and III, a significant ($p < 0.05$) an increase in the Ca content in OF ($0.98 \pm 0.03$ mmol/l and $1.11 \pm 0.15$ mmol/l, respectively) after application of the TPC, and these indicators are also significant ($p < 0.05$) differed from the CTS indices ($0.69 \pm 0.08$ mmol/l and $0.76 \pm 0.06$ mmol/l) (Table 3).

Table 3. The content of calcium and phosphorus in the oral fluid of children 7-11 years before treatment and after 12 months

<table>
<thead>
<tr>
<th>Groups</th>
<th>Indicators</th>
<th>I</th>
<th>II</th>
<th>III</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Ca, mmol / l before TPS</td>
<td>0.68±0.04</td>
<td>0.59±0.03</td>
<td>0.66±0.10</td>
</tr>
<tr>
<td></td>
<td>P, mmol / l before TPS</td>
<td>0.73±0.15</td>
<td>0.69±0.08*</td>
<td>0.98±0.03</td>
</tr>
<tr>
<td></td>
<td>P, mmol / l after TPS</td>
<td>4.67±0.48</td>
<td>4.77±0.23</td>
<td>5.04±0.35</td>
</tr>
<tr>
<td></td>
<td>P, mmol / l after TPS</td>
<td>4.58±0.44</td>
<td>4.19±0.39</td>
<td>4.47±0.32</td>
</tr>
</tbody>
</table>

Note. 1. p – the reliability of the difference in scores in children of each subgroup before and after the TPC; 2. * – $p < 0.05$ – the reliability of the differences between children CTS and MMS after the TPC.

It is known that acid phosphatase and alkaline phosphatase participate in calcium-phosphorus metabolism; they separate phosphate from phosphoric acid compounds, thereby providing mineralization of bones and teeth (22). In the presence of inflammatory diseases, the activity of these enzymes increases sharply (21, 25). At the beginning of the study, an increased activity of ACPH in OF was established in children of CTS and MMS of groups II and III compared with children of group I.

Regarding alkaline phosphatase, compared with the indicator of children of group I, an increase in the activity of this enzyme was observed in children from MMS of group II and from CTS and MMS of group III. 12 months after the use of TPS in children from MMS of groups II and III determined a significant ($p < 0.05$) decrease in the activity of ACPH ($0.11 \pm 0.02$ mmcat/l and $0.10 \pm 0.02$ mmcat/l) and ALPH ($0.35 \pm 0.03$ mmcat/l and $0.35 \pm 0.04$ mmcat/l, respectively) in OF compared with the results in children with CTS of these groups (ACPH - $0.28 \pm 0.02$ mmcat/l and $0.26 \pm 0.01$ mmcat/l, ALPH - $0.32 \pm 0.01$ mmcat/l and $0.59 \pm 0.09$ mmcat/l, respectively) (Table 4).
The relationship between the micro- and macroelement composition of MW in certain regions of Ukraine and the main dental diseases in children living in these territories indicates the importance of this factor in the development of caries (15, 30, 31).

In recent years, 285 publications have devoted the problem of the relationship between endemic caries and the salt composition of drinking water, but most of them are devoted to the influence of fluoride in the water on the development of dental caries (32). Children living in regions with an optimal or excessive concentration of this halogen in water have significantly lower caries compared to children living in regions with insufficient fluoride (6, 33, 34). The prophylactic effect of physiological concentrations of fluoride in drinking water on the prevalence of caries in children has been proven (35, 36). On the other hand, an excessive concentration of fluoride in drinking water leads to the occurrence of fluorosis (34). Significantly less scientific work is devoted to other macro- and microelements of drinking water. Despite the low intake of Ca and other trace elements with drinking water, which hypothetically can affect the mineralization processes, most researchers are skeptical about the possible role of the mineral composition of drinking water in the prevalence of caries (32).

However, a survey of 440 children 7, 12 and 15 years old in different settlements of Transcarpathia (v. Kvasy., G. Svalyava., G. Rakhov), with climatic and geographical differences in the macro- and microelemental composition of drinking MW, indicates that the prevalence and caries intensity to a certain extent depends on the composition of drinking water (37).

The largest number of studies is devoted to the possibility of local application of MW of Transcarpathian region in the comprehensive prevention of caries, due to the presence of a significant amount of fluorine and Ca in them (especially in carbonic and iodine-bromine waters). According to AM. Potapchuk (1991), the use of highly mineralized “Paseka” MW as a local prophylactic together with controlled hygiene of PR significantly reduces the solubility of the surface layers of enamel, the yield of Ca and P ions, increases the enamel resistance and the ability to remineralize (38).

The analysis shows that when using Transcarpathian mineral waters in the form of hydro
procedures, the buffer capacity of OF increases, and the permeability of tooth enamel and the molar ratio decrease and stabilize. The bactericidal effect on plaque microflora deserves special attention (39).

Conclusions. Thus, the use of MW with a Ca$^{2+}$ content of 50 to 200 mg/l as a source of natural Ca intake can be recommended as an aid to the comprehensive prevention of caries in schoolchildren. Given the importance of endogenous prevention of dental caries in children, the use of natural table MW with a Ca content of 50 to 200 mg/l as a safe prophylactic is relevant and requires additional study since it will significantly reduce the need for drug therapy and significantly increase the effectiveness of treatment without contraindications.

Authors declare no conflict of interest

References


17. Trubka IO. Investigation of the influence of mineral waters with vitamin D$_3$ on biochemical parameters in rats with experimental caries. Visn


