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Citation for the manuscript:

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Submitted to: BMC pediatrics

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Acidity and *in vitro* effects on dental hard tissues of pharmaceutical preparations used in paediatric cardiology

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**Abstract**

**Background:** The knowledge of oral health effects caused by long-term medication in medically compromised children is sparse. Besides the effects on salivary secretion, pharmacotherapy may also act directly on the dental hard tissues, with dental caries and/or erosive lesions as possible outcomes of their acid and fermentable sugar content. **Methods:** Thirteen pharmaceutical preparations commonly used on a long-term basis in paediatric cardiology were selected. The endogenous pH of water solutions of tablets, capsules, and liquid medicines were measured with a pH meter. The titratable acidity and the dissolution of calcium and phosphate after immersion of tooth specimens were quantified for preparations with an endogenous pH below 5.5. **Results:** The endogenous pH values varied between 3.03 and 9.02. Six of the 13 preparations (46%) had an endogenous pH below the critical value for enamel dissolution (pH 5.5). The captopril (12.5 mg) tablet water solution had the lowest pH while the propranolol hydrochloride mixture displayed the highest titratable acidity. The highest dissolved calcium and phosphate was displayed for captopril (12.5 mg) tablet water solution followed by acetylsalicylic acid (75 mg) tablet water solution. **Conclusion:** It is concluded that some pharmaceutical preparations that are commonly used on a long-term basis in paediatric cardiology may pose a hazardous threat to dental hard tissues due to their acidity.

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Background
The knowledge of oral health effects caused by long-term medication in medically compromised children is sparse [1]. Besides the effects on salivary secretion, pharmacotherapy may also act directly on the dental hard tissues, with dental caries and/or erosive lesions as possible outcomes of their acid and fermentable sugar content [2, 3].

To maintain chemical stability, control tonicity, and physiological compatibility in pharmaceutical preparations, acids are frequently used as buffering agents [1]. Often, medicines are administered to children in the liquid dosage form to facilitate administration [4]. But also tablets and capsules are used and to facilitate swallowing in small children crushed tablets and the contents of capsules often are given separately or in sugary products. Fermentable sugars such as sucrose can also be added in paediatric pharmaceutical preparations to disguise unpleasant taste, and thereby, facilitate compliance. However, the presence of sugars in medicines may result in a pH drop in the oral biofilm as a result of their fermentation by acid-producing bacteria [5], and the acid content may help to prolong the pH drop after a sugar challenge. Increased acidity in the biofilm changes homeostasis with a selection towards bacteria associated with dental caries [6]. Acids in medicines may also act directly on dental hard tissues and increase their solubility [7, 8].

Many medicines used on a long-term basis may have a low pH, high acidity, and contain sugar [7]. In a study on the erosive potential of 97 medicines used regularly and long term by children, 57% had an endogenous pH below 5.5 [4]. A pH below 5.5 is considered critical for enamel demineralisation and increases the cariogenic and erosive potential. Medicines used for the cardiovascular system had the lowest mean endogenous pH and it was 4.05.

Children with severe congenital heart disease are a steadily growing patient group due to increased survival as a result of technical development and early interventions. However, children with very complex diseases frequently require regular long-term medication after surgery [9]. The need for regular medicine intake during the day, and before bedtime may consequently constitute a potential threat to the maintenance of a good oral health in these children.

The finding that children with severe congenital heart disease display a severe caries situation, especially in the primary dentition, with the highest prevalence in children on digoxin medication administered in a sugar-containing syrup [10], calls for an exploration of oral health effects of medicines. Microbiological findings indicate a more acidic oral environment in this group of children [11].

The aim of the present study was to investigate the
endogenous pH and titratable acidity of and the dissolution of calcium and phosphate from dental hard tissues submitted to pharmaceutical preparations used regularly and on a long-term basis in paediatric cardiology.

**Methods**

*Pharmaceutical preparations, sample preparation, and pH measurement*

Thirteen pharmaceutical preparations commonly used in paediatric cardiology were selected. The selected pharmaceutical preparations were solid tablets (n = 7), capsules (n = 2), and mixtures (n = 4). Citric acid (10 mM) was used as control. Tablets and contents of capsules were crushed in a mortar and dissolved in 10 ml distilled water. For liquid preparations, 10 ml sample was taken. The endogenous pH of the water solutions of tablets, capsules, or liquid preparations were measured twice using the Mettler seven easy pH meter with the pH-electrode DG 115-SC (Mettler Toledo, 8603 Schwerzenbach, Switzerland).

*Titration*

A sodium hydroxide (NaOH) solution was prepared from the content of an ampoule of J. T Baker 4689 dilut-it (Mallinckrodt, Baker BV, Deventer, Holland). The content of the ampoule was diluted with MQ distilled water according to the manufacturer’s instructions to achieve a total volume of 1.0 l. From this, a 10 times dilution was made to achieve 0.1 M of NaOH. Each sample was titrated by adding 0.01 ml aliquots of NaOH solution by using a titrator. The pH value achieved for each 0.01 ml NaOH added was read with the pH meter and recorded. The titration was performed until a pH value of 7.0 was reached. Samples with an endogenous pH value ≥ 5.5 were not titrated. All titrations were performed on two different occasions.

*Loss of calcium and phosphate after immersion of enamel in pharmaceutical preparations*

For pharmaceutical preparations with a pH below 5.5, the dissolution of calcium and phosphate after immersion of tooth specimens were quantified. Specimens were prepared from the mid-part crown of healthy primary canines extracted for orthodontic reasons and with no cracks in the enamel (n = 8). Prior to the extractions the patients and their parents were informed that the teeth should be used for research purposes and consent were obtained. The teeth were embedded in Epofix (Struers, Ballerup, Denmark), and 3 transverse slices (thickness, 80 µm) from each crown were cut using a low-speed microtome (Leitz, Wetzlar, Germany) under water cooling. Prior to the experiment, the tooth specimens were conditioned by immersion in a mineral solution (1 mM CaHPO₄ + 1 mM NaF, pH 7) overnight at room temperature. Tablets were crushed in a mortar, diluted in 2 ml MQ-water, and rocked overnight in room
temperature. Calcium was measured by atomic absorption (Varian Techtron AA-6DB, Varian Techtron Pty Ltd, Melbourne, Australia) at 422.7 respectively 285.2 nm with an acetylene flame, with a standard curve in the range of 0.8–1.5 mM total calcium. Phosphate was determined spectrophotometrically (Ultrospec 100 pro, Amersham Biosciences, Biochrom Ltd, Cambridge, England) at 700 nm with a standard curve in the range of 2.2–5.7 mM total phosphate. First, the baseline calcium and phosphate content of medicine solutions were measured in 100 µl of each sample. Thereafter, 500 µl of each medicine solution was transferred into cell cultivation chambers and 2 chambers were filled with each medicine (Nucleon surface, Cat. no. 142475, Nalge Nunc International, Rochester, USA). Each well had a diameter of 15 mm. One tooth disc was put into each chamber, and the plate was placed in 37°C and rocked. One hundred µl were collected from each chamber after 30 minutes of immersion and analysed. The experiment was duplicated in two separate runs.

Results
The endogenous pH values varied between 3.03 and 9.02 (Table 1). Six of the 13 tested pharmaceutical preparations (46%) had an endogenous pH below 5.5. The lowest pH was recorded for the captopril (12.5 mg) water solution while the propranolol hydrochloride mixture displayed the highest titratable acidity (Fig 1). The highest dissolved calcium and phosphate was displayed for captopril (12.5 mg) tablet water solution 1.9 mM and 1.1 mM, respectively. From the acetylsalicylic acid (75 mg) tablet water solution 1.6 mM calcium and 0.7 mM phosphate were dissolved (Table 2 and Fig 2).

Discussion
The present study was performed in order to increase the knowledge of possible effects on dental hard tissues caused by pharmaceutical preparations used regularly and on a long-term basis in children with severe heart disease. As the composition of teeth is variable, due to genetic influences, environmental conditions and post-eruptive maturation and dentin sclerosis such differences may lead to large variations in their response under acidic challenges. The tooth specimens were conditioned in a 1 mM calcium phosphate medium in order to standardize the specimens prior to measurements. Fluoride was added to the medium as we had no control of the fluoride exposure prior to the extractions. Due to the in vitro design, the magnitude of the clinical dental effects of low pH and titratable acidity can only be speculated on but are depending on the intimate interplay between chemical factors such as endogenous pH and titratable acidity, biological factors such as the properties of saliva, and behavioural factors like diet, oral hygiene, vomiting, and regular medication [13].
The results display that some pharmaceutical preparations pose a threat to oral health, with caries and erosions as possible outcomes. One of the most important factors in the defence against dental erosion and caries is the quality and quantity of saliva. With its buffering capacity, saliva protects the oral cavity from damaging pH changes by neutralizing acids in plaque, diluting acids, transporting the acid away from the oral cavity, and contributing minerals to the remineralisation process [14, 15]. In low salivary secretion, the buffering capacity of saliva is reduced mainly because of the reduction of bicarbonate, which aids in the neutralization of acids.

A strong correlation between xerostomia and pharmacological treatment has been shown [16]. Xerostomia increases the risk of developing both caries and dental erosion [17]. The medication in children with severe heart disease is often regular, long-term, and multiple. In order to treat the build-up of excess fluid in the body that occurs in congestive heart failure and to relieve the over-load on the heart, diuretics are the first treatment of choice. Solely or as a complement treatment ACE-inhibitors and or glycosides as digoxin are also used. We showed that 29% of a group of children with severe heart disease on medication for heart failure displayed reduced salivary secretion rate, especially in early ages, compared to healthy age and gender-matched controls [11]. If the findings from the present study are taken together with the fact that some of these medicines also may contain fermentable sugars such as sucrose, it is clear that they may pose a hazardous threat to the dental hard tissues, particularly in individuals with reduced salivary secretion.

Minerals in dental hard tissue are dominated by calcium and phosphate mainly organised in hydroxyapatite crystals with a critical dissolution pH of 5.5 [5]. Any substance with an endogenous pH below this value may cause ionic dissolution of the hard tissue, resulting in caries and erosion [18]. In dental caries, there is localized destruction of the tooth surface covered by dental plaque. The process is initiated by decalcification of the enamel, followed by enzymatic lysis of organic structures, and finally cavity formation. Dental erosion is a multifactorial condition often defined as ‘the dissolution of the tooth by acids when the surrounding aqueous phase is unsaturated with respect to tooth mineral [19]. The importance of endogenous pH for mineral loss is illustrated in our in vitro finding, which shows that the loss of calcium and phosphate after immersion of the tooth specimens was highest for the captopril (12.5 mg) tablet water solution, which also displayed the lowest endogenous pH of the tested pharmaceutical preparations. The mixture of the non-selective beta-blocker propranolol hydrochloride \( \text{C}_{16}\text{H}_{21}\text{NO}_2 \cdot \text{HCl} \) with an endogenous
pH of 3.84 displayed a higher titratable acidity than citric acid, while the mineral loss, especially for calcium, was much less than for the citric acid solution, which had a pH of 2.55. Each acid anion has a different strength of calcium complexation, which is dependent on the structure of the molecule and how easily it can attract the calcium ion. Thus, acids such as citric acid have double actions on the hydroxyapatite crystals: the hydrogen ion directly attacks the crystal surface and may also complex with calcium and remove it from the crystal surface [20].

Because many children with severe heart disease already lead a heavy state of life with feeding problems, long periods of hospitalisation, and life-long medication, caries and dental erosions are unfavourable factors that may pose an additional burden. Compromised oral health in this group may also be a reason for delay of scheduled heart surgery. Thus, it is of great importance that paediatric cardiologists and paediatric dentists are aware that the pharmaceutical preparations commonly used in this group may constitute challenges for dental hard tissues with both caries and erosions as possible outcomes. The importance of close cooperation between the medical and the dental care with referrals to the paediatric dentist before the child is 1 year of age is underlined. A preventive approach for oral health is strongly recommended.

**Conclusion**

It is concluded that some pharmaceutical preparations that are used on a long-term basis in paediatric cardiology may pose a hazardous threat to dental hard tissues due to their acidity.

**Competing interests**

Linda Rosén has no competing interests to declare.

Annika Rydberg has no competing interests to declare.

Inger Sjöström has no competing interests to declare.

Ted Lundgren has no competing interests to declare.

Christina Stecksén-Blicks has no competing interests to declare.

**Author contributions**

Linda Rosén and Christina Stecksén-Blicks designed the study. Linda Rosén and Inger Sjöström performed the laboratory work. Ted Lundgren contributed with preparation of the tooth specimens. Linda Rosén drafted the manuscript. Annika Rydberg, Ted Lundgren, and Christina Stecksén-Blicks critically revised the manuscript.
Acknowledgement
We acknowledge the Swedish Heart-Children’s Association (Hjärtebarnsfonden) who financially supported the study.

References


Table 1. Endogenous pH and titratable acidity to reach pH 7.0 for pharmaceutical preparations. Pharmaceutical preparations with pH >5.5 were not titrated. Citric acid (10 mM) was used as the control.

<table>
<thead>
<tr>
<th>Dosage form, generic name/substance, (trade-name), dose</th>
<th>pH</th>
<th>Titratable acidity (μM)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Citric acid, 10 mM (control)</td>
<td>2.55</td>
<td>300</td>
</tr>
<tr>
<td>Tablet, captopril (Captopril), 12.5 mg</td>
<td>3.03</td>
<td>54</td>
</tr>
<tr>
<td>Capsule, captopril (Captopril), 2 mg</td>
<td>3.55</td>
<td>9</td>
</tr>
<tr>
<td>Capsule, captopril (Captopril), 0.5 mg</td>
<td>3.95</td>
<td>3</td>
</tr>
<tr>
<td>Mixture, propranolol hydrochloride (Propranolol ), 1 mg/ml</td>
<td>3.84</td>
<td>385</td>
</tr>
<tr>
<td>Tablet, acetylsalicylic acid (Tromblyl ), 75 mg</td>
<td>3.97</td>
<td>115</td>
</tr>
<tr>
<td>Tablet, furosemide (Furix ), 20 mg</td>
<td>5.40</td>
<td>50</td>
</tr>
<tr>
<td>Mixture, potassium citrate (Kajos ), 33 mg K⁺/ml</td>
<td>5.89</td>
<td>n a</td>
</tr>
<tr>
<td>Tablet, spironolactone (Spironolakton ), 25 mg</td>
<td>6.80</td>
<td>n a</td>
</tr>
<tr>
<td>Tablet, metoprolol (Seloken ), 50 mg</td>
<td>6.92</td>
<td>n a</td>
</tr>
<tr>
<td>Tablet, enalapril (Enalapril ), 5 mg</td>
<td>7.11</td>
<td>n a</td>
</tr>
<tr>
<td>Mixture, digoxin (Lanoxin ), 50 μg/ml</td>
<td>7.14</td>
<td>n a</td>
</tr>
<tr>
<td>Tablet, warfarin (Waran ), 2.5 mg</td>
<td>7.56</td>
<td>n a</td>
</tr>
<tr>
<td>Mixture, furosemide (Impugan ), 10 mg/ml</td>
<td>9.02</td>
<td>n a</td>
</tr>
</tbody>
</table>

n a not analyzed
Table 2. Dissolved calcium and phosphate from tooth specimens in pharmaceutical preparations. Effects of pharmaceutical preparations with pH >5.5 were not analysed. The tooth specimens were pretreated with CaHPO$_4$ + 1 mM NaF overnight prior to immersion. Citric acid (10 mM) and MQ-water as controls.

<table>
<thead>
<tr>
<th>Dosage form/generic name/substance/dose</th>
<th>pH</th>
<th>Phosphate (mM) Baseline</th>
<th>After 30 min of immersion at 37°C</th>
<th>Calcium (mM) Baseline</th>
<th>After 30 min of immersion at 37°C</th>
</tr>
</thead>
<tbody>
<tr>
<td>MQ-water (Control)</td>
<td>-</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>Citric acid 10mM (Control)</td>
<td>2.55</td>
<td>0.0</td>
<td>1.2</td>
<td>0.0</td>
<td>4.6</td>
</tr>
<tr>
<td>Tablet captopril (12.5 mg)</td>
<td>3.03</td>
<td>0.0</td>
<td>1.1</td>
<td>0.0</td>
<td>1.9</td>
</tr>
<tr>
<td>Capsule captopril (2 mg)</td>
<td>3.55</td>
<td>0.1</td>
<td>0.6</td>
<td>0.0</td>
<td>1.3</td>
</tr>
<tr>
<td>Capsule captopril (0.5 mg)</td>
<td>3.95</td>
<td>0.1</td>
<td>0.3</td>
<td>0.0</td>
<td>0.3</td>
</tr>
<tr>
<td>Mixture propranolol hydrochloride (1 mg/ml)</td>
<td>3.84</td>
<td>0.0</td>
<td>0.5</td>
<td>0.1</td>
<td>0.9</td>
</tr>
<tr>
<td>Tablet acetylsalicylic acid (75 mg)</td>
<td>3.97</td>
<td>0.0</td>
<td>0.7</td>
<td>0.1</td>
<td>1.6</td>
</tr>
<tr>
<td>Tablet furosemide (20 mg)</td>
<td>5.40</td>
<td>0.1</td>
<td>0.1</td>
<td>0.1</td>
<td>0.1</td>
</tr>
</tbody>
</table>
Figure legends

Fig 1. Titratable acidity from endogenous pH to reach pH 7.0 for pharmaceutical preparations. Citric acid (10 mM) was used as control.

Fig 2. Dissolved calcium and phosphate from tooth specimens in pharmaceutical preparations. The tooth specimens were conditioned with CaHPO$_4$ + 1 mM NaF overnight prior to immersion. Citric acid (10 mM) was used as control.
Fig 1.
Fig 2.