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Analysis of sister chromatid exchanges in mint factory workers.

Original Article

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SUMMARY.

Introduction. Sister chromatid exchanges (SCEs) were analysed in the peripheral lymphocytes of mint factory workers exposed to metal alloys of aluminium, copper, nickel and magnesium.

Materials and methods. Heparinised blood samples were collected from mint factory workers (63 nonsmokers and 67 smokers), 54 nonsmokers and 42 smokers belonging to the same socioeconomic group and not exposed to toxic chemicals. All the samples were studied for frequency of sister chromatid exchanges. The exposed population was subgrouped into 3 categories based on duration of exposure.

Results. There was a significant increase in the frequency of SCEs in the exposed group when compared to controls. The frequency of SCEs increased with duration of exposure to metal dusts. Further more the increase was significant at all durations of exposure when compared to control group.

Discussion. The present study reveals that prolonged exposure of man to these metals may result in genetic damage. (*Rev Biomed 2003; 14:11-15*)

Key words: Metal alloys, sister chromatid exchanges, genotoxicity, metal dusts, occupational exposure.

RESUMEN.

Análisis de intercambios cromátides hermanas en empleados de las casas de moneda.

Introducción. Se analizaron intercambios de cromátides hermanas (ICH) en los linfocitos periféricos de los empleados de una fábrica expuestos a las mezclas metálicas de aluminio, cobre, níquel y magnesio.

Materiales y métodos. Se coleccionaron muestras de sangre de empleados de una fábrica (63 no fumadores y 67 fumadores), 54 no fumadores y 42 fumadores quienes pertenecían al mismo grupo socioeconómico y no expuesto a químicas tóxicas. Todas las muestras fueron estudiadas por la frecuencia de los ICH. La población expuesta fue subdividida en tres categorías basadas en la duración de la exposición.

Resultados. Hubo un incremento significativo en la frecuencia de ICH en el grupo expuesto comparado con los controles. La frecuencia de los ICH se

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incremento con la duración de la exposición a polvos metálicos. Además, el incremento fue significativo en todas las duraciones de exposición cuando se comparó al grupo de control.

Discusión. Este estudio revela que la exposición prolongada de personas a estos metales puede producir daño genético.

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Palabras clave: Mezclas de metales, intercambio de cromátides hermanas, genotoxicidad, polvos metálicos, exposición ocupacional.

INTRODUCTION.

Industrial chemicals constitute one of the major areas of human exposure. Many are apparently proved to be potential mutagens, allergens, carcinogens and teratogens. A large fraction of the human population is continuously being exposed to potentially toxic chemicals such as drugs, pesticides, industrial chemicals, metals, organic solvents, etc. Apart from this, metals are also released into the environment from various anthropogenic sources such as the smelting of metallic ores, industrial fabrication, and the commercial application of metals in a wide spectra of industries. The genotoxic effects of metals are reported in various test systems ranging from microorganisms to occupationally exposed workers.

A high incidence of chromosomal aberrations were reported in rats fed with aluminium sulphate (1). Increase of fetal loss was seen in swiss mice fed with aluminium hydroxide (2). High mortality due to malignant neoplasms and lung cancer was observed in aluminium plant workers (3). Teratogenic effects of coppersulphate, benlate and dichlofluanic was reported (4) in drosophila. Injections of cupric acetate to fresh water fish clarius induced teratogenic responses in the level of hormones (5). Tachon (6) reported that cupric ions brought single strand breakage when tested on purified DNA. High incidence of lung cancer was recorded in plating factory workers exposed to chromium, copper and nickel (7).

Nickel is a well established carcinogen. Furst and Schldauder (8) reported malignant tumors in hamsters treated with metallic nickel powder. Jacquet and Mayence (9) reported reduced fertilisation in mice treated with nickel nitrate. Senft *et al.* (10) revealed a high incidence of chromosomal aberrations in workers occupationally exposed to nickel.

As several earlier studies indicated the frequency of sister chromatid exchanges increases when cells, animals or humans are exposed to toxic chemicals the present study was conducted to observe the frequency of sister chromatid exchanges (SCE) in peripheral lymphocytes of mint factory workers exposed to metal alloys-alumag (aluminium and magnesium) and cupronickel (copper and nickel).

MATERIALS AND METHODS.

Occupational exposure of workers to metal alloys.

In the mint factory the metal alloys were heated (alumag was heated to 660°C and cupronickel was heated to 1450°C) in open fire furnaces. The metal alloys were then cooled (annealing) and made into sheets. The sheets were cut and processed into currency coins of different denominations. Thus the workers were exposed to dust and fumes of metal alloys.

Selection of subjects.

The exposed group comprised of one hundred and thirty workers out of which sixty three were nonsmokers and sixty seven were smokers. The duration of their service ranged from 3-27 years and they worked for 8 h/day. The workers were further divided into three groups based on the duration of exposure. For comparison ninety-six age matched subjects belonging to the same socioeconomic group as that of workers were selected. Out of the ninety-six subjects fifty four were nonsmokers and forty two were smokers. The control subjects were not exposed to any toxic chemicals.

Blood Collection.

The blood was collected from one hundred and thirty workers and ninety-six age matched controls. 2

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mL of venous blood was collected from each individual in heparinized and sterile bottles.

Culture Methods.

Heparinised whole blood (0.5 mL) was added to RPMI 1640 medium supplemented with 25% human AB serum, 0.5% phytohemagglutinin, 0.25% discristinin and 0.25% gentamycin. 3µg/mL of bromodeoxyuridine (Brdu) was added to the cultures at the time of initiation. The cultures were then kept in incubation at 37° C for 72 h and harvested according to the standard method (11). All the slides were kept in dark for aging . Three days old slides were stained using the fluorescence plus giemsa technique of Perry and Wolff (12).

RESULTS.

Table 1 depicts the data of SCE in the exposed nonsmokers. The incidence of SCE in exposed smokers is shown in table 2.

In the control group there was a significant increase in the mean SCE rate per cell in the smokers (control II) (6.48) when compared to non smokers (control I) (2.00).

In the nonsmokers exposed to metal alloys a significant increase of mean SCE rate per cell (6.12) was recorded when compared to non smoker controls (Control I) (2.00). The increase in mean SCE rate per cell (6.36) in the 11-20 years exposed group was insignificant when compared to mean SCE rate per cell (5.14) recorded in the 3-10 years exposure group. Similarly the increase in the mean SCE rate per cell recorded in the 21-27 years exposed group (7.34) was also insignificant when compared to the data recorded from 11-20 years exposure group. The increase in the mean SCE rate per cell at all time intervals was significant when compared to the controls.

The increase in mean SCE rate per cell in the exposed smokers (8.54) was significantly high when compared to control group -I and II. The increase of mean SCE rate per cell recorded were 8.51, 9.31, 13.50 in the 3-10 years, 11-20 years and 21-27 years exposed group respectively. The increase of mean

SCE rate per cell when compared between the different exposed groups was insignificant whereas a significant increase of mean SCE rate per cell was observed at all the intervals, when compared to control subjects.

In both the control group and exposed group a high incidence of sisterchromatid exchanges was recorded in smokers when compared to non smokers.

DISCUSSION.

Our study has revealed a significant increase of SCE in the exposed group when compared to the control subjects. Although reports on cytogenetic damage in mint factory workers are not available the clastogenic potential of each metal constituting metal alloys used in mint factory is well recorded.

Enhanced levels of aromatic DNA adducts were observed in workers of an aluminium plant (13). High incidence of carcinogenic DNA adducts were determined by enzyme linked immunosorbent assay in the peripheral lymphocytes of aluminium industry workers (13). Szyba *et al.* (14) reported mutagenic potential of copper II chromate and dichromate in microorganisms. A high incidence of chromosomal oberrations and sisterchromatid exchanges were reported in electroplating workers exposed to nickel (10).

Carcinogenic potential of metals like aluminium, nickel and copper is also well recorded. Sunderman (15) recorded high incidence of sarcomas in rats treated with nickel copper oxide. Kusiak *et al.* (16) reported an increased risk of stomach cancer in workers occupationally exposed to aluminium powder. Sandstrom and Wall (17) reported high incidence of lung cancer in copper smelter workers. A high occurrence of lung cancer was recorded in nickel mine workers (18).

As all these reports provide strong evidence of clastogenic potential of different metals involved in a mint factory the high incidence of SCE in the periheral lymphocytes of mint factory workers observed in the present study can be attributed to the mutagenic effect of metals used in mint factories. As the metals are used as alloys the mutagenic effect can be regarded as a

Table 1
Frequency of sisterchromatid exchanges (SCE) in nonsmokers employed in mint factory.

Group & duration of exposure in years	Number of samples	Number of metaphases screened	Total number of SCEs	SCE/Cell \pm SD
Control Group - I	54	2430	6321	2.00 \pm 0.04
Exposed Group				
3 - 10 Y	25	1125	5789	*5.14 \pm 0.09
11 - 20 Y	22	990	6300	*6.36 \pm 0.12
21 - 20 Y	16	720	5286	*7.34 \pm 0.14
Total				
3 - 27 Y	63	2835	17375	*6.12 \pm 0.07

* P < 0.05 Control I = nonsmokers; Control II = Smokers; 45 metaphases were analysed for each sample.

cumulative effect of different metals used in mint factories.

The present study also revealed a significant SCE in the smokers when compared to non smokers. The effect can be attributed to the synergistic effect of smoking and exposure to metal alloys. Our study confirms previous reports which revealed high incidence of chromosomal aberrations in the workers exposed to pesticides (19). The results also agree with our

report where high incidence of chromosomal aberrations were reported in the smokers employed in mint factory (20).

This present study suggests that undue exposure of man to metal dusts may result in genetic damage.

REFERENCES.

1.- Roy AK, Sharma A, Talukdar G. Effects of aluminium salts on bone marrow chromosomes in rats *invivo* cytobios 1991;

Table 2
Frequency of sisterchromatid exchanges (SCE) in smokers employed in mint factory.

Group & duration of exposure in years	Number of Samples	Number of Metaphases Screened	Total Number of SCEs	SCE/Cell \pm SD
Control Group - I	54	2430	6321	2.00 \pm 0.04
Control Group - II	42	1890	12248	*6.48 \pm 0.07
Exposed Group				
3 - 10 Y	22	990	8432	*8.51 \pm 0.11
11 - 20 Y	30	1350	12580	*9.31 \pm 0.10
21 - 27 Y	15	675	7090	*13.50 \pm 0.14
Total				
3 - 27 Y	67	3015	25753	*8.54 \pm 0.07

* P < 0.05 Control I = nonsmokers; Control II = Smokers; 45 metaphases were analysed for each sample.

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- 66:105-11.
- 2.- Colomina MT, Gomez M, Domingo JL, Corbell J. Teratogenic effect of aluminium hydroxide in Swiss albino mice. *Pharmacol Toxicol* 1994; 74:236-9.
- 3.- Carta P, Couo PL, Flora C, Pau M, Grussu M, Cherchi P. Mortality in workers of a primary aluminium foundry in Portovesme in Sardinia. *Med Lav* 1992; 83:530-5.
- 4.- Merchal-Seault D. The effect of commercial preparations of benlate, dithane M-45, copper sulphate and dichlofluanid on the fertility of *Drosophila melanogaster*. *J Environ Sci Health* 1993; 28:397-411.
- 5.- Panigrahi A, Darmahapatra AK, Medda AK. Effect of lead, zinc, mercury and copper with and without estrogen on serum vitellogenin level in magur fish (*Clarius batrachus*). *Geenbaurs Morphol Jahrb* 1990; 136:775-80.
- 6.- Tachon P. DNA single strand breakage by H₂O₂ and ferric or cupric ions, its modulation by histidine. *Free-Radic Res Common* 1990; 9:39-47.
- 7.- Horiguchi S, Morinaga K, Endo G. Epidemiological study of mortality from cancer among chromium platers. *Asia Pac J Public Health* 1990; 4:169-72.
- 8.- Furst A, Schlauder MC. The hamster as a model for metal carcinogenesis. *Proc West Pharmacol* 1971; 16:150-3.
- 9.- Jaquet P, Mayence A. Application of the in vitro embryo culture to the study of the mutagenic effects of nickel in male germ cells. *Toxicol Lett* 1982; 11:193-7.
- 10.- Senft V, Losan F, Tusek M. Cytogenetic analysis of chromosomal aberrations of peripheral lymphocytes in workers occupationally exposed to nickel. *Mut Res* 1992; 279:171-9.
- 11.- Moorhead PS, Nowell PS, Mellmann WJ, Battups DM, Hungerford DA. Chromosomal preparation of leukocytes cultured from human peripheral blood. *Exp Cell Res* 1960; 20:613-5.
- 12.- Perry P, Wolff S. New giemsa method for the differential staining of sister chromatids. *Nature* 1974; 251:156-8.
- 13.- Schoket B. Monitoring occupational to exposure carcinogens. *IARC Sci Publ* 1993; 124:341-7.
- 14.- Szyba K, Golonka MC, Gaslorowski K, Urban J. Mutagenic activity of copper (II) chromate and dichromate complexes. with polypyridines. *Biometals* 1992; 5:157-61.
- 15.- Sunderman FN. Evolution of clinical science. An overview. *Ann Clin Lab Sci* 1993; 23:231-48.
- 16.- Kusiak RA, Rilchie AC, Springer J, Muller J. Mortality from stomach cancer in ontario miners. *Por J Ind Med* 1993; 50:117-26.
- 17.- Sandstrom A, Wall B. Cancer incidence among male salaried employees at a smeltery in northern Sweden. *Acta Oncol* 1993; 32:9-14.
- 18.- Arrouisalfz FZ, Marzin D, Pestel J. Differences in genotoxic activity of alpha-Ni 352 on human lymphocytes from nickel-hypersensitized and nickel unsensitized donors. *Mutagenesis* 1992; 34:183-7.
- 19.- Rupa DS, Reddy PP, Reddy OS. Chromosomal aberrations in peripheral lymphocytes of cotton field workers exposed to pesticides. *Environ Res* 1989; 49:1-6.
- 20.- Padmaja-Reddy T, Ramana-Devi ChV, Vidyullata V, Hema-Prasad M, Reddy PP. Chromosomal aberrations in smokers exposed to metallic dust in mint factory. *Rev Biomed* 2000; 11:87-90.