



Occupational Exposure in Dental Laboratory Technicians May Induce Nuclear Abnormalities in Buccal Mucosa Cells: A Preliminary Study

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ABSTRACT

Background and Aim: Increased rate of micronucleus in buccal mucosa cells and its correlation with carcinogenesis is worth consideration. Recently, occupation in dental laboratories has been proposed as a predisposing factor. Therefore, the present study aimed to evaluate the effect of occupational exposure on buccal mucosa cells of dental laboratory technicians.

Methods and Materials: This historical-cohort study was conducted on 16 male dental laboratory technicians and 16 males were selected as the control group. All samples were matched according to age. The samples neither had any recent viral diseases nor were consumers of any specific medications. Cigarette smokers and alcoholics, individuals with risky occupations or with a history of radiotherapy were excluded. Buccal mucosa cells were sampled by use of a plastic spatula and were stained with Papanicolaou stain. Micronucleus frequency was evaluated under light microscope (400×). T-test was used for statistical analysis. Significance level was set at 0.05.

Results: Micronucleus frequency equaled 69 ± 70 and 27 ± 8.6 in case and control samples, respectively; which is 2.6 times higher in the case group. T-test showed that the difference in micronucleus frequency was significant between the two groups. ($P < 0.001$)

Conclusion: The present study showed that the frequency of micronucleated cells in buccal mucosa of dental laboratory technicians is 2.6 times higher than that of the control subjects. Therefore, the mentioned occupation may increase the risk of induction of oral malignant transformations.

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Introduction:

Dental laboratory technicians are regularly exposed to dust and organic solutions which may jeopardize their health.⁽¹⁾ Dental technicians use various materials during the fabrication of dental prostheses. Metal alloys (Cobalt, Chromium, and Nickel) and MMA (methyl methacrylate)-based monomers and polymers are used frequently in dental laboratories. The combination of Chromium and Nickel is known to be correlated with lung cancer. Also, Cobalt and its compounds may be carcinogenic.⁽²⁾ Exposure to MMA can cause asthma, contact dermatitis and headache.⁽³⁾ On the other hand, pneumoconiosis has been reported in dental technicians.⁽⁴⁾ Therefore, dental technicians are advised to wear face masks, eye shields and to use efficient ventilation systems.⁽⁵⁾ Micronucleus is a very small nuclear body which is breaking away from the main nucleus during interphase in cell cycle. Micronuclei are separated from the main nucleus but still added to the cell. They have either complete chromosomes or chromosomal fragments. Micronuclei are one of the biological signs of genotoxicity in human erythrocytes, lymphocytes, reticulocytes and buccal mucosa cells. Increase of micronucleus rate has been a criterion for measurement and diagnosis of aneugenicity and clastogenicity⁽³⁻⁵⁾ and also for studying the genotoxicity of various chemicals.⁽⁶⁻⁸⁾ Micronucleus evaluation has been used in human epidemiology and animal experiments to study clastogene and aneuploidy in injuries caused by occupational and environmental factors.⁽⁹⁾

Many researchers have investigated the effects and disadvantages of laboratory materials and have revealed various results^(9, 10). To the best of our knowledge, there is no comprehensive study assessing the diagnostic value of micronucleus frequency in buccal mucosa cells of dental technicians.

Considering the gap of information and the importance of early diagnosis of DNA injury which may lead to carcinogenesis, this study aimed to evaluate the frequency of micronucleus in buccal mucosa cells of dental technicians, at dental branch of Islamic Azad University in 2013.

Methods and Materials:

This historical-cohort study was designed following a pilot study with 95% confidence rate and 90% power of analysis. The study design has been approved by the ethical committee of Islamic Azad University of Medical Sciences. Sixteen dental laboratory technicians were placed in the case group and sixteen individuals served as the control group. The subjects in case group had been working for at least a year in dental laboratories of school of dentistry of Islamic Azad University of Medical Sciences. The laboratory was equipped with ordinary ventilation systems. All subjects were matched according to gender, age, residing city (Tehran) and socio-economic status. All subjects signed an informed consent. Individuals with recent viral diseases, consumers of specific medications, cigarette smokers, and alcoholics, individuals with addiction to narcotics, risky occupations and a history of radiotherapy were excluded from the study.

First, subjects of case group were requested to wash their mouth thoroughly with water before sampling. Buccal mucosa cells were scraped by a wet plastic spatula and were smeared on clean microscope glass slides. The smears were fixed by Pathofix spray (Padtan Teb co., Tehran, Iran) and were dried at room temperature. Afterwards, the smears were stained with Papanicolaou stain for cytological assessment. Micronucleus assessment was performed under light microscope (400×). The criteria used by Talbert et al. were adapted for micronucleus evaluation.⁽¹¹⁾ 500 cells were counted in each sample and micronucleus frequency was assessed,⁽¹²⁾ (Fig. 1)



Figure 1- Papanicolaou staining of buccal mucosa cells showed a cell with micronucleus under light microscope (400×)

T-test was used for statistical analysis. Significance level was set at 0.05.

Results:

The present study was conducted on sixteen male dental laboratory technicians with the average age of 36 ± 19 years as well as sixteen male control samples with the average age of 39 ± 88 years. Frequency of micronucleus in buccal mucosa cells is shown in Table 1.

Table1- Micronucleus frequency in buccal mucosa cells

Micronucleus frequency	Mean \pm SD	P-value
Dental laboratory technicians	69 \pm 70	P<0.001
Control group	27 \pm 86	

Micronucleus frequency equaled 69 ± 70 and 27 ± 8.6 in case and control samples, respectively; which is 2.6 times higher in the case group. T-test showed that the difference in micronucleus frequency was significant between the two groups. ($P < 0.001$) Variation coefficient or homogeneity rate of micronucleus in both groups was almost similar and equaled 25%.

Discussion:

The present study showed that the frequency of micronucleated cells was significantly higher in the case group. Dust scattering during technical laboratory procedures and exposure to organic solutions can jeopardize dental technician's health.⁽¹⁾ Various materials are used in prosthesis fabrication procedures that contain metal alloys (cobalt, chromium, nickel) and MMA-based monomers and polymers. Exposure to combinations of chromium and nickel has been reported to correlate with cancer. Moreover, Cobalt and its compounds may be carcinogenic to humans.⁽²⁾

Considering the expensive and complicated treatment procedures, also prolonged and challenging rehabilitation processes during the treatment of malignant lesions, nowadays, research methods are based on early diagnosis and more importantly, screening of high risk population. Moreover,

noninvasive methods such as micronucleus assessment of buccal mucosa cells may be helpful in the early detection of nuclear changes in oral mucosa cells which can be an indicator of dysplastic alteration before clinical manifestations. The present study assessed micronucleus frequency in buccal mucosa of dental laboratory technicians and it showed that the average percentage of micronucleated cells was significantly higher in the case group. This finding is in line with the results of a similar study showing that dental laboratory technicians presented higher micronucleus frequency in their cells.^(2, 3, 4) Azhar et al. studied cell toxicity of MMA monomers by use of micronucleus assay in dental technicians. They studied dental laboratory technicians that had been exposed to MMA and concluded that micronucleus frequency in dental technicians doesn't differ from that of the control group⁽¹⁰⁾. Other studies have shown that the occupation period is an important factor which increases the rate of micronucleated cells.⁽¹³⁾

Use of efficient ventilation systems can be the reason for the differences between the results of our study and Azhar's. Another important issue is the consideration of safety principles such as hand washing and wearing face masks and hand gloves in the laboratory. Washing the hands is an effective way to decrease absorption of organic solutions and monomers by the skin.⁽¹⁰⁾ Ishikawa et al. showed that washing hands in work environment significantly decreases the rate of micronucleus however use of hand gloves doesn't have a significant effect. In the mentioned study, individuals used surgical face masks which are efficient against liquids while using filtered and gas face masks is recommended for protection against gas and dust. Nevertheless, the researchers suggested that wearing face mask is beneficial⁽⁹⁾. The mentioned study evaluated peripheral blood cells and showed that occupation as a dental technician was a risk factor that significantly increased micronucleus frequency.⁽¹⁴⁾

Staining methods used in the detection of micronucleus in exfoliated cells of oral mucosa varies in different researches and include Feulgen-Fast Green, fluorescent colors such as diamidino-2-phenylindole (DAPI), Acridine orange, Propidium iodide, May-Grunwald-Giemsa and Papanicolaou⁽¹⁴⁾. Feulgen-Fast Green is more commonly used due to its specificity for DNA and its clear

staining of cytoplasm which eases micronucleus count. ⁽¹⁵⁾ Papanicolaou stain has been also used in many recent studies ^(10, 16, 17) with acceptable results. This method excludes complex processes such as cell culture, metaphase preparation and DNA specific staining. In a research by Pradeep et al. one μ lit concentration of MMA prevented cell growth, decreased the number of vital cells and increased cell death ⁽¹⁸⁾ which proves the toxic effect of this material. On the other hand, some controversies are noticeable in various studies regarding the role of gender since some showed higher micronucleus rates in males and some others showed them to be higher in females. ⁽¹⁹⁻²¹⁾ The present study only involved male subjects in order to exclude the confounding effect of gender. As a final point, it seems that the majority of the previous studies are in line with our results and have reported increased micronucleus frequency in buccal mucosa cells of dental laboratory technicians. However, further investigations with greater sample volume are recommended.

Conclusions:

The present study showed that the average percentage of micronucleated cells was significantly higher in buccal mucosa cells of dental laboratory technicians compared with that of the control group. Therefore, the mentioned occupation may be a predisposing factor in the induction of nuclear changes in oral mucosa cells with increased susceptibility to oral malignant transformations.

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