

## Risk of Chromosomal Aberrations associated with Residential Proximity to an Industrial Area

**Sridevi Tirumalaraju<sup>1</sup>, Chandana Lakkaraju<sup>3</sup>, Dhanesh Mandam kulathil<sup>1</sup>, Vanitha Baluka<sup>2</sup>, Chiliveri .Prashanth<sup>2</sup> & U. Indira Priyadarshini<sup>2</sup>, P.P.Reddy<sup>1,2</sup>.**

<sup>1</sup>School of Biotechnology, Mahatma Gandhi National Institute of Research and Social Action;

<sup>2</sup>Department of Genetics and Environmental Toxicology, BhagawanMahavir Medical Research Centre

<sup>3</sup>Institute of Genetics and Hospital for Genetic Diseases, Hyderabad.

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### ABSTRACT

*A study on the cytogenetic damage in residents living in and around the Patancheru and Bollaram Industrial area, Hyderabad, Telangana state was carried out using chromosomal aberrations as parameter. For comparison, people who were living in the non-industrial area were included (control group). The subjects were clinically examined and information on demographic characteristics such as age, BMI, habits and duration of stay was collected. Blood samples from the residents of industrial area and non-industrial area were collected and analysed for chromosomal aberrations. The results showed a high frequency of chromosomal aberrations in the peripheral lymphocytes of the residents of industrial area (8.07%) and compared to the frequency of the chromosomal aberrations observed in the residents of the non-industrial area (2.61%). The results further showed a gradual increase with duration of stay in the residents of the industrial area. The statistical analysis of the results showed that the increase in the frequency of chromosomal aberrations in residents of the industrial area was significant ( $P$  value  $< 0.01$  &  $< 0.05$ ) compared to the control subjects. The cytogenetic effects in the residents might be due to exposure to the industrial pollutants in this area. The present study suggests that chromosomal aberration analysis may be considered as a suitable biomarker to predict the cancer risk in the populations exposed to industrial pollution.*

**Keywords:** Industrial Pollution, Chromosomal aberrations, lymphocytes, blood

### 1. INTRODUCTION

Population monitoring for health effects of environmental pollution is a critical problem especially in the case of mixed, low level chemical exposures. Studies have also reported chromosomal abnormalities in offspring associated with a maternal residence near waste sites (Brender et al., 2008), very few studies have examined the health effects in people living near industrial facilities (Hans Orru et al., 2018, Adil Alwahaibi and Ariana Zeka, 2016, Prashanth et al., 2017). Since it is not possible to control the background exposure conditions, it is difficult to identify the pollutants responsible for the observed effects in residents.

There have also been limited studies of the mutagenic effects on non-occupational exposure to individual chemicals (Infante, 1976). Mixed exposure studies have been conducted mainly in populations residing in the vicinity of old chemical dumps (Vrijheid, 2000, Heath, 1982). Many of the animal model (Iji and Adegun, 2014) and human population studies (Leelavathi and Mathivanan, 2015) showed relationship between exposure to industrial pollution and chromosomal aberrations. The epidemiologic studies showed increased risks of chromosomal aberrations in children of women lived near hazardous waste sites (Dolk et al., 1998; Sandra et al., 1992; Lisa A et al., 1997).

In several animal models including wild rats (Eckl and Riegler, 1997, Chibuisi Alimba et al., 2009) and mice (Yahya et al., 2017) pollutants at hazardous waste sites and in solid waste leachates are known to induce chromosomal aberrations and DNA damage.

Epidemiological studies were carried out all over the world established ill health effects (Subodh Kumar Rastogi et al., 2008) and cytogenetic damage (Anusha Pawar et al., 2016, Srinivas Chinde et al., 2014, Solange Costa et al., 2015) in the industrial workers. Thus it is highly pertinent to understand the health problems and cytogenetic effects in people living in and nearby industrial areas.

Chemical exposure to both women and men may cause chromosomal abnormalities or gene mutations in the offspring and also disturb the development of the fetus through heritable changes (epigenetic mechanisms) in egg or semen cells (Marja-Liisa Lindbohm and Markku Sallmén, 2017).

### 2. NEED FOR THE STUDY ON PEOPLE OF PATANCHERU AND BOLLARUM INDUSTRIAL AREAS

The present study was conducted in the residents of Patancheru-Bollaram industrial area to evaluate the risk of cytogenetic damage.

The Patancheru-Bollarum industrial Estate was established in the year 1975 in Medak district of Telangana state and is about 30 kms from Hyderabad. It is one of the most industrially polluted areas. Patancheru-Bollarum cluster is one of the most important industrial areas having an enormous number of industries manufacturing pharmaceutical drugs, paints, pesticides, steel, plastics, textiles, leather, rubber, bulk drug manufacturing units, etc., earning the State the nickname 'bulk drug capital' of the country. These industries release a lot of effluents, which are toxic and contaminate the air, soil and water. The impact is such that a pungent and acrid odour is released in this area. This region has traditionally relied on ground water for sustenance and is supported by seasonal streams like the NakkaVagu and IskaVagu. Recent reports suggest that both groundwater of the region and the surface water in NakkaVagu are heavily polluted due to domestic sewage, industrial effluents and also due to partially treated effluents from the common effluent treatment plant (CETP) of this region. As per the study of CPCB, the Patancheru –Bollaram industrial area was considered as a critically polluted area (ACTION PLAN-PTN-BLM., 2010). Very few studies were carried out on the health hazards in people living in this industrial area. Thus in the present study an attempt is made to understand the cytogenetic damage in the residents of this area.

### 3. METHODOLOGY

#### Study population

195 people living in and around Patancheru–Bollarum industrial areas were enrolled for the study (exposed group). For comparison 199 people belonging to the same age group and socioeconomic status and who were not occupationally exposed to chemical compounds and living away from the industrial area were selected (control group). This study was approved by the institutional ethics committee.

All participants were informed about the objectives of the investigation and written consent was obtained from each person. Personal information on the age, weight, height, duration of stay, habits (smoking & nonsmoking) were collected from the subjects from both the groups and entered in a questionnaire specially proposed for this purpose.

The following inclusion and exclusion criteria were followed for the selection of subjects .

**Inclusion:** 1. Residents with a minimum stay of 1 year

2. Residents not occupationally exposed to physical or biological agents

**Exclusion:** 1. Residents with less than one year stay

2. Industrial workers

3. Couples moved to the industrial area after conceiving elsewhere.

#### Analysis of chromosomal aberrations in peripheral blood of the industrial residents

Cytogenetic damage was assessed by analysing the chromosomal aberrations in the residents of both industrial area and nonindustrial areas.

3 ml blood sample was drawn from each subject from the residents of both the industrial and non-industrial areas and cultures were setup following the method of Moorhead et al., (1960). 0.5 ml blood from each individual was added to 5ml medium and each sample was maintained in duplicate. A control batch of cultures was always set up simultaneously for each batch of the study group and all the cultures were incubated at 37°C for 72 hours. 2 hours prior to the termination at the 70<sup>th</sup> hour colchicine (0.3 µl/ml) was added to all the cultures to inhibit the spindle formation. At the end of 72 hours each culture was decanted into 15 ml centrifuge tube and centrifuged at 1000 rpm for 10 minutes. The supernatant was discarded. 5ml of pre-warmed hypotonic solution (0.75 M KCl) was added to the pellet and the cells were incubated at 37°C for 20 minutes. After the hypotonic treatment, the cell suspension was centrifuged at 1000 rpm for 10 minutes. The supernatant was removed and to the pellet the pre-chilled fixative (3:1 methanol and acetic acid) was added carefully along the walls, to avoid clumping of the cells. The cells were allowed to stand for 10 min. After 10 minutes the cell suspension was centrifuged at 1000 rpm for 10 min. The supernatant was removed and fresh fixative was added to the pellet. This process was repeated for 3 to 4 times and the final cell suspension was made in 1ml of fresh fixative.

#### Preparation of the slides.

The micro slides were cleaned and kept in chilled distilled water before use. Two or three drops of cell suspension were dropped on each micro slide using a Pasteur pipette. The slides were air dried. All the slides were coded and stained in 2% Giemsa (2 ml of Giemsa, 2ml of Sorenson's buffer and 46 ml of distilled water) for 5 minutes. After staining, the slides were rinsed in distilled water and dried. The slides were then observed under the microscope.

### Scoring of chromosomal aberrations

For each individual 100 well spread metaphases were screened and scored for structural (gaps, breaks, dicentric) and numerical aberrations (polyploids). However, gaps and polyploids were not included in the total number of aberrations. Well spread metaphases were micro photographed.

### Statistical analysis

The differences in the frequencies of chromosomal aberrations and the demographic data between the residents of the industrial area and non-industrial area were analysed for significance using chi-square value and t-test.

## 4. RESULTS

The present study was conducted to evaluate cytogenetic effects in 195 subjects from the Patancheru and Bollaram industrial area and 199 subjects from non-industrial area (control group) and the results are as follows.

The demographic characteristics of the study population are represented in **Table 1**.

The mean age of the residents of the industrial area was 33.18 yrs. as against 36 yrs. of the residents of the non-industrial area. Nearly 51% of the residents of the industrial area were obese having BMI  $\geq 25$  Kg/m<sup>2</sup> while only 43.21 % were obese in control group. The results also show a higher prevalence of underweights (4.61%) in industrial residents than in people who were living in non-industrial area (3.01%). The Mean weight of industrial residents was 65 kg and that of control subjects was 63 kg. 43.58% of industrial residents had a normal weight and 53.26% of control subjects have normal weights. 94.87 % were non-vegetarians among study subjects as against 85.42% in controls. 69.23% literates and 23.07% smokers were observed among the study subjects and 83.92% literates, and 7% smokers were observed among the control group.

**Table - 1. Demographic data of the Residents of industrial and non-industrial areas**

S.No.	Parameter	Industrial residents	Non industrial residents	P-Value
1	Age (Years)			
	Mean $\pm$ SD	33.18 $\pm$ 10.59	36.0 $\pm$ 13.29	0.020*
2	Height (cm)			
	Mean $\pm$ SD	158.29 $\pm$ 8.00	160.09 $\pm$ 7.00	0.017*
3	Weight (Kg)			
	Mean $\pm$ SD	65.0 $\pm$ 12.00	63+10.39	0.008**
4	BMI (Kg/m <sup>2</sup> )			
	Under weight (<18.5)	9 (4.61%)	6(3.01)	0.14
	Normal (18.5-24.9)	85 (43.58%)	106(53.26)	
	Obese (>25)	100 (51.28%)	86(43.21)	
5	Veg	13 (6.66 %)	29(14.57)	0.009**
	Non-Veg	185(94.87%)	170(85.42)	
6	Literates	135 (69.23%)	167(83.92)	0.000**
	Illiterates	60(30.76%)	32(16.08)	
7	Duration of Exposure (Years)			
	Range	0.9 -30 years		
8	Smokers	45(23.07)	14(7)	0.000**
	Non-smokers	150(76.92)	185(93)	

\*p<0.05 Significant

\*\*p<0.01 highly significant

The results on the incidence of chromosomal aberrations in the residents living in the industrial area are presented in Tables 2. An increased incidence in the frequency of chromosomal aberration was observed in the industrial residents when compared to the control group. While the frequency of aberrations

was 2.61% in the residents of non-industrial area it has increased to 8.07% in the residents of the industrial area. The results in both the groups were categorized as smokers and nonsmokers. The frequency of chromosomal aberrations was assessed to understand the influence of smoking. The results showed an increased percentage of chromosomal aberrations (8.85) among the smokers of industrial residents as against the frequency (3.83) observed among the smokers from non-industrial area (Table-2).

Table-2 Frequency of Chromosomal aberrations in the residents of the Patancheru- Bollaram Industrial area

Group	No. of Samples	No. of Metaphases screened	Chromosomal aberrations				Total no. of Aberrations
			Gaps	Breaks	Dicentrics	Polyploids	
1. Residents of non-industrial area (Controls group)	199	19900	182(0.91)	339(1.70)	182(0.91)	122(0.61)	521(2.61)
Smokers	12	1200	17(1.41)	25(2.08)	21(1.75)	16(1.33)	46(3.83)
Non Smokers	187	18700	154(0.82)	277(1.48)	144(0.77)	101(0.54)	421(2.251)
2. Residents of industrial area (Exposed group)	195	19500	524(2.68)	987(5.06)	587(3.01)	384(1.97)	1574(8.07)
Smokers	33	3300	100(3.03)	148(5.48)	91(3.37)	64(2.37)	239(8.85)
Non-Smokers	168	16800	430(2.55)	849(5.05)	505(3.00)	335(1.99)	1354(8.05)
3. P-value			0.006*	0.003 *	0.414	0.534	0.004 *

\*p<0.05 Significant

\*\*p<0.01 highly significant

Values in the parenthesis are percentages.

Table 3. Frequency of chromosomal aberrations in the duration of residence in the Patancheru - Bollaram Industrial area.

Group	No. of Samples	No. of Metaphases	Chromosomal aberrations				Total no. of Aberrations
			Gaps	Breaks	Dicentrics	Polyploids	
1. Residents of the non-industrial area(controls)	199	19900	182 (0.91)	339(1.70)	182 (0.91)	122 (0.61)	521(2.61)
2. Residents of Industrial Area (by duration of residence)	1-5 yrs.	92	137 (1.48)	259 (2.87)	168 (1.82)	128 (1.39)	427 (4.65)
	6-10 yrs.	54	162* (3.00)	323** (5.98)	181** (3.35)	114* (2.10)	504** (9.33)
	11-15 yrs.	21	78** (3.71)	158** (7.52)	85 (4.04)	52** (2.48)	243** (11.57)
	>15 yrs.	28	141** (5.03)	247** (8.82)	153 (5.46)	90** (3.20)	400** (14.28)

\*p<0.05 Significant

\*\*p<0.01 highly significant

Values in the parenthesis are percentages.

The effect of duration of residence (stay) on the chromosomal aberrations in the residents of the Patancheru- Bollaram industrial area was studied and the results are presented in Table 3.

The frequency of chromosomal aberrations showed a gradual increase in the residents of the industrial area with the increase in the duration of residence. While the frequency of chromosomal aberrations was 4.65 % in the residents with 1- 5 years of stay, it has increased to 9.33% in the residents with 6-10 years. Further increase in the incidence of chromosomal aberrations was seen in the residents with 11-15 years residence (11.57%) and more than 15 years (14.28%) of residence.

The statistical analysis of the results using chi square showed that the increase in the chromosomal aberrations in the residents with increased years of their stay was significant. Suggesting dosage effect.

#### 4. DISCUSSION

Acquisition of scientific knowledge and technologies, their application in industrialization, rapid urbanization and increasing needs of man has resulted in pollution of the environment and affected all parts of the biosphere making survival of living organisms precarious. Industrial pollution has become more significant and a global reality. Hence there is a high risk in the population living in villages nearby industrial areas for adverse health effects due to contamination of air, water and soil with industrial pollutants and nearby villages are contaminated with high levels of heavy metals. Extensive studies have clearly showed the health problems (Heron and Pickering, 2003) in workers of pharmaceutical industry, cytogenetic effects in cement (Divvyapriya and suja, 2012, Calistus Jude, 2002) and leather (Subodh Kumar Rastogi et al., 2008; Anusha Pawar et al., 2016), agricultural (Fatih Taspinar and Ismail toroz, 2017) industries. However studies on the adverse effects in people living in and around industrial areas are very scanty. Studies carried out on people living near to industrial dumps and in children playing close to industrial waste indicated certain health problems (Ihedioha et al., 2017).

In this context a study was taken up to know the possible detrimental effects in the residents of Patancheru and Bollaram area where many industries are located. The present study carried out in the residents of the Patancheru and Bollaram industrial area showed a significant increase in the chromosomal aberrations, thus suggesting cytogenetic damage compared to the controls.

In our study we observed significant increase of chromosomal aberrations in the residents of this area thus induced effects have an attributions. Smoking also considered as an important factor in cytogenetic studies on occupational and industrial exposures, a significant increase in chromosomal aberrations were observed in smokers when compared to non-smokers. Our results confirm previous reports (Vladimir et al., 2015, Anusha C Pawar et al., 2016). Thus data generated clearly provided evidence for the cytogenetic damage in the residents of the industrial area due to undue exposure to industrial pollution at Patancheru and Bollaram industrial area where a number of pharmaceutical, rubber, iron, bulk drug, steel, leather, cement and paint industries are located. The effects might be attributed to the indirect effect of exposure to pollutants liberated from the industries.

Chromosomal aberrations have been reported in human population groups with occupational exposure to specific chemicals, such as Lead (Urmisha Das and Madhusnata De., 2013), vinyl chloride (Ducatman et al., 1975, Czeizel et al., 1977) benzene (Watanabe et al., 1980) pesticides (Brian et al (2009), nickel (MahaFahmy et al., 2014, Waksvik et al., 1982) and styrene (Watanabe et al., 1983).

The industrial management has to take proper measures to control the release of industrial gases and wastes in to the environment. The results of the above study showed adverse effect on chromosomes. Further studies are needed to understand the consequences of exposure of children living in this area.

## 5. CONCLUSION

The results clearly showed induced chromosomal aberrations in the residents of the industrial area compared to matched controls and the same can be attributed to environmental pollution with industrial pollutants. The study also demonstrates that factors such as cigarette smoking and duration of residence are associated with the increased frequencies of chromosomal aberrations in the industrial residents. Thus the study suggested that long term exposure to the industrial pollution and stay in the industrial area might cause adverse genetic consequences in the population resulting the health of future generation.

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