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INVESTIGATION OF GENOTOXIC EFFECT OF CHEMOTHERAPY IN PATIENTS OF THE PULMOLOGY CLINIC IN A HOSPITAL OF PRISHTINA

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Summary

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Our aim was to investigate the adverse effects of occupational exposure to medicaments and in a control group. In the present investigation, 15 patient and 15 control subjects were enrolled for DNA damage analysis in buccal cells by micronucleus assay. Patients showed a significant increase in micronucleated cells when compared to control group.

Key words: genotoxic effect, medicaments

INTRODUCTION

Within epidemiologic cancer research, molecular epidemiology is an area of increasingly intense activity and interest, combining molecular biology and epidemiologic methods to strengthen epidemiologic evidence.

Molecular epidemiology research focuses on three types of biomarkers: biomarkers of exposure (eg, cytogenetic endpoints – chromosomal aberrations, micronuclei, and sister chromatid exchanges), biomarkers of susceptibility (eg, genetic polymorphisms), and biomarkers of disease (eg, tumour biomarkers).

Casartelli *et al.* (2000) observed MN frequencies in exfoliated buccal cells in normal mucosa, precancerous lesions, and squamous cell carcinoma. They concluded that the gradual increase in MN counts from normal mucosal to precancerous lesions to carcinoma suggested a link of this biomarker with neoplastic progression. It is notable that the first studies of Stich *et al.* (1983; 1984) conducted between 1983 and 1984 had higher baseline MN frequencies than subsequent studies.

MATERIAL AND METHODS

The study population consisted of 15 patients (13 males and 2 female) and a control group of 15 healthy subjects. The youngest patient was 17 years old, and the oldest – 71 years old. The patients were subjected to chemotherapy to cure cancer in lungs. Buccal cells were sampled with toothbrush from the inside of the cheeks and placed in physiological solution (NaCl 0.9%). The cells were washed thrice in the buffer solution by centrifugation. After centrifugation (10 min at 1000 rpm), the pellet was fixed in 3:1 methanol/acetic-acid for 10 minutes. Five slides were prepared for each subject and 1000

Table 1. Frequency of micronuclei (MN) in patients after chemotherapy, scored to 1,000 buccal cells

	MN in chemotherapy patients	MN in control group	Significance
Total MN number (female and male)	108 MN	41 MN	
Mean MN (female and male)	7.20 MN	2.73 MN	P=0.022
Total MN number (females)	13 MN	5 MN	
Mean MN (females)	6.5 MN	2.5 MN	P=0.005
Total MN number (males)	95 MN	43 MN	
Mean MN (males)	7.30 MN	3.3 MN	P=0.040
Oldest group	5.25 MN	4.87 MN	
Youngest group	4.14 MN	2.57 MN	

cells are scoring (at 100× magnification), from each subject were examined. The cells were stained in 10% Giemsa solution.

RESULTS

The results were presented in Table 1 and Fig. 1–3. The results were separated according to gender and age.

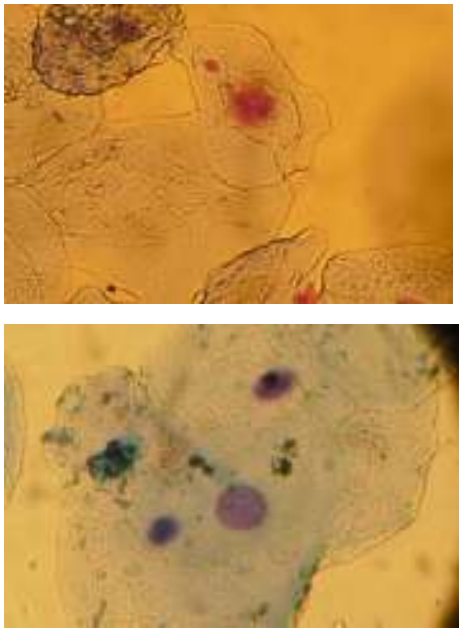

Fig. 1. Micronucleus in buccal cells.

The frequency of micronuclei in buccal cells after chemotherapy in patients with cancer lung was 7.20 MN/1000 buccal cells. It was statistically significantly higher (P=0.022) compared with MN in the control group (2.73 MN/1000 buccal cells).

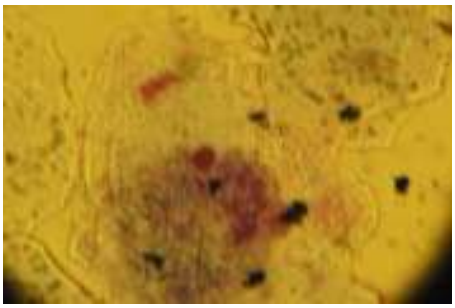

Fig. 2. Kariolysis and pyknosis.

When we analysed the frequency of MN after separating the patients according the gender we found a higher but not statistically significant frequency of MN in males (7.33 MN/1000 cells) compared with females (6.5 MN/1000 cells). Also, the frequency of micronuclei in male subjects (7.33 MN/1000 cells) was statistically significantly higher compared with male controls (3.3 MN/cells; P=0.040). The same was valid for the difference between females and female controls (P=0.005).

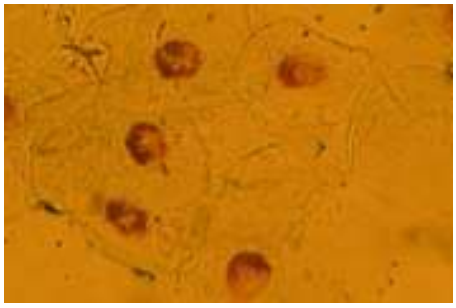

Fig. 3. Karrhyorhexis.

DISCUSSION

In the last 15–20 years, the MN assay has been applied to evaluate chromosomal damage for biologic monitoring of human populations exposed to a variety of mutagenic and carcinogenic chemical or physical agents, such as antineoplastic drugs used for hospital staffs, areca nut chewers, arsenic in drinking water, dioxin as fertilizer, ethylene oxide, formaldehyde, lead oxide, solvents, benzene, ozone, polycyclic aromatic hydrocarbons, chlorants, toxic gases, pesticide mixtures, toluene, hexane, acetone, methyl-ethyl-ketone, 2-trans-hexol, and all forms of tobacco.

A broad range of baseline MN frequencies has been reported (0.05–11.5 MN/1000 cells) with the majority of values between 0.5 and 2.5 MN/1000 cells. There is no clear pattern of the variations among laboratories from different countries. Many studies report a statistically significant elevation of MN levels in exposed individuals compared with control groups, although the observed effects are relatively small, ranging between 1.1 and 4–fold (Holland *et al.,* 2007) and many other studies report changes that were not statistically significant (Machado-Santelli *et al,*. 1994; Martinez, 2005; Pastor *et al.*, 2002). A 12-fold increase in MN frequency was observed in a study by Suruda *et al.* (1993). This increase was unusually large, but has been confirmed by independent analyses using a different staining procedure.

CONCLUSIONS

According to this investigation, we can conclude: 1) Chemotherapy induced increased number of micronuclei in the buccal cells but not statistically significant compared with control group; 2) Males had greater average number of MN (7.33 MN) compared with females (6.5); 3) Higher frequency of micronuclei was observed in the oldest group (5.25 MN) compared with the youngest group (4.14 MN), but the difference was not statistically significant. 4) Age correlated with micronucleus frequency.

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