Ferum and Magnesium Levels in the Plaque and Saliva of Children with Dental Black Stain Who Consume UHT Milk

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Abstract
This study measured the levels of ferum and magnesium in the plaque and saliva of children with dental black stain. The subjects were 16 children aged 4 to 8 years with dental black stain. Samples consisted of the black stain plaque and saliva of children with dental black stain. Ferum and magnesium levels were measured using atomic absorption spectrophotometry. The results showed ferum levels of 30.50% (plaque) and 7.68 ppm (saliva) and magnesium levels of 0.23% (plaque) and 1.59 ppm (saliva). Higher ferum levels were found in the saliva of children with dental black stain who consumed ultra-high-temperature milk.

Keywords: Dental black stain, dental plaque, ferum, magnesium, saliva.

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Introduction
Extrinsic discoloration is the discoloration of the teeth caused by any substance or material, metal or nonmetal, that is attached to the outer surface of the teeth or to the pellicle.¹ Extrinsic discoloration that occurs on the tooth varies in color from dark red, orange, yellow, yellowish-brown, and green to brownish-gold, purplish-black, brown, and black.¹ Discoloration that is black in color is known as dental black stain. Dental black stain is characterized by pigmented black dots or a dark line located in the cervical third of a tooth’s crown. The stain generally cannot be cleaned using regular oral hygiene procedures; instead, professionals must use special instruments and polish the tooth.² Several studies have been conducted to assess the prevalence of dental black stain in various countries. The prevalence of dental black stain in Switzerland reached 19.9%, UK 21%, Poland 4.5%, Germany 4%, Italy 6.3%, Brazil (2003) 14.8%, Spain (2005) 7.54%, Philippines 16%, India 18%, Brazil (2012) 3.5%, Greece 2.4%, Spain (2013) 3.1%, China 9.9%, and Istanbul 18.5%.³-⁶ However, the prevalence of dental black stain in Indonesia is still unknown.

Dental black stain is basically a dental plaque. However, it is a special form of dental plaque because it contains certain substances not found in regular dental plaque. One of these substances is generally insoluble ferric salt, or iron sulfide. This substance is believed to be the cause of the black color seen in dental black stain. Iron sulfide is formed by the interaction between hydrogen sulfide and ferum. Hydrogen sulfide is the result of bacterial metabolism, whereas ferum is a mineral contained in saliva or gingiva fluid. Other than iron sulfide, dental black stain also contains high concentrations of calcium and phosphate.³ Ferum is a mineral that can cause the dark discoloration of teeth.¹ Usually, this mineral is found in saliva, and it enters the body when a person consumes foods or beverages that contain ferum.⁷ Other than ferum, another mineral that can cause tooth discoloration is magnesium.¹ In the research conducted in India, magnesium was found in dental black stain.⁸ This mineral is also usually found in saliva, and it gets into the body when a person consumes magnesium-containing foods or beverages.⁹

Ultra-high-temperature (UHT) milk is processed using a high-temperature sterilization technique that kills microorganisms without negatively affecting the quality of the milk. The process involves quickly running the UHT milk...
through the heating and cooling phases. UHT milk contains both macro- and micronutrients. Some of the micronutrients in UHT milk are minerals such as ferum and magnesium. Until now, research on the relationship between ferum and magnesium content in the plaque and saliva of children with black dental stain and the consumption of UHT milk did not exist. To address this lack of research, the author performed a study on ferum and magnesium levels in the plaque and saliva of children with dental black stain who consume UHT milk regularly.

Materials and methods

The study applied the laboratory observation method of descriptive research. The research subjects were elementary school children (aged 4–8 years) from five elementary schools in Jakarta, Indonesia, including Jubilee Elementary School, St. Cicilia Ancol Elementary School, Strada Van Lith Elementary School, Ibnu Sina Elementary School, and Insan Mandiri Elementary School. The quantity of subjects was determined using the convenience sampling method. This method is a special nonprobability sampling technique that relies on collecting data from the population participating in the study. The sample of the study was the black stain plaque and saliva from children with dental black stain who consume UHT milk regularly.

Sampling began after the researcher described the study to the children’s parents and gave them informed consent sheets. Subject selection was done in school using intraoral examinations to determine which children presented with the black stain plaque. The researcher then took samples of both the black stain plaque and saliva from those who met the inclusion criteria. Plaque samples were taken by scraping each subject’s teeth and then inserting the material into a microcentrifuge tube containing phosphate buffer saline. Saliva samples were obtained by having each subject spit into a 15-ml tube. The first laboratory procedure involved manufacturing ferum and magnesium standard solution with concentrations of 2, 4, 6, 8, and 10 ppm. Standard solution was prepared from Fe2+ 100 ppm and Mg2+ 1,000 ppm and then diluted using DM-AQUA (Dikma Technologies Inc., Lake Forest, CA). Afterward, the plaque and saliva samples were processed.

The plaque samples were weighed and then dissolved using acid. These samples were diluted by adding DM-AQUA. Saliva sample tubes were inserted into the centrifuge, and 1 ml of liquid from each tube was taken. The salivary fluid was diluted by adding DM-AQUA.

After the sample processing was completed, ferum and magnesium levels were measured using atomic absorption spectrophotometry. The ferum calibration was performed using a standard solution of ferum with concentrations of 2, 4, 6, 8, and 10 ppm. After a calibration curve for ferum was completed, the ferum level was measured in the whole sample. The neutralization was done by inserting a tube into DM-AQUA before calibrating the magnesium curve. The magnesium calibration was performed using a standard solution of magnesium with concentrations of 2, 4, 6, 8, and 10 ppm. After a calibration curve for magnesium was completed, the level of magnesium was measured in the whole sample. The results of the plaque samples were measured in percentages (b/b), whereas the results of the saliva samples were measured in ppm. The data obtained from the laboratory procedures were processed using descriptive statistical test software.

Results

Table 1 shows the measurement of ferum and magnesium levels taken from 50 teeth of 12 children who drank UHT milk on a regular basis. The data indicated ferum levels of 30.50% and magnesium levels of 0.23% in black stain plaque.

<table>
<thead>
<tr>
<th>Metal</th>
<th>N</th>
<th>Levels (% (b/b))</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fe</td>
<td>50 teeth (12 children)</td>
<td>30.50</td>
</tr>
<tr>
<td>Mg</td>
<td>50 teeth (12 children)</td>
<td>0.23</td>
</tr>
</tbody>
</table>

Table 1. Measurement of Ferum and Magnesium levels in black stain plaque.

Table 2 shows the average ferum levels in the saliva of children with dental black stain who were drinking UHT milk at 7.68 ppm and the average magnesium levels in the saliva of children with dental black stain who were drinking UHT milk at 1.59 ppm.

<table>
<thead>
<tr>
<th>Metal</th>
<th>N</th>
<th>Mean ± SD (ppm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fe</td>
<td>16</td>
<td>7.68 ± 1.25</td>
</tr>
<tr>
<td>Mg</td>
<td>16</td>
<td>1.59 ± 0.09</td>
</tr>
</tbody>
</table>

Table 2. Measurement of Ferum and Magnesium levels in saliva of children with dental black stain.
Discussion

In this study, the ferum level of black stain plaque in children who consumed UHT milk was 30.50%. When compared with the research conducted in India, where the level of ferum in black stain plaque was 2.56%, the ferum level in this study was much higher.8 However, when compared with studies conducted in Israel, where the ferum level in black stain plaque was around 40%, the result in this study was lower.12 The percentage of ferum level difference in black stain plaque was likely to be caused by dietary factors.

The level of magnesium in black stain plaque in children who consume UHT milk was 0.23%. When compared with the research conducted in India, where the magnesium level in black stain plaque was 0.72%, the magnesium level in this study was lower.10 The level of magnesium in children without dental black stain was 0.23%, again the same result as in this study.12

The mean ferum level in the saliva of children who consume UHT milk was 7.68 ppm and the mean level of magnesium in the saliva of children who consume UHT milk was 1.59 ppm. These results cannot be compared with those of previous studies because studies to measure the magnesium level in the saliva of children with dental black stain have not yet been done.

Conclusion

In this study, the ferum level in plaque was 30.50% and the ferum level in saliva was 7.68 ppm in children with dental black stain who consume UHT milk on a regular basis. The magnesium level in plaque was 0.23% and the magnesium level in saliva was 1.59 ppm in children with dental black stain who consume UHT milk on a regular basis. These results lead to the conclusion that UHT milk, which has ferum levels of 4% to 25%, can cause precipitation of ferum, particularly in children. Based on these findings, children who consume UHT milk on a regular basis are advised to rinse their mouths with water immediately after drinking UHT milk to reduce their risk of dental black stain. The sampling process will also focus more on the black stain plaque because it is more representative than saliva. Further research is needed to determine other dietary risk factors that may contribute to high ferum levels and, subsequently, dental black stain.

Declaration of Interest

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References