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RESPONSE TO THE LETTER TO THE EDITOR

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The growing importance of cytogenetic biomonitoring studies in buccal mucosa of dental laboratory technicians

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We are grateful to the author of the Letter to the Editor for providing feedback about our manuscript titled "Determination of cytogenetic abnormalities in buccal mucosa of dental laboratory technicians"[1] and to the Editors for giving us an opportunity to respond. We believe that our response will be of interest to readers of the Archives of Environmental & Occupational Health.

In our article, we focused on the micronucleus (MN) analysis formed in the nucleus of the buccal cells of the technicians working in the dental prosthesis laboratory but focused on the anomaly cells that may occur as a result of exposure to fine particles, and the toxic and carcinogenic metals they contain during the construction of the dental prosthesis. As mentioned in the Letter to the Editor, the cells of the oral mucosa were examined, and micronucleus counts were determined. In addition, the types of nuclear anomalies (micronucleus, Pyknotic nuclei, Chromatin condensation, Broken-egg, Karyolytic cells, Karyorrhexis, Binucleated cells, Nuclear Bud, etc.) and their frequencies were investigated. Micronucleus formations were generally observed within these nuclear anomalies. The author stated that May-Grunwald dye should not be used while performing micronucleus analyzes in these anomaly cells. However, previous studies utilizing micronucleus analyzes in buccal cells performed with May-Grunwald is abundant in the literature.[2–7] To give an example, in Bonassi et al.'s[8] study, 14 (30%) of 46 laboratories stated that they prepared smear preparations for buccal cells with May–Grunwald Giemsa. However, as recommended, in future studies, micronucleus and cell anomalies can be examined with dyes such as acridine orange, DAPI, and Feulgen, etc. In order to avoid false positivity such as cytoplasmic granules, leukocytes and bacteria, in our study, attention was paid to the preparation and oral hygiene during the sample collection. In addition, anomalies in cells that were only seen one-to-one, except for groups of cells stacked on top of each other in sample examinations, were examined. Our cells in Figure 1 (Yuksel & Arsal Yildirim, 2021; original article) were examined at 100 magnification and analyzed on the cells observed individually in the preparation.[1]

Various factors affecting MN in buccal cells contain cell collection (timing and instruments used), selection of counted cells, number and scoring criteria, fixation and staining techniques, and differences between further nuclear anomalies in normal and degenerate cells. Several these methodological things for buccal cells overlap with testing for MN in lymphocytes, however changes in tissues may impact to changeability. Therefore, the statistics obtained on the MN test in human lymphocytes have been reported by investigators that a specific validation study is needed for buccal cells.[5,9]

The author in the Letter to the Editor mentioned that collecting samples from different parts of the mouth might reveal different findings depending on the oral cavity region. However, this is only argued as a personal opinion without basing it on any external reference. However, in literature reviews, it was stated by various researchers that buccal mucosal cells could be collected by wiping or gently scraping the mucosa from one or both cheek areas with a wooden tongue depressor, a metal spatula, toothpick or toothbrushes (e.g., Theda et al., 2018).[5,10,11–17] As can be seen from the studies published in reputable journals in the field, collecting samples from different parts of the mouth may not cause an erroneous result in the data.

As mentioned at the beginning of our article, our study investigated the genotoxic effects of the fine particles and the toxic and carcinogenic metals they contain on the buccal mucosa cells during the production of dental prosthesis and we aimed to raise awareness regarding the occupational risks of dental

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laboratory technicians. We tried to detect anomalies in buccal cells rather than micronucleus analysis. By examining the oral mucosa cells, the types of core anomalies (micronucleus, Pyknotic nuclei, Chromatin condensation, Broken-egg, Karyolytic cells, Karyorrhexis, Binucleated cells, Nuclear Bud) and their frequencies were investigated. All experiments were performed in two replicates. A total of 1000 cells were counted, 500 in each preparation. A total of 15,000 cells were analyzed from an oral site of 30 controls, 30 DPL workers. Thus, 30,000 cells for each group and 60,000 cells for both groups were examined. Statistical analyzes were run using these data. We believe that our study makes an original contribution to the field, albeit its limitations, and we are glad that it receives the attention of the scholars in our field. We hope to contribute more to this line of research and attract the attention of the researchers on the hazards faced by the DPL workers.

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