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RESEARCH ARTICLE

INCREASED FREQUENCY OF MICRONUCLEI AND SISTER CHROMATID EXCHANGE IN CEMENT FACTORY WORKERS

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The population most exposed to cement dust pollution includes workers and managers in cement plants and factories, families of workers and managers living in staff houses of factories, and other neighbourhood habitations. Children studying in the schools situated in proximity to factories are particularly prone to cement dust exposure. In this study, the micronuclei (MN) and sister chormatid exchange (SCE) was applied in peripheral blood lymphocytes cells, in order to assess the genotoxic risk associated with occupational exposure of cement factory workers. In the present study were analyzed the samples of 62 people working in cement factory including 27 smokers and 35 non-smokers and compared with 62 controls (no exposure to cement dust or any known physical or chemical agent) of the same age group were analyzed using standard micronucleus test protocol. The mean frequencies of MN in the exposed group were significantly higher ($P < 0.000$) when compared to the control group. A significant increase in the micronuclei are observed in the residents with smoking habit (22.351 ± 0.331), when compared to the residents without smoking habit (18.380 ± 0.325) and SCE results also was observed with smoking habit (12.965 ± 0.222) without smoking habit (10.808 ± 0.275). In this study concluded that high number of blood lymphocytes cell micronuclei and sister chromatid exchange induction is possible in case of long term exposure to cement dust and that may cause adverse health impact.

Key words: Cement dust; Micronuclei; Sister chromatid exchange; Smokers, Non smokers.

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INTRODUCTION

Cement factory workers are exposed to dust at various manufacturing and production processes, such as quarrying and handling of raw materials, during grinding the clinker, blending, packing and shipping of the finished products (Abudhaise *et al.,* 1997). High concentration and / or prolonged inhalation of cement dust in cement industry workers can provoke clinical symptoms and inflammatory response that may result in functional and structural abnormalities (Short and Petsonk, 1996). The most frequently reported clinical features in cement mill workers are chronic cough and phlegm production, impairment of lung function, chest tightness, obstructive and restrictive lung disease, skin irritation, conjunctivitis, stomach ache, headache, fatigue (Abou Taleb,1995) and carcinoma of lung, stomach and colon(Rafnsson, 1997).Cement production is invariably a dusty operation resulting in the exposure of factory workers to cement dust. Cement is known to be a mixture of calcium oxide (60-67%), silicon oxide (17-25%), aluminium trioxide (3-8%), and ferric oxide (0-5%) (Fell *et al*., 2003), and is been implicated as a causative agent of silicosis. Silicosis is one civilization's oldest known occupational disease, and is induced by inhalation of crystalline silica (Ding *et al*., 2002). Exposure to silica can result in or contribute to several other diseases including acute silicosis, pulmonary tuberculosis, interstitial fibrosis, rheumatoid complications, vascular diseases, glomerulonephritis, and immunological reactions (Ding *et al*., 2002). Though inflammatory responses resulting from workplace exposures are usually observed in specific target organs, such as the lungs, skin and liver and if persistent, may progress to fibrosis, granulomatous diseases and even cancer (Aminian *et al*., 2008). On the basis of new knowledge, it is possible that cement dust may have effects on the immune and liver functions. People of cement dust zone area badly affected by respiratory problems, gastrointestinal diseases etc (Adak *et al*., 2007).

A study to evaluate the mutagenic effects of occupational exposure to cement dust in such workers concludes that the chromosomal damage was more pronounced in the workers who are also smokers when compared with the non-smokers both in control and exposed groups. A significant increase in the frequency of chromosomal aberrations was also observed with increase in age in both control and exposed subjects (Fatima *et al*., 2001). There is good evidence for cement dust exposure acting as a tobacco, alcohol and asbestos independent risk factor for laryngeal carcinoma (Dietz *et al*., 2004).It has also been found to have mutagenic properties. Chromosomal abnormalities (CA) were induced by exposure to asbestos in ovarian cells and lung cells from Chinese hamsters (Trosic *et al*., 1988) Chrysotile has been reported to induce chromosomal abnormalities in cultured human lymphocytes (Valerio *et al*., 1983). One of the study results was found that asbestos workers had a raised mean sister chromatid exchange (SCE) rate compared with that of controls. In another study a slightly higher incidence of chromosomal aberrations in asbestos exposed factory workers compared with controls was found (Srb, 1984). Our preliminary studies showed enhanced SCE and chromosomal aberrations in human lymphocytes in vitro

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treated with dust from an Indian asbestos cement factory. In the present study the cytogenetic effects of asbestos dust on workers who include both smokers and non-smokers in the same asbestos cement factory and exposed to the same dust have been examined.

Micronuclei MN assay is one of the most sensitive markers for detecting DNA damage, and has been used to investigate genotoxicity of a variety of chemicals. MN testing with interphase cells is more suited as a cytogenetic marker because it is not limited to metaphases, and has the advantage of allowing rapid screening of a larger numbers of cells than in studies with SCEs or CAs. MN analysis therefore appears to be a good tool for investigating the effects of clastogens and aneuploidogens in occupational and environmental exposure in human epidemiological studies. Genetic damage when present in the cells of the germline may be expressed many generations after the primary induction of the genetic change the mutation. (Norppa *et al*., 1993). (Smailyte *et al*., 2004) analyzed the cancer risk in Lithuanian cement producing workers and found that exposure to cement dust may increase lung and bladder cancer. He further reported a dose related risk for stomach cancers (Fatima *et al*., 1997) have reported chromosomal abnormalities in asbestos cement factory workers. (Rahman *et al*., 1996) found chromosomal aberrations, sister chromatid exchanges and micronuclei formation in the blood lymphocytes of asbestos cement factory workers in comparison to their controls. Dusinska *et al*., 2004 investigated chromosomal and DNA damage in former asbestos cement plant workers. The present study was aimed at evaluating and predicting the risk of cytotoxic and genotoxic in subjects exposed to cement dust. The main objective of this study was to evaluate**,** through the MN and SCE in peripheral blood lymphocytes in culture**,** the effect of such exposure in workers from the cement factory.

MATERIALS AND METHODS

Study Sample

The study was conducted on 124 healthy male individuals, 62 cement factory workers and 62 control individuals, participated voluntarily and gave informed consent to the study. Blood samples were obtained by venipuncture using heparinized syringes and needles during a routine health check up only from healthy persons selected by a physician. Samples were taken from the subjects living in and around Chennai and nearby villages. All participants completed a questionnaire and provided a blood sample. The questionnaire elicited demographic data, medical status, drinking, exercising and smoking habits, and prior or current exposure to medication or environmental agents that could affect the MN and SCE assay.

Cytokinesis – blocked micronucleus assay (CBMN)

Blood was obtained from each subject by venipuncture using heparinized vacutainers. lymphocyte cultures were initiated by adding 0.5 ml of whole blood to 5 ml of RPMI 1640 medium supplemented with 15% heat-inactivated fetal bovine serum, 1% antibiotics (penicillin and streptomycin), and L-glutamine. Lymphocytes were stimulated by 1% of phytohemagglutinin and incubated for 72 hrs at 37°C. Two cultures per subject were established. A final concentration of 6.0 µg/ml of cytochalasin B (Sigma, Germany) was added to the cultures 44 hr later to arrest cytokinesis. At 72 hrs of incubation the cultures were harvested by centrifugation at 800 rpm for 8 min and treated with a hypotonic solution (2-3 min in 0.075 M KCl at 4°C). Cells then were centrifuged and a methanol-acetic acid (3:1, v/v) solution was gently added. This fixation step was repeated twice and the resulting cells were resuspended in a small volume of fixative solution and dropped onto clean. Slides were Stained with 10% Giemsa in phosphate buffer (pH 6.8) for 10 min and scored. The frequency of binucleated cells with micronuclei (BNMN) and the total number of MN in lymphocytes (MNL) were determined by blind scoring and slides were stained using giemsa, a total of 1000 binucleated cells with well-preserved cytoplasm for each subject.

Sister chromatid exchange assay (SCE)

Heparinised syringes were used to obtain venous blood samples, which were immediately taken to the laboratory for processing; 0.5 ml of blood were added to 3 ml of RPMI 1640 culture medium with L-glutamine (Gibco) containing 10% fetal bovine serum, 1% phytohemagglutinin-M (4 %, Gibco). The cultures were incubated at 37℃ for 72 h. At 24 h of incubation, we added 5- bromodeoxyuridine (BrdU, Sigma), at a final concentration of 5 mg/mL, to the medium, and 2 h prior to the harvest, we added colchicine (0. 1 µg/ml, Sigma). Metaphase cells were harvested by centrifugation, treated with 0.075 M KCl (Himedia).Cells were then fixed in freshly prepared methanol-glacial acetic acid (3:1) (Merck). The cell suspension was washed twice in fixative, and slides were prepared. A smear was made with these cells, after which the slides were stained by the fluorescence plus-Giemsa technique (Perry and Wolff, 1974). For calculating frequency of SCE per cell, 30 metaphases were analysed as per international practice.

Criteria for selecting binucleated cells which can be scored for micronucleus frequency

The cytokinesis-blocked cells that may be scored for MN frequency should have the following characteristics: (a) The cells should be binucleated; (b) The two nuclei in a binucleated cell should have intact nuclear membranes and be situated within the same cytoplasmic boundary; (c) The two nuclei in a binucleated cell should be approximately equal in size, staining pattern and staining intensity; (d) The two nuclei within a BN cell may be attached by a fine nucleoplasmic bridge which is no wider than 1/4th of the nuclear diameter. (e) The two main nuclei in a BN cell may touch but ideally should not overlap each other. A cell with two overlapping nuclei can be scored only if the nuclear boundaries of each nucleus are distinguishable. (f) The cytoplasmic boundary or membrane of a binucleated cell should be intact and clearly distinguishable from the cytoplasmic boundary of adjacent cells.

Criteria for scoring micronuclei

MNi are morphologically identical to but smaller than nuclei. They also have the following characteristics:(a) The diameter of MNi in human lymphocytes usually varies between 1/16th and 1/3rd of the mean diameter of the main nuclei which corresponds to 1/256th and 1/9th of the area of one of the main nuclei in a BN cell, respectively. (b) MNi are non-refractile

and they can therefore be readily distinguished from artefact such as staining particles; (c) MNi are not linked or connected to the main nuclei; (d) MNi may touch but not overlap the main nuclei and the micro nuclear boundary should be distinguishable from the nuclear boundary; (e) MNi usually have the same staining intensity as the main nuclei but occasionally staining may be more intense. The scoring criteria used in this study were according to the criteria adopted by Fenech *et al*., 2003.

Examination of slides and assessment of MN frequency

Slides are best examined at 400X magnification using a light or fluorescence microscope. Slides should be coded before analysis so that the scorer is not aware of the identity of the slide. A score should be obtained for slides from each duplicate culture. The number of cells scored should be determined depending on the level of change in the MN index that the experiment is intended to detect and the expected standard deviation of the estimate.

Slide scoring

For cytogenetic analysis, preparations were coded and scored blind. Scoring of MN was limited to binucleate lymphocytes only with preserved cytoplasm (Fenech and Morley, 1985), according to the criteria proposed by (Countryman and Heddle, 1976). Identification of binucleate cells in cell groups required careful visual examination of the individual cell boundaries. The results are expressed as the average percentage of micronucleated cells per 1000 binucleate cells on the two different slides from the same culture. When the cytokinesis-block assay is used, it should be kept in mind that some cells may escape the cytokinesis block. Thus some mononucleate cells may actually have divided in the culture, while some binucleate cells may have divided more than once. Care should be taken not to include binucleate cells with irregular shapes or sizes of the main nuclei, as these cells may represent the latter category; neither should binucleate cells be confused with poorly spread multinucleate cells. Cells containing more than two main nuclei should not be analysed for micronuclei as the baseline micronucleus frequency is higher in these cells owing to non-genotoxic effects such as nuclear disintegration.

Statistical analysis

All data analyses were performed using the SPSS Version 17.0. Data were double entered, and the resulting data sets were compared and checked for completeness and accuracy of entry. The significance of differences between the mean for the two groups was assessed with Student's t-test, the differences in means were illustrated using box plots, relationships between micronuclei and chromatid exchange levels were assessed using correlation, and discriminant analysis has been used to differentiate the control and pan-chewers groups using micronuclei and chromatid levels. A p value < 0.05 was considered to be statistically significant. Descriptive statistics were applied for calculating the distribution of various characteristics.

RESULTS

Cytogenetic analysis of micronuclei and sister chromatid exchange have been applied to the study of individuals result was showed in the exposure of cement factory workers and compare to healthy controls. The present study results of mean and standard error (SE) of age, MN, and SCE levels of cement workers of the two groups and the validate independent samples t-tests are given in Table 1. The mean level of micronuclei was significantly higher in cement factory workers (20.750 ± 0.343) than controls (6.373 ± 1.406) and the level of sisterchromatid exchange also was observed higher in the exposure group (12.095 ± 0.218) than controls (5.371 ± 1.268). Overall the mean frequencies of MN and SCE in the exposed group were significantly higher ($P < 0.000$) when compared to the control group. The mean ages of study groups (45.145±1.378) than controls (42.887 ± 1.433). The increased effects of aging on MN and SCE frequency might reflect accumated genetic damage occurring during the life span.

Table 1. Summary Statistics and t-test values: Control and Study Groups

Parameters	Control (N = 62) Mean ± SE	Study (N = 62) Mean ± SE	Independent Samples t-test t	Sig.
Age	42.887 ± 1.433	45.145 ± 1.378	-1.136	.258
Micronuclei	6.373 ± 1.406	20.750 ± 0.343	-37.177	.000
Sister Chromatid Exchange	5.371 ± 1.268	12.095 ± 0.218	-24.796	.000

The t-tests indicate that the mean ages of the workers in the study and normal group remain the same since the P-value is greater than 0.05 (i.e., P = 0.258). The mean values of MN and SCE are found to be statistically different between normal and control group. The P values of the two parameters are less than 0.001 (i.e., P = 0.000). It is evident from the study that both MN and SCE levels are higher among people working in cement industries compared to control. The Box Plots results were given evident from the plots that the mean values of age, MN, and SCE are found to be higher in the study group and compared to the control group as shown in Figure1. The t-tests indicate that the mean ages of the workers in the study and normal group remain the same since the P-value is greater than 0.05 (i.e., P = 0.258). The mean values of MN and SCE are found to be statistically different between normal and control group. The P values of the two parameters are less than 0.001 (i.e., P = 0.000). It is evident from the study that both MN and SCE levels are higher among people working in cement industries compared to control. The Box Plots results were given evident from the plots that the mean values of age, MN, and SCE are found to be higher in the study group and compared to the control group as shown in Figure1. For the conformation study to find out whether there is relationship between age, MN, and SCE levels in the two groups, correlation analysis has been carried out and the results are presented in Table 2. It is clearly evident from the correlation analysis that age, MN, and SCE are highly correlated ($p < 0.01$) with one another among workers in both the groups. To test whether exposure to cement particles and smoking has got anything to do with the MN and SCE levels of the cement workers, the following analysis were carried out: independent samples t-tests and Analysis of Variance (ANOVA). To test whether smoking has got any effect on the cement workers, the independent samples t-test has been applied. The exposure duration has been classified in to three categories, 10 years or less, 11 to 15 years, and more than 15 years. To test whether mean levels of MN and SCE differ between the three groups of exposure duration, ANOVA has been applied. The summary information of age, MN, SCE levels of smokers and non-

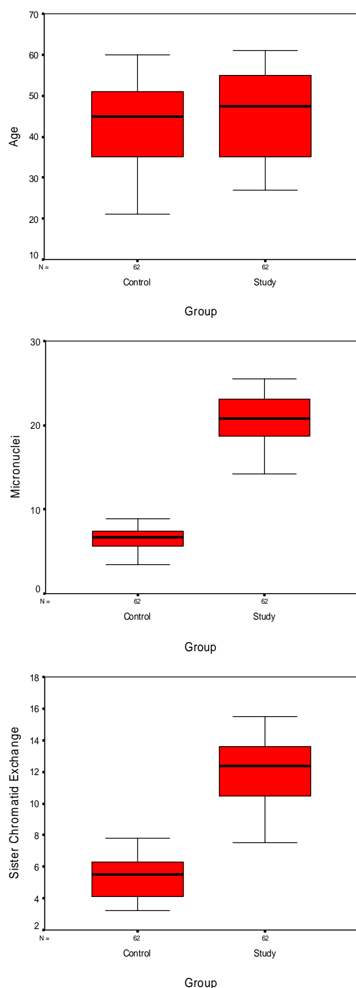

Figure 1: Box Plots - Comparison of age, micronuclei and sister chromatid exchange

Table 2: Correlations analysis in Study and Control Groups

Group		Micronuclei	Sister Chromatid Exchange
Control	Age	.873 **	.864 **
	Micronuclei	1	.893 **
Study	Age	.895 **	.793 **
	Micronuclei	1	.777 **

** Correlation is significant at the 0.01 level (2-tailed).

smokers working in cement industry is given in Figure 2. A significant increase in the micronuclei are observed in the cement workers with smoking habit 22.351 ± 0.331), when compared to without smoking habit (18.380 ± 0.325) and SCE results also was observed with smoking habit (12.965 ± 0.222) without smoking habit (10.808 ± 0.275). The mean levels of MN and SCE frequencies with smokers had significantly higher than non-smokers (P < 0.000). The t test values for smokers and non-smokers of the study group are given in Table 3. The average age of a cement worker having the habit of smoking is nearly 53 years and that of a non-smoker working in the same industry is around 34 years. Micronuclei level of a smoker is nearly 22 whereas it is 18 in the case of a non-smoker. Similarly, the average SCE level of a smoker working in Cement industry is about 13 and that of a non-smoker is about 11. The t-tests indicate that the mean values of the three parameters studied are found to be

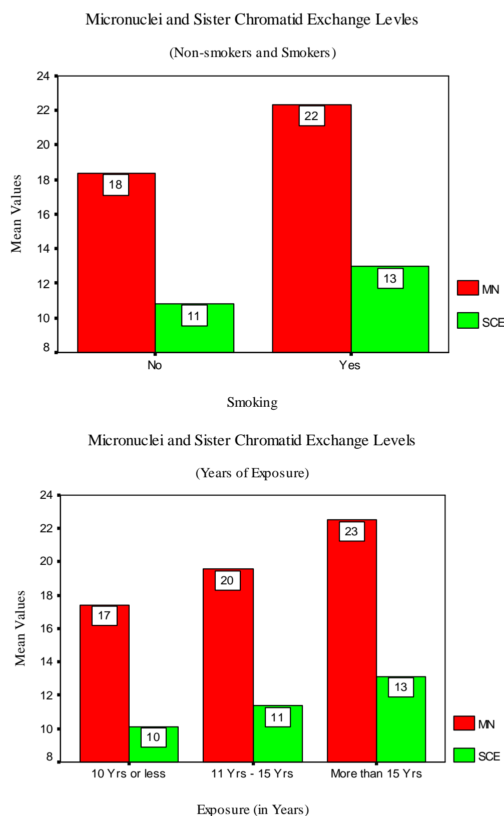

Fig. 2: Micronuclei and Sister Chromatid Exchange Levels among (i) Non-smokers and Smokers and (ii) Different years of Exposure

Table 3: Summary Statistics and t-test values – Smokers and Non smokers in Study Group

Study Group - Parameters	Smokers (N = 37) Mean ± SE	Non-smokers (N = 25) Mean ± SE	Independent Samples t-test t	Sig.
Age	52.676 ± 0.941	34.000 ± 1.156	12.551	.000
Micronuclei	22.351 ± 0.331	18.380 ± 0.325	8.204	.000
Sister Chromatid Exchange	12.965 ± 0.222	10.808 ± 0.275	6.136	.000

Table 4: Correlations - Study Group

	Micronuclei	Sister Chromatid Exchange
Exposure	.769**	.716**
Micronuclei	1	.777**

** Correlation is significant at the 0.01 level (2-tailed).

significantly different between smokers and non-smokers. The P values are all less than 0,001 (i.e., P = 0.000) in all the four parameters. A close examination of the mean values indicate

Table 5: Comparison of Exposure duration and MN and SCE – Study Group

	10 Years or less (N = 11)	11 Yrs – 15 Yrs (N = 18)	More than 15 Yrs (N = 33)	ANOVA	
	Mean ± SE	Mean ± SE	Mean ± SE	F	Sig.
Micronuclei	17.427 ± 0.432	19.567 ± 0.369	22.503 ± 0.357	37.148	.000
Sister Chromatid Exchange	10.136 ± 0.371	11.378 ± 0.262	13.139 ± 0.231	27.874	.000

that smokers have higher micronuclei and sister chromatid exchange levels compared to non-smokers working in cement industry. Correlation analysis of exposure, micronuclei, and sister chromatid exchange levels of study group is presented in Table 4. It is evident from the analysis that correlation, micronuclei, and sister chromatid exchange are interrelated with one another. Higher the exposure to cement particles among workers of the cement industry results in higher levels of micronuclei and sister chromatid exchange. Similarly when there is an increase in micronuclei level, there is an increase in the sister chromatid exchange levels and vice versa ($p < 0.01$).
It is of interest to know whether years of exposure has got any influence over the MN and SCE levels of workers of cement factory. This has been tested using Analysis of Variance (ANOVA). It has been hypothesized that the mean levels of MN and SCE remain the same irrespective of years of exposure in cement factories. The analysis indicates that both mean micronuclei and sister chromatid levels differ with different years of exposure in Table 5. Post Hoc tests indicated that mean values of the three exposure periods are significantly different from one another both in Micronuclei and Sister Chromatid Exchange levels. It can be concluded that the mean MN and SCE levels vary according to period of exposure among workers of cement industry. A close examination of the table indicates that as the years of exposure to cement particles increases, the mean MN and SCE levels also increases.

DISCUSSION

Cement is produced through a series of processes that includes quarrying, crushing, raw milling, blending, kiln burning to form clinker, cement milling, and packing. Substantial dust is emitted during these processes, exposing workers to dust. Our findings confirm the previously reported higher prevalence of respiratory symptoms among exposed cement workers than among controls. Some studies have also investigated the association between current dust exposure levels and respiratory symptoms among cement workers, with differing results (Abou taleb, 1995). Cement industry is considered as a major pollution problem because of dust and particulate matter emitted at various steps of cement production. The workers are usually exposed to dust through dermal, respiratory routes and to a lesser extent via ingestion. In cement plant workers, the prevalence of chronic phlegm increased with tenure whereas the prevalence of wheezing increased with both tenure and the current dust level. (Abrons *et al*., 1998). The micronucleus is a well-known marker of genotoxicity and any reduction in the frequency of the genotoxic endpoint gives an indication of the antigenotoxicity of a particular compound (Albertini *et al*., 2000). We examined the relationship between demographic variables and lifestyle factors and clastogenicity, as measured by the MN frequency level, in 62 cement factory workers individuals. Our study confirmed by univariate analysis that the frequency of MN in peripheral blood lymphocytes is associated with age, gender and smoking status. The mean level of micronuclei was significantly higher in cement factory workers (20.750 ± 0.343) than controls (6.373 ± 1.406) and the level of sister chromatid exchange also was observed higher in the exposure group (12.095 ± 0.218) than controls (5.371 ± 1.268). Overall the mean frequencies of MN and SCE in the exposed group were significantly higher ($p < 0.000$) when compared to the control group. The higher MN frequencies in males might be due to the more damageable characteristics of Y chromosomes compared with other chromosomes, or due to contributions of unknown factors. The effects of smoking on MN frequencies in this study are supported by previous studies (Tomanin *et al*., 1991).

A Medline search of studies performed in subjects environmentally or professionally exposed to genotoxins (and their controls) that also analyzed the role of cigarette smoking, identified 33 publications out of the 37 evaluated (89.2%) that did not find an association between MN frequency and smoking habits (Da Cruz *et al*., 1994 ; Yager *et al*., 1988).Therefore, if the MN assay perfectly reflects the carcinogenic effect of smoking, then only a small percentage of smokers would be expected to have an increased frequency of MN caused by smoking. The ever-smokers were about three times more likely to report chronic bronchitis and dyspnea than the never- smokers. Therefore, the relative contribution of smoking in the development of respiratory symptoms should be of concern in the cement industry (Abu dhaise *et al*., 1997; Laraqui hossini, 2002). The frequency of micronuclei and sister chromatid was significantly higher in the exposed group than in the controls because of chronic occupational exposure to wood dust leads to an increased risk of genetic damage among workers of wooden furniture industries (Elavarasi *et al*., 2002). One of the study was clearly indicates that pan chewing greatly affect the cells level of micronuclei, sister chromatid exchange and causes chromosomal aberrations such as gaps, breaks, acentric fragments, exchanges, and dicentrics. The results of such studies could have a significant impact on the future use to detect DNA damage in any kind of gonotoxicity and toxicology biomarker (Ramakrishnan *et al*., 2011). Sister chromatid exchange (SCE) is a more sensitive indicator of genotoxic effects (Tucker and Preston 1996). There is an excellent correlation between carcinogenicity and SCE inducing ability of large number of chemicals (Gebhart, 1981). Previous reports have shown that smoking and age are associated with an increase in the SCE frequency (Husum *et al*., 1986). We also found that cigarette-smoking, age and duration of long exposure was associated with increased SCE frequency in this study ($P < 0.000$). The validity of our study was strengthened by the high response rate by both the exposed workers and the controls. Calcium silicate, a component of most silicious dusts including that of cement, significantly increased the frequency of chromosomal aberrations and sister chromatid exchanges (Aslam *et al*., 1993). It was shown previously that asbestos samples collected from an asbestos factory enhanced sister chromatid exchanges and chromosomal aberrations in vitro using human lymphocytes (Rom *et al*., 1983; Fatima *et al*., 2001). Asbestos workers had a raised mean SCE rate and increased numbers of chromosomal aberrations compared with a control population. Most of the chromosomal aberrations were chromatid gap and

break types (Trosic, 1997). All extracts of particulates induced a significant increase of sister chromatid exchanges in a dose dependent manner. Samples from the industrialized area revealed the highest activities (Hadnagy *et al*., 1989). The present study was designed to assess the DNA damage among cement industry workers who are occupationally exposed to cement dust contains heavy metals like nickel, cobalt, lead, chromium, pollutants hazardous to the biotic environment, with adverse impact for vegetation, human and animal health and ecosystems. (Baby *et al*., 2008). Occupational cement dust exposure has been associated with an increased risk of liver abnormalities, pulmonary disorders, and carcinogenesis (Aydin *et al*., 2010). On the basis of the above literature described, it has been demonstrated that cement dust causes chronic obstructive lung disease, restrictive lung disease, lung function impairment, pneumoconiosis, and carcinoma of larynx, lungs, stomach and colon. In addition, it has been suggested that, the components of cement dust also enter into blood stream and may also reaching the different tissues of the body including heart, liver, spleen, bone, muscles and hairs, and affecting their micro-structure and physiological performance. This study is to gather the potential toxic effects of cement dust and to minimize the health risks in cement factory workers by providing them with information about the hazards of cement dust.

CONCLUSION

Cytogenetic biomarkers such as, micronuclei and sister chromatid exchanges have been applied to the study of individuals exposed to known or potential genotoxic agents. The present study suggests that workers exposed to cement dust are prone to genetic damage. The factors that contribute to increased MN and SCE levels are age, years of exposure and smoking habits. Smokers have a synergic effect on inducing DNA damage among cement industry workers. In the cytogenetic analysis, the frequency of micronuclei and SCE analysis was found to increase with the increase in period of exposure to cement dust. Hence there is a directly proportional relationship between the frequency of micronuclei and the period of exposure to cement dust. Also, safety measures have to be improved to prevent occupational health hazard in the cement factory workers.

REFERENCES

Abou Taleb, A.N.M., Musaniger, AO. and Abdel moneim, R.B. 1995.Health status of cement workers in the United Arab Emirates. *J. Roy. Soc. Health*., 2: 378-383.

Abrons, H.L., Peterson, M.R..and Sanderson, W.T. 1998.Symptoms, ventilator function and environmental exposures in Portland cement workers. *Br J Ind Med*., 45: 368-375.

Abudhaise, BA., Rabi, A.Z., Zwairy, M.A.A., Hader, A.F.E. and Qaderi, S.E. 1997. Pulmonary manifestation in cement workers in Jordan. *Int. J. Occup. Med. Environ. Health*.,10: 417-428.

Adak, M. D., Adak, S. and K. M. Purohit, K.M. 2007. Ambient air quality and health hazards near mini cement plants. *Pollution Research*., 26 (3): 361- 364.

Aminian, O., Sharifian, S.A., Mehrdad, R., Narooey, A.K. and Giahi, O. 2008. Humoral Immune system alterations in silica exposed workers. *Iranian J Publ Health*., 3:142-145.

Albertini, R.J., Anderson, D., Douglas, G.R., Hagmar, L, Heminiki, K., Merelo, F., Natarajan, A.T., Norppa, H., Shuker, D.E.G., Tice, R., Walters, N.D. and Aitio, A. 2000. IPCS guideline for the monitoring of genotoxic effects of carcinogens in humans. *Mutat. Res.,* 463: 111-172.

Aydin, S., Croteau, G., Sahin, I. and Citil, C. 2010. Ghrelin, Nitrite and Paraoxonase/Arylesterase concentrations in cement plant workers. *Journal of Medical Biochemistry*., 29(2): 78-83.

Baby, S., Singh, N. A., Shrivastava, P., Nath, S. R., Kumar, S. S., Singh, D. and Vivek K., 2008. Impact of dust emission on plant vegetation of vicinity of cement plant. *Environmental Engineering and Management Journal*., 7(1): 31-35.

Collins, M., Kazimirova, A., Barancokova, M., Harrington, V., Volkovova, K., Staruchova, M., Horska, A., Wsolova, L., Kocan, A., Petrik, J., Machata, M., Ratcliffe, B. and Kyrtopoulos, S.2004. Genotoxic effects of asbestos in humans. *Mutat Res*., 553(1–2):91-102.

Countryman, P.L. and Heddle, J.A. 1976. The production of micronuclei from chromosome aberrations in irradiated cultures of human lymphocytes. *Mutat. Res.*, 41: 321-332.

Da Cruz, A.D., McArthur, A.G., Silva, C.C., Curado, M.P. and Glickman, B.W. 1994.Human micronucleus counts are correlated with age, smoking, and cesium-137 dose in the Goiania (Brazil) radiological accident, *Mutat. Res*., 313: 57–68.

Dietz, A., Ramroth, H., Urban, T., Ahrens, W. and H. Becher, H. 2004. "Exposure to cement dust, related occupational groups and laryngeal cancer risk: Results of a population based case-control study." *International Journal of Cancer*., 108(6): 907-911.

Ding, M., Chen, F., Shi X., Yucesory, B., Mossman, B. and Vallyathan V. 2002.Diseases caused by silica: mechanisms of injury and disease development. *International Immunopharmacology*., 2:173-182.

Elavarasi, D., Ramakrishnan, V., Subramoniam, T., Ramesh, A., Cherian, K., M. and Emmanuel, C.2002. Genotoxicity study in lymphocytes of workers in wooden furniture industry. *Current science*, 82:869 -873.

Fatima, N., Jain, A.K. and ahman Q. 1991 Frequency of sister chromatid exchange and chromosomal aberrations in asbestos cement workers. *Br J Ind Med*., 48:103-105.

Fatima, S. K., Prabhavathi, P. A., Padmavathi, P. and Reddy, P. P. 2001. Analysis of chromosomal aberrations in men occupationally exposed to cement dust. *Mutation Research/Genetic Toxicology and Environmental Mutagenesis*., 490(2): 179-186.

Fenech, M. and Morley, A.A. 1985. Measurement of micronuclei in human lymphocytes". *Mutation Res*., 148: 29-36.

Fenech, M., Chang, W.P., Kirsch-Volders, M., Holland, N., Bonassi, S. and Zeiger, E. 2003. HUMN project: detailed description of the scoring criteria for the cytokinesis-block micronucleus assay using isolated human lymphocyte cultures. *Mutation Research*., 534: 65-75.

Fell, A.K., Thomassen, T.R, Kristensen, P., Egeland, T. and Kongland, J. 2003. Respiratory Symptoms and Ventilatory Function in Workers Exposed to Portland Cement Dust., *JOEM* 45:1008-1014.

Gebhart, E. 1981. Sister chromatid exchange (SCE) and structural chromosomes aberrations in mutagenicity testing. *Hum. Genet*., 58: 235-254.

Husum, B. H.C. and Wulf, E.1986. Niebuhr, Sister chromatid exchange frequency correlates with age, sex and cigarette smoking in a 5-year material of 553 healthy adults. *Hereditas*., 105: 17–21.

Hadnagy, W., Seemayer, N.H., Tomingas, R. and Ivanfy, K. 1989. Comparative study of sister chromatid exchanges and chromosomal aberrations induced by airborne particulates from an urban and highly industrialised location in human lymphocyte cultures. *Mut. Res*., 225: 27-32.

Laraqui Hossini, C.H., Laraqui Hossini, O., Rahhali, A.E., Tripodi, D., Caubet, A. and Belamallem, I. 2002. Symptores respiratoires et troubles ventilatoires chez les travailleurs dans une cimenterie au Maroc [Respiratory symptoms and respiratory disorders among a group of cement workers in Morocco]. *Rev Mal Respir.,* 19:183–189.

Norppa,H., Luomahaara,S., Heikanen, H., Roth, S., Sorsa, M., Renzi L. and Lindholm, C. 1993.Micronucleus assay in lymphocytes as a tool to biomonitor human exposure to aneuploidogens and clastogens . *Environ Mol Mutagen*., 101: 139-143.

Perry, P. and Wolff, S. 1974. New giemsa method for the differential staining of sister chromatids. *Nature*. 251:156-158.

Rafnsson, V., Gunnarsdottir, H. and Kiilunen M.1997. Risk of lung cancer among masons in Iceland. *Occup. Environ. Med*., 54: 184-188.

Rahman, Q., Prasad, R., Das, M., Pandey, U.S., Ashquin, M. and Lohani, M.1996. Follow up study in an asbestos cement factory with special emphasis on preventive and diagnostic measures project., *Industrial Toxicology research Centre, India, Annual Report*.

Ramakrishnan, V., Gowtham kumar,S. and govindaraju,S. 2011. Cytogenetic analysis of micronuclei, sister chromatid exchange and chromosomal aberrations in pan masala chewers. *International Journal of Pharma and Bio Sciences*., 2(3):122- 134.

Rom, W.N., Livingston, G.K., Casey, K.R., Wood, S.D., Egger, M.J, Chiu, G.L.and Jerominski, L. 1983. Sister chromatid exchange frequency in asbestos workers., *J Natl Cancer Inst*, 70: 45-48.

Short, S. and Petsonk, E. L.1996. Non-fibrous inorganic dusts. In: Philip Harber, Marc B Schenker and John R Balmes. Occupational and environmental respiratory disease. *London. Mosby.*, 356.

Smailyte, G., Kurtinaitis, J.and Andersen, A. 2004. Mortality and cancer incidence among Lithuanian cement producing workers. *Occup Environ Med.,* 61(6): 529-534.

Srb, V.L., Kubzova, E. and Musil, M.1984. Testing genotoxic activity in exposure to asbestos I. Cytogenetic examination of lymphocytes of human peripheral blood. *Proc Leuk*., 36: 175-8.

Tomanin, R., Ballarin, C., Nardini, B., Mastrangelo, G. and F Sarto.1991. Influence of smoking habit on the frequency of micronuclei in human lymphocytes by the cytokinesis block mehod. *Mutagenesis*., 6: 123-126.

Trosic, I., Horvat, D., Stilinovic, L., Pisl, Z.1988. Cytotoxic, haemolytic and mutagenic issue caused by chrysotile asbestos in vitro. In: Mossman BT, Begin RO, eds. Effect of mineral dusts on cells. *New York: Springer-Verlag.*, 423-37.

Trosic, I., Brumen, V. and Horvat, D. 1997. In vitro assessment of asbestos fibers genotoxicity. Zentralbl Hyg Umweltmed, 558-567.

Valerio, F., De Ferrari, M., Ottaggio, L., Repetto, E. and Santi, L.1983. Chromosomal aberrations induced by chrysotile and crocidolite in human lymphocytes in vitro. *Mutat Res*., 122:397-402.

Yager, J.W. M., Sorsa, S. Selvin., Bartsch, H. and Hemminki, I.K., 1988. Micronuclei in cytokinesis-blocked lymphocytes as an index of occupational exposure to alkylating cytostatic drugsMethods for Detecting DNA Damaging Agents in Humans: Applications in Cancer Epidemiology and Prevention. *IARC, Lyon*., 89: 213–216.

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