

# Relationship between Fluor Concentration and Structure Pattern of Enamel Prism in Enamel Surface after Coffee and Black Tea Exposure

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## ABSTRACT

Coffee and tea are often consumed by Indonesian and Aceh people. Based on previous research reported that concentration of fluor in coffee and tea affects pattern structure of enamel prism. Currently, little explanation about relationship between fluor concentrations and pattern structure of enamel prism in enamel surface after coffee and tea drinking water exposure. The purpose of this study is to explain relationship between fluor concentrations and pattern structure of enamel prism in enamel surface after coffee and tea drinking water exposure.

**Materials and methods:** Eight maxillary premolars were divided into control and treatment group. Positive control group exposure to NaCl 0.9%, negative control group exposure to sulfuric acid 50%, treatment groups are exposure to coffee and tea drinking water. This study conducted by scanning electron microscopy/energy dispersive X-ray spectroscopy (SEM/EDX). Data analysis is descriptive qualitative.

**Results:** Fluor in tea increases the density of the enamel prism structure pattern. Surface enamel that exposed to tea has described better arrangement of enamel prism pattern than enamel surface that exposed to coffee.

**Conclusion:** Fluor concentration in drinking water has relationship with structure patterns of enamel prism. The structure patterns of enamel prism that exposed to tea have better arrangement than enamel that exposed to coffee.

**Keywords:** Enamel prism, Fluor, SEM/EDX.

**How to cite this article:** Nasution AI, Gani BA, Pospos RM. Relationship between Fluor Concentration and Structure Pattern of Enamel Prism in Enamel Surface after Coffee and Black Tea Exposure. *World J Dent* 2012;3(4):284-289.

**Source of support:** Nil

**Conflict of interest:** None declared

## INTRODUCTION

Coffee and tea is a type of drink that has long been known and widely consumed worldwide, including in Indonesian.<sup>1</sup> Commerce market was estimate the level of coffee consumption in Indonesia is an attractive market. According to Food and Agriculture Organization of the United Nations (FAO) in several countries, including Indonesia, the consumption of black tea is more than 140 thousand tons per year.<sup>2</sup> Some of chemical compound in both beverages can improve the taste of coffee and tea.<sup>3,4</sup>

Factly, scientific studies conducted to examine coffee and tea.<sup>3-5</sup> Published studies reported that coffee and tea has health benefits for the human body. Both of these drinks

considered as a beverage that can reduce risk of some diseases including the risk of caries.<sup>3-7</sup> Research results in 2009 in Bengaluru reported the score of decayed missing filled surface (DMFS) in people who do not consume coffee is 4, whereas in people who consumed coffee is 2.9.<sup>4</sup> Research data from research of dental caries prevalence 2001 in London, England, also showed that the prevalence of caries in people consumed tea are lower than people do not consumed tea.<sup>7</sup>

Fluor can strengthening an enamel prism structure pattern and assist the remineralization process in the structure of enamel prism to increase density of enamel surface.<sup>8,9</sup> United States Department of Agriculture National Fluoride Database (USDA) reported the concentration of fluor in coffee and tea is difference.<sup>10</sup> Fluor in arabica is 0.52 to 1.25 mg per liter, while in black tea is 9 mg per liter.<sup>11</sup> Based on the results of studies using fluoride-specific electrode (FSE) reported that there are differences in concentration of fluor in coffee and tea, but not significant.<sup>12</sup>

Enamel is structured by enamel prism which formed by crystal apatite.<sup>13,14</sup> The apatite is  $\text{Ca}_{10}(\text{PO}_4)_6(\text{X})$ . The (X) elements could be hydroxyl, fluor, carbonate or magnesium.<sup>15,16</sup> The apatite is very tolerant from substitutions. Enamel structure which formed by fluor apatite (FA) demonstrate a different structure than HA.<sup>17</sup> The fluor substitution indicated the change of apatite crystal structure as enamel former.<sup>18</sup> Research study using scanning electron microscopy (SEM) known that fluor has significant effect on apatit.<sup>19</sup> Substitution of fluor into apatite, influence the pattern of the surface of the prism structure.<sup>8,9</sup>

Based on fluor content in coffee and tea can affect the macroscopic of enamel surface, that important to conduct study regarding to relationship between structure of prism patterns and fluor concentration in enamel surface after exposure to coffee and tea. The aim of this study is to explain the relationship between patterns of enamel prism and fluor concentration in enamel surface after exposure to coffee and tea.

## MATERIALS AND METHODS

This laboratory experimental conducted at Laboratory of Metallurgy Faculty of Engineering University of Indonesia and Laboratory of Chemistry Faculty of Mathematics and Natural Sciences University of Syiah Kuala. This research was conducted in March 2011.

Specimens are eight premolars that collected from orthodontic treatment that extracted from various dentistry clinics in Banda Aceh-Indonesia. The specimens were free from dental caries, abrasion, attrition and erosion. The specimens were randomly separated into four groups: Control positive, control negative (applied with 50% sulfuric acid), and two treatment groups (applied with 20 gm coffee and tea in 150 cc water). Each group consisted of two specimens: Every group wrapped in ringer-cotton for storage.

SEM analysis conducted with a secondary electron emission for image-acquisition of both surfaces of the specimens. Two separate magnifications were used: 20× and 500×, EDX/EDS analysis conducted with backscattered emission, i-Probe 380 to 450 pA, accelerating voltage 12 kV. Wavelength is under 1,000 mA. Result analysis is fluor percentage rate of EDX/EDX standard.

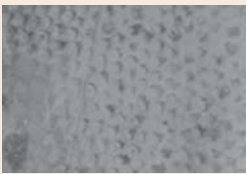

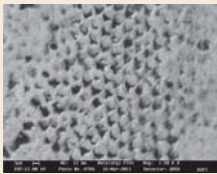
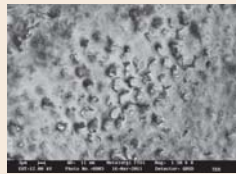
## RESULTS

Positive control (Fig. 1) should that exposure with 0.9% NaCl solution based on SEM observation (magnification 1,500×), showed enamel prism structure in regular pattern with head and tail of enamel rods in normal appearance.

Generally, the pattern of enamel prism in working distance (WD) 12 mm was 82% have good density. The picture showed more clearly in magnification of 3,000× (Fig. 1). The pattern of enamel rods showed a keyhole shape. Picture also showed demarcated line on peripheral, no gaps on rod sheath, and rod cores not porous. EDX analysis of positive control showed ratio between calcium and phosphate is 2:1 (Ca = 39.83% and p = 22.97%). EDX analysis showed fluor content as 12.18%\*. Sign (\*) that found on fluor concentration in EDX is mean fluor concentration on the surface of enamel equal to zero. EDX analysis can be see on Table 1 below.

Scanning electron microscopy of negative control in magnification 1,500× that exposed with 50% sulfuric acid for 180 minutes showed irregularly pattern of enamel prism. Enamel prism mainly showed a damage area on part of head of enamel prism. The pattern of the enamel showed prismless zone. Picture in magnification 3,000× (Fig. 2) generally showed no demarcated line on peripheral part, rod sheath showed fibrous-like and indicate damage of enamel as well as highly porous on part of rod core enamel. Some areas showed decreasing in enamel density. SEM picture on WD 12 mm of showed only 14% undamaged

Table 1: SEM/EDX analysis

Unit	Positive control (NaCl 0.9%)	Negative control (sulfuric acid 50%)	Treatment 1 (coffee)	Treatment 2 (tea)
Ca	39.83	92.83	63.95	35.04
P	22.97	30.14	30.93	21.46
F	-12.18*	-32.42*	-11.76*	1.19
Ca:P	2:1	3:1	2:1	2:1
SEM Mag 3,000x				
Structure of prism	<ol style="list-style-type: none"> <li>1. Regular prism</li> <li>2. Structure of head and tail of enamel prism clearly visible without lost of structure</li> <li>3. Good density, in WD = 12 mm 82% prism visible in good condition</li> <li>4. Keyhole shape</li> <li>5. Part of perifer demarcated</li> <li>6. No gaps on rod sheath</li> <li>7. No porous on rod core</li> </ol>	<ol style="list-style-type: none"> <li>1. Irregular prism</li> <li>2. Damage of head and tail of enamel prism</li> <li>3. Bad density, in WD = 12 mm only 14% prism presented good condition</li> <li>4. Damage of shape</li> <li>5. Part of perifer undemarcated</li> <li>6. Rod sheath fibrous-like and damage</li> <li>7. Rod core very porous</li> </ol>	<ol style="list-style-type: none"> <li>1. Regular prism</li> <li>2. Both structure of head and tail begin to lose</li> <li>3. Bad density, in WD = 12 mm, only 37% prism presented good condition</li> <li>4. Outline shape: Fish head</li> <li>5. Structure of perifer begins to lose and tend to independent</li> <li>6. Rod sheath begin to damage/crack visible in various surface</li> <li>7. Rod core porous</li> </ol>	<ol style="list-style-type: none"> <li>1. Regular prism</li> <li>2. Both structure of head and tail presented good condition</li> <li>3. Good density WD = 12 mm 50% prism presented good condition</li> <li>4. Outline shape: Keyhole and arcade</li> <li>5. Structure of periphery is depended several area presented good density</li> <li>6. Rod sheath begin to damage/crack visible in various surface</li> <li>7. Rod core not porous and several surface presented improving pattern</li> </ol>

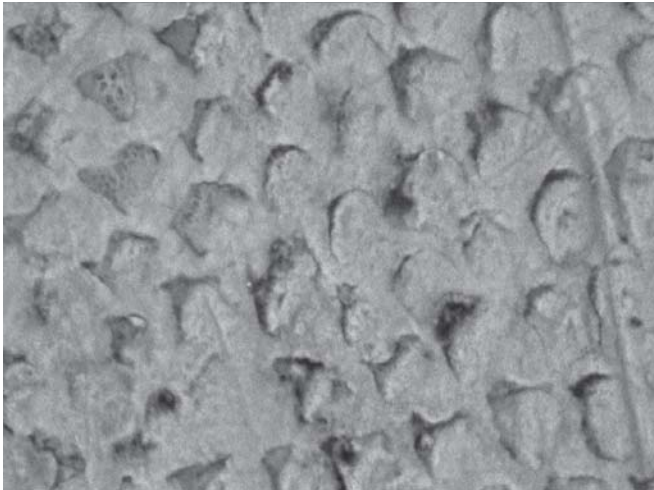


Fig. 1: SEM picture of positive control (Magnification: 3,000×)



Fig. 2: SEM picture of negative control (Magnification: 3,000×)

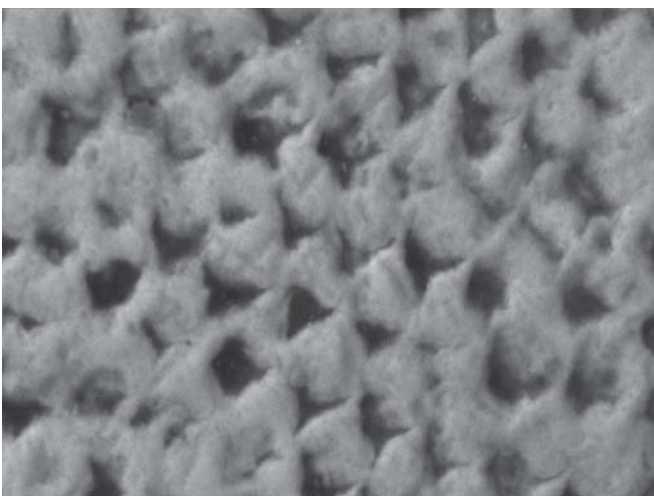


Fig. 3: SEM picture of coffee exposure specimen (Magnification: 3,000×)



Fig. 4: SEM picture of tea exposure specimen (Magnification: 3,000×)

enamel prism. EDX analysis of negative control showed ratio between calcium and phosphate to be 3:1 (Ca = 92.83% and p = 30.14%). EDX analysis showed fluor is -32.42%\*.

SEM of coffee exposure in 15 minutes in magnification 1,500× showed a pattern of enamel rods that arranged in head and tail of enamel rods even in regular shape but it was beginning to loss of structure. Prism pattern on the enamel surface showed decreasing of density of enamel. SEM picture on WD = 12 mm of the pattern of enamel prism shows only 37% in clear condition. SEM picture in magnification 3,000× (Fig. 3) generally showed visible the outline shape that resembles of fish scale. Some areas indicate porous areas. The peripheral parts indicate lost of rod structure. Generally, damaged pattern occurred in the head of rod sheath. This picture shows a decrease of enamel density as shown in negative control. EDX analysis of coffee exposure showed ratio between calcium and phosphate is 2:1 (Ca = 63.95% and p = 30.93%). EDX also showed fluor is -11.76%\*.

SEM of tea exposure in 15 minutes in magnification 1,500× showed the pattern of prism structure that arranged on a regular shape with a prism shape with head and tail is still clearly visible. Generally, the pattern of prism structures showed an increasing of enamel density. Picture of the pattern of the enamel surface showed pattern of the surface structure same as enamel prism on positive control. Density relatively 50% in good condition as shown in WD = 12 mm. SEM picture in magnification 3,000× (Fig. 4) showed the enamel prism structure as keyhole shape. SEM generally show the demarcated peripheral, no gaps on rod sheath, and rod cores are not porous. EDX of tea exposure showed ratio between calcium and phosphate is 2:1 (Ca = 35.04% and p = 21.46%). EDX also showed fluor is 1.19%.

## DISCUSSION

Enamel consists of a prism that extends from the dentinoenamel junction (DEJ) to outer surface of enamel.



Each prism consists of crystal hydroxy apatite.<sup>20</sup> Calcium and phosphate ions dominate prismatic structures. From the results of this study, it is known that calcium and phosphate ratio is 2:1. According to Robinson et al study, it was reported that average ratio of calcium and phosphorus in the enamel surface is 2:1.<sup>20</sup>

SEM/EDX observations in this study suggests that the negative control group (50% sulfuric acid for 180 minutes) shown demineralization has occurred in this specimen. The structural of chemical properties of the mineral apatite allow the substitution of ions in enamel, especially calcium and hydroxyl. The mineral content of prism structure determine pattern the surface of the enamel. Little fluor content on the surface of enamel indicates worse prismatic structure. Decreasing of fluor content in enamel surface were initiating the formation of cavities (caries) and demineralization.<sup>20</sup> This condition, greatly affects the pattern of the structure of enamel prism.<sup>21</sup> Many scientific studies have proven that fluoride is the most important in caries prevention.<sup>8,17,19,22-24</sup> Fluor is believed to be able to strengthening the structure of the enamel because fluoride is able to stimulate the remineralization.<sup>17</sup>

The results of this study proved that fluor which exposed on the enamel surface after tea exposure improving the structure of enamel prism pattern, especially in rod core and peripheral rods. According to Simpson et al that reported there is an interaction between the fluor in black tea against hydroxyl apatite.<sup>21</sup> Several studies also support this result that the incorporation of fluor ions into the apatite are significantly effective in inhibiting demineralization.<sup>17,20</sup> From these studies obtained a conclusion that the decrease in the amount of minerals, especially fluor ions affect the structure of the prism. However, these results differ with previous research that reported fluor substitution on hydroxyl ions cannot affect the enamel surface and does not prevent demineralization.<sup>17</sup>

The surface of enamel prism that exposed to fluor showed more consistent morphological structure. Fluor acts as a catalyst and increases the level of formation of hydroxy apatite (HA) into fluor hydroxy apatite (FHA) or fluor apatite (FA).<sup>16</sup> This substitution was stimulated, the crystal size becomes smaller. As a result, an increasing number of crystals after fluoride application will increase enamel prism structure and tend to be more resistant to acid. Similar with this study, previous study reported that fluor affects the surface structure of enamel prism pattern.<sup>24</sup>

Result of this study was also supported by the results of research by Okazaki. They proved that there is a significant influence on hydroxy apatite after the incorporation of fluor ions. Incorporation of fluor ions into apatite crystal size

may improve the enamel crystal. Substitution of fluor ions into apatite crystals was reported can stimulate the increasing of enamel prism structural.<sup>17</sup> This answer is in line with result of research that was conducted by various researches.<sup>17,25,26</sup>

A mineral component in enamel was substituted of calcium hydroxyl apatite. Hydroxy apatite compounds  $CA_{10}(PO_4)_6(X)$ . The X mean in specimen were packed by hydroxyl ions.<sup>15,16</sup> Hydroxyl ions are highly susceptible to substitution by some other ions, such as fluor. As apatite, FHA and FA were very tolerant of substitution of ions in the lattice of crystal apatite. The substitution of other ions into apatite will improve shape and physical properties of hydroxy apatite.<sup>27</sup> The number of hydroxyl ions 20 to 30% less than the apatite. Defects in hydroxy apatite are frequent, particularly the trend and experienced demineralization soluble at low pH (acid).<sup>20</sup> Enamels that have remineralization is more resistant to acid than is healthy because enamel is not easily soluble fluorine minerals and crystals deposit back enamel.<sup>28</sup>

Fluor is the most electronegative atom and a reactive element. The substitution of fluor ions into apatite will replace hydroxyl ions because fluor has a smaller atom than hydroxyl. Two ions that are similar, have same components, and have the smaller atom can lead the substitution of hydroxy apatite crystal lattice and thus affects the pattern of prism structure of enamel.<sup>27</sup> Increased prism structure can occur due to an increase in the amount of fluor. The fluor position is in c-axis in crystal apatite. This structure was closer to calcium that causes the energy of crystal that required in apatite composition becomes lower. This condition made crystal structure more stable than apatite that arranged by hydroxyl on c-axis. This condition was proved that substitution of fluor into hydroxyl position made crystal structure becomes more resistant to acid.<sup>20</sup>

The results of this study also showed the different pattern of the prism structure after exposure to coffee and tea. Prism structure on the enamel surface after exposure to coffee is more likely to show demineralization than tea exposure. The prism structure after exposure to coffee for 15 minutes showed the structure of prism with the peripheral part of independent and surface fibrous-like texture. According to research results of Nicholl, Judd and Ansell, the exposure of acid into enamel would influence the demineralization. The prism damage in the peripheral after exposure to coffee is a reaction of prism to acid attack.<sup>26</sup> As shown by SEM, the prism pattern more has porous with a destruction of half prism. That usually occurs in the head and rod sheath. Coffee, factly is an acidic consumption that initiated demineralization of enamel.

The statement was similar with the research of Simmelink and Abrigo on morphology and decalcification hydroxy apatite crystals exposure with acid in different duration.<sup>29</sup> This study also not contradict with the results that stated there are different reactions in the peripheral regions (rod periphery) to acid attack into enamel.<sup>26</sup> SEM of the prism structure after exposure to sulfuric acid and coffee visible organic material (enamelin) in interrod higher than other specimen. At low pH (acid) that contained enamel often visible organic material on tooth surface, after exposure by acid.<sup>20</sup>

Results of this study also obtained picture of normal prism structure as shown in Figure 1 on positive control. SEM observations at 1,500× magnification show the pattern of prism structure without damage. Detailed description that shown in magnification 3000× looks prisms are interconnected with each other to form a keyhole-like outline, hexagonal, head of the fish (fish scale) and arcade. Demarcated peripheral (dependent) of rod still looks clear and without gaps as well as the rod is not porous, but another studies shown there is a gap between the prism enamel that called interprismatic.<sup>30</sup>

Changes in the structure of the prism pattern seen in the SEM observations in this study generally show changes in the pattern of the enamel structure begins at the peripheral part and continues to head the prism, then extends along the lateral edge, and eventually spread to the inferior region and affect cervical rod cores. This statement is in accordance with the results on SEM studies on the prism enamels with multiple treatments using acids and fluor.<sup>31</sup> Based on this study, it can be proved that prism structure pattern on the enamel surface showed an increase prism structure because of the addition of fluor ions. Thus, it can be stated that the substitution of fluor ions can affect the pattern of enamel surface prism structures, especially in the core and peripheral rods.

## CONCLUSION

The exposure of coffee and black tea affected prism pattern. Fluor ions that contain in black tea affect the better pattern of enamel surface prism structures, especially in the core and peripheral rods.

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