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Saliva parameter analysis on smoker and non smoker patients exposed to intraoral radiography

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Abstract

Objective: To analyze saliva parameter such as salivary pH, total protein, buffer capacity, as well as sodium and potassium level on smoker and non smoker patients after exposed to intraoral radiography.
Material and Methods: This study was an observational study with cross-sectional design. Samples were smoker and non smoker patients who were referred to Dental Hospital Hasanuddin University. Saliva samples were taken by draining method. Samples were transferred immediately to Laboratory of Biochemistry, Faculty of Mathematics and Science to be processed regarding saliva pH, saliva total protein, buffer capacity, as well as saliva sodium and potassium level. Saliva pH were measured with pH meter Hanna instruments, total protein with Lowry's method, buffer capacity with Ericcson's method, sodium and potassium level with atomic absorption spectrophotometer Parking Elmer A400. Data were then analyzed with statistical test Kolmogorov Smirnov, independent t-test, and oneway Anova with $p<0.05$ were considered statistically significant.
Results: There were no significant difference on salivary pH, buffer capacity and potassium level between smoker group and non smoker group ($p>0.05$) before exposed to intraoral radiography, but we found significant difference on saliva total protein and sodium level between smoker and non smoker group ($p<0.05$) before radiography exposure. There were no significant change on salivary pH, total protein, buffer capacity, sodium and potassium level after exposed to intraoral radiography ($p>0.05$) on both smoker and non smoker group.
Conclusion: Exposure of intraoral radiographs did not have significant effect on salivary pH, total protein, buffer capacity, as well as sodium and potassium level.

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Introduction

Saliva is the first biological fluid that is exposed to cigarette smoke, containing numerous toxic compositions responsible for structural and functional changes in saliva. Beside cigarettes, there are several external factors that affect salivary glands, one of which is dental x-ray.[1,2] X-rays affected cells by ionizing and forming free radicals within 10-13 seconds after its exposure. The biological effect can be a temporary disruption of endothelial cells function. X-rays affected blood vessels permeability by vasoconstriction and vasodilatation resulting in vascular permeability changes.[3]

Radiographs are essential in dentistry for the diagnosis, treatment planning, treatment monitoring and follow-up of patients. One of the most commonly used technique is intraoral radiography. It is easily accessible and the cost, in terms of both radiation dose and monetary, value, is low compared with other radiographic techniques.[4]

Although the radiological doses used by dentists are low individually, patients are often exposured to many repeat dental radiographic examinations. During radiographs assessment, salivary glands and oral mucosal received the highest absorbed doses.[5,6] Salivary glands are exquisitely sensitive to radiation, yet unlike classically radiosensitive tissues, they proliferate slowly and are made up of highly differentiated cells that could lead to changes in acinar cells and affect saliva composition, structure, and volume.[5]

Numerous studies have been done on saliva analysis after high dose radiation exposure but still very limited on low dose radiation, especially those accompanied by cigarette smoke exposure accumulation. Hence, the authors were interested to analyze saliva parameter such as salivary pH, total protein, buffer capacity, as well as sodium and potassium level on smoker and non smoker patients after exposed to intraoral radiography.

Material and Methods

This study was an observational study with cross-sectional design on smoker and non smoker patients who were referred by their dentist to Dental Radiology Department, Faculty of Dentistry, Hasanuddin University to undergo intraoral radiographs. Ethical clearance was approved by Ethical Committee Faculty of Dentistry, Hasanuddin University (0129/P1.09/KEPKFKG-RSGM UNHAS/2019). Patients were interviewed and informed consent were obtained from patients fulfilled

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the inclusion criteria and willing to participate in this study.

Patients were divided into 2 groups: smoker patients undergo intraoral radiographs for endodontic treatment, 5 patients; non smoker patients undergo intraoral radiographs for endodontic treatment, 5 patients.

Saliva samples were taken on 9-11 pm to avoid saliva concentration bias caused by circadian rhythm using draining method.[5] Samples were taken based on radiographs schedule of endodontic treatment; (P1) Before exposure, (P2) Immediately after first and second exposure, (P3) 10+2 days after first and second exposure/before third and fourth exposure, (P4) Immediately after third and fourth exposure and (P5) 10+2 days after third and fourth exposure.

The subject was made to sit quietly with the head bent down and the mouth open to allow the saliva to drip passively from the lower lip into the graduated sterile tubes. Test tubes were then labelled with group code and time taken, stored in cooler box and immediately transferred to the laboratory.

Saliva parameter analysis were done in Laboratory of Biochemistry, Faculty of Mathematics and Science, Hasanuddin University. Salivary pH was measured with pH meter Hanna Instruments, total protein with Lowry's method, buffer capacity with Ericcson's method, sodium and potassium level were measured with atomic absorption spectrophotometer parking elmer A400.

The data were then processed with computerized statistical test using Kolmogorov Smirnov and oneway Anova. P values <0.5 were considered statistically significant.

Table 1 **Oneway ANOVA test result of saliva parameter (pH, total protein, buffer capacity, sodium and potassium) on smoker and non smoker group exposed to intraoral radiograph**

Group		Smoker	
Saliva Parameter		Non Smoker p	p
Saliva pH	P1-P5	0.968	0.99
Total protein	P1-P5	0.978	0.785
Buffer capacity	P1-P5	0.997	0.994
Sodium	P1-P5	0.934	0.941
Potassium	P1-P5	0.996	0.997

*p<0.05 = statistically significant

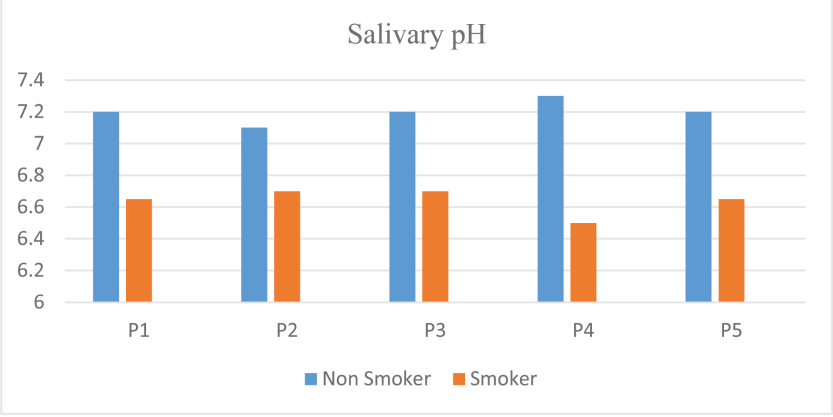

Figure 1 Salivary pH on smoker group compared to non smoker group exposed to intraoral radiography

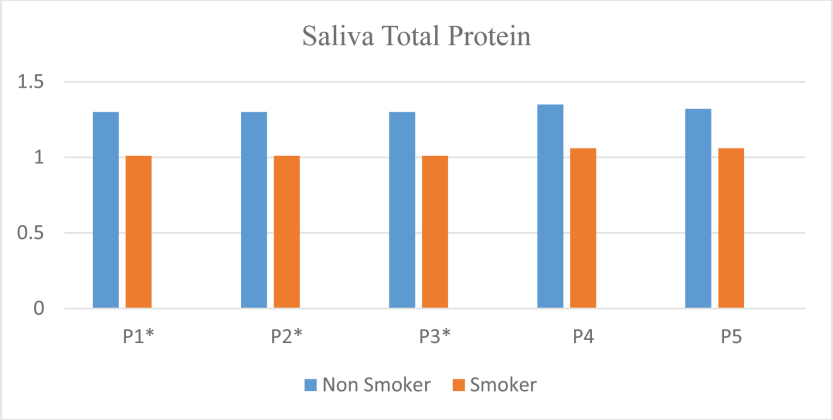

Figure 2 Saliva total protein on smoker group compared to non smoker group exposed to intraoral radiography

Results

The result of saliva parameter analysis showed lower mean of salivary pH, total protein, buffer capacity, as well as sodium and potassium on smoker group compared to non smoker group as shown on figure 1- figure 5.

Result of independent t-test showed there was no significant difference (p>0.05) on salivary pH figure 1, total protein figure 2, buffer capacity figure 3, sodium figure 4 and potassium level figure 5 between smoker and non smoker group, but there was significant difference (p<0.05) on saliva total protein figure 2 and sodium level figure 4 between smoker and non smoker group before exposed to radiograph (P1), immediately after first and second exposure (P2 and P3), and several days after first and second exposure (P3 and P5).

Oneway ANOVA test result showed no significant change (p>0.05) for saliva parameter (pH, total protein, buffer capacity, sodium and potassium) on smoker and non smoker group exposed to intraoral radiographs, as seen on below table 1.

There were slight change on salivary pH (p>0.05), total protein (p>0.05), buffer capacity (p >0.05), sodium (P>0.05) and potassium (P>0.05) for P2, P3, P4, and P5 smoker group and non smoker group. This result showed that intraoral radiograph exposure do not have significant effect on salivary pH, total protein, buffer capacity, sodium and potassium both on smoker and non smoker group.

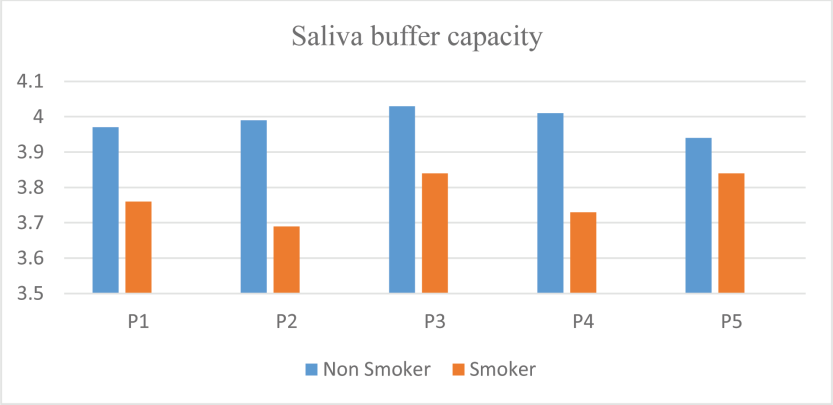

Figure 3 Saliva buffer capacity on smoker group compared to non smoker group exposed to intraoral radiography

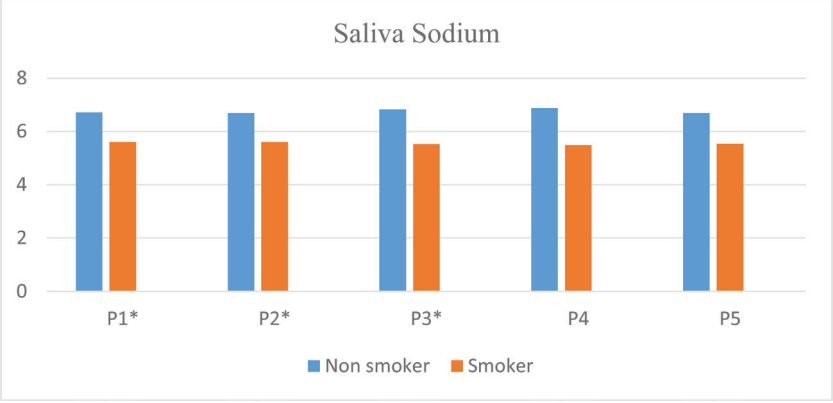

Figure 4 Saliva sodium level on smoker group compared to non smoker group exposed to intraoral radiography

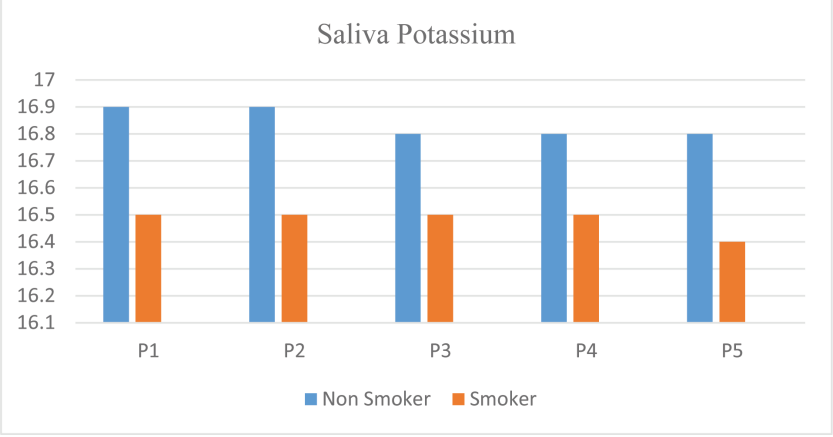

Figure 5 Saliva potassium level on smoker group compared to non smoker group exposed to intraoral radiography

Discussion

The result of this study showed lower salivary pH, total protein, buffer capacity, sodium and potassium level on smoker group compared to non smoker group ($p>0.05$ for salivary pH, buffer capacity, and potassium level; $p<0.05$ for total protein and sodium level). Because nicotine in cigarettes work on certain cholinergic receptors in the brain that will affect central nervous system activity which trigger changes in saliva secretions.[7] Changes in salivary secretion will then affect the flow of saliva and salivary pH in smokers. In long-term smoking, the taste receptors, a primary site for salivary secretion, are repeatedly exposed to tobacco for long-time thus presumably affecting the salivary reflex.[8] A study conducted by Khan et al[7] showed lower mean of salivary pH on smoker group (6.8+0.11) compared to non smoker group (7.03+0.14). The decrease in salivary flow rates alters salivary pH by decreasing bicarbonate secretion and this decrease in saliva bicarbonate in turn decreases the salivary pH. The decrease in salivary flow rates on smoker also affect on saliva total protein ($p<0.05$) and electrolyte composition (sodium $p<0.05$; potasium $p>0.05$).[8,9]

Total protein level in our study was significantly decreased in smokers as compared to non smoker. This could be explained by the facts that, as most of the proteins are secreted by acinar cells, any situation that impairs the acinar activity probably results in secretion of smaller volume of total proteins.[10] In addition, injury to acinar, ductal secretory unit caused by tobacco-related toxic products might be responsible for the reduction in total proteins. The decrease in total salivary protein levels in smokers subjects leads to a decrease in the levels of immunoglobulin and enzymes that work on saliva and a decrease in glutathione which acts as the main antioxidant in the mouth.[11,12] Aldehyde component in cigarette smoke can bind to -SH group in salivary protein and reduce its function. A decrease in total salivary protein levels can result in tissue damage and an increased risk of oral cavity infection which increases the risk of oral disease in smokers.[13] In a study conducted by Jethlia et al for 50 smokers and 50 non-smokers, a significant difference in total protein was obtained in the smoker group (1.08 + 0.65) compared to the control group (1.41). + 0.75). This can also be caused by trauma to the ductal secretion unit due to toxic cigarette products, which in turn will cause a decrease in total protein levels.[14]

The result of this study showed that intraoral radiograph exposure did not have significant effects on salivary pH, total protein, buffer capacity, sodium and potassium level on smoker and non smoker group ($p>0.05$ for all saliva parameter). The initial interaction between ionizing radiation and salivary glands occur at the level of the electron within the first 10–13 seconds after exposure. These changes result in modification of biologic molecules within the following seconds to hours. The molecular changes may lead to alterations in cells and organisms that persist for hours, decades, and possibly generations. These changes may result in cell injury

or cell death.[15] X-ray radiation exposes its energy in the form of free electrons to cellular components such as water and undergoes a process of free radical formation that releases into H_2O_2 which will cause apoptosis of salivary gland secretion cells, especially serous asini cells. Because serous asini cells are more radiosensitive than mucous asini cells, saliva become stickier and thicker. The initial effect of radiation on the salivary gland is damage to the acinar cell plasma membrane, which in turn affects the muscarinic receptors which stimulate salivary secretion.[16]

A study conducted by Pow et al.[17] showed reduced saliva buffer capacity and total protein as many as 44% on patients underwent head and neck radiotherapy (radiation dose >70 Gy), while a study conducted by Galih et al.[18] showed a slight difference (0.25 mean difference) of salivary pH before and after panoramic radiography (radiation dose 35.8–103.2 mGy). Adverse effect of radiation are greatly determined by radiation doses,[5] where radiation dose for intraoral radiographs is 1.45–4.45 mGy,[5] very low compared radiotherapy doses (>70 Gy with fractioned radiation 1.8-2 Gy per day), hence the different doses of radiation greatly affect saliva parameter changes[16] and this explained the result of this study that showed no significant change in saliva pH, total protein, buffer capacity, as well as sodium and potassium level after exposed to intraoral radiography.

Conclusion

Exposure of intraoral radiographs did not have significant effect on salivary pH, total protein, buffer capacity, as well as sodium and potassium level immediately after exposure and several days after exposure. However, repeat dental radiographic examinations should be clinically justified and each exposure should be expected to give patient a positive net benefit.

Acknowledgment

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Conflict of Interest

The authors report no conflict of interest.

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