UDC 575.224. 23:613.6:622

DOI: 10.15372/KhUR20160414

Genotoxic Effects of Coal Dust on Kuzbass Workers

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Abstract

The DNA damage in peripheral blood lymphocytes in employees of Kuzbass coal enterprises and men, non-exposed to the coal dust, was studied. In total, 501 donors were included in our research (137 coal miners, the average length of service is 28 years and 104 thermal power plant workers, the average length of service is 25 years). The average concentration of coal dust in miners' workplace reached to 190 mg/m^3, the content in power plant workers varied from 11 to 23 mg/m^3. As a control group, 260 non-exposed men of similar age living in the same area were studied. Their blood samples were obtained from the ulnar vein. Cell cultivation, slides preparation and chromosomal aberrations assessment were performed in a similar way for all groups. It was shown, that the frequency cells with chromosomal aberrations in coal miners and thermal power plant workers was significantly increased compared to the control. Increasing the frequency of chromosomal breaks (chromosome as well as chromatid type) and chromosome exchanges was registered. The results obtained are the evidence of the expressed genotoxic exposure in the workers occupationally associated with the coal dust.

Key words: coal dust, chromosomal aberrations, coal miners, thermal power plants

INTRODUCTION

The coal dust is a mixture of various organic and inorganic chemical agents that can induce the DNA damage. This mixture consists of quartz, metals, hydrocarbons transforming into cancerogenic polycyclic aromatic hydrocarbons (PAHs) under high temperatures. Moreover, coal contains native radioactive agents from uranic, aktinoutranic and thorium series. Combined exposure to these radioactive and chemical factors can induce various types of DNA damage, formation of adducts, single and double DNA strand breaks, structural and quantitative chromosomal abnormalities.

The experiments showed that exposure to coal dust could induce molecular, cellular and histopathological alterations in exposed laboratory mice [1]. The investigation of animals caught in the mining regions indicated cytotoxic and genotoxic effects of the exposure to coal dust. The increased degree of the DNA damage was discovered in wild rodents and *Iguana iguana* from the coal mining areas of Brazil and Colombia [2, 3].

Investigation the results of underground and open-cast miners performed in various countries (Turkey, Brazil, Colombia, the Netherlands, Peru and India) testify a high level of chromosomal alterations (CAs) in coal miners. Such alterations are accumulated in various cells (buccal epithelium and blood lymphocytes) and can be registered using such tests as chromosomal aberrations and sister chromatid exchanges (SCE) assessment, micronucleus assay and DNA comet assay. This fact evidences of the multiple character of the genotoxic agents exposing coal miners [4–9].

In the Kemerovo Region a positive correlation between air pollution from industrial wastes and lung cancer incidences were registered [10].

According to these data, studying the level of the DNA damage in the residents of the Kemerovo Region working in conditions of high concentrations of the coal dust is important.

MATERIALS AND METHODS

A group of 137 coal miners working at the coal mines of the south of Kemerovo Region (drift miners, diggers, steigers; the average length of service is 28 years) was examined. Average concentration of coal dust in miners' workplace reached to 190 mg/m^3. Also 104 thermal power plant workers working at the Novo-Kemerovskaya Thermal Power Plant (the average length of service is 25 years) were included in the exposed group. Average concentration of coal dust in power plant workers' workplace varied from 11 to 23 mg/m^3. As a control group 260 non-exposed men of similar age (50 years) living in the same area were studied.

Cytogenetic investigations were performed using the routine test for the assessment of CAs [11] with certain modifications. The whole blood obtained from the ulnar vein was cultured. Volumes of 0.5 mL blood, 0.1 mL phytohemagglutinin (PanEco, Moscow, Russia), 6 mL RPMI-1640 (PanEco, Moscow, Russian Federation) and 1.5 mL embryonic veal serum (PanEco, Moscow, Russia) were added to a culture flask. The duration of the cultivation was 48 h. Then, colchicine (PanEco, Moscow, Russia) was added to the culture at a final concentration of 0.5 μg/mL, and the flasks were placed in an incubator for 2 h. At the end of the cultivation cycle, the preparations were centrifuged for 10 min at 1000 rpm, the supernatant was removed, and the pellet was resuspended. The pellets were placed in a hypotonic solution of 0.55 % KCI for 10–15 min at 37 °C. The fixation of the material was performed in a cooled fresh Carnoy fixative (methanol and acetic acid in the ratio of 3 : 1). The cell suspension was pipetted onto clean, cooled slides moistened with water. The preparations were encoded and stained with a 2 % Giemsa solution. Counting of the aberrations was performed using light microscopy at 1000 magnification (oil immersion) without karyotyping. The selection of metaphases included in the analy-

TABLE 1

Quantitative characteristics of chromosomal alterations in the studied groups

Characteristics	Coal miners n = 137		Thermal power plant workers n = 104		Non-exposed control n = 260	
	Me	min–max	Me	min–max	Me	min–max
Aberrant metaphases (%)	5.5*#	1.5–11.5	5.5*	0–11.5	1.0	0–5.0
Aberration/100 cells	4.0*#	1.5–11.5	6.0*	0–11.5	1.0	0–5.5
Single fragments (%)	4.0*	0–8.5	4.0*	0–8.5	0.6	0–5.5
Chromatid interchanges (%)	0**#	0–1.0	0	0–1.0	1.0	0–1.0
Chromatid-type aberrations (%)	4.0*	0.5–8.5	4.0*	0.5–8.5	0.5	0–5.5
Acentric fragments (%)	1.0*#	0–3.5	1.0	0–3.5	0	0–2.0
Dicentrics + fragments (%)	0***#	0–1.0	0***	0–1.0	0	0–0.7
Dicentrics without fragments (%)	0***	0–1.5	0**	0–1.5	0	0–0.5
Ring chromosomes (%)	0*#	0–0.5	0	0–0.5	0	0–1.0
Atypical monocentrics (%)	0*#	0–2.0	0	0–2.0	0	0–0.5
Chromosome-type aberrations (%)	0*#	0–2.0	0**	0–3.0	0	0–2.0

Note. Me – median.

*$p < 0.0001$.

**$p < 0.01$.

***$p < 0.05$ – significant differences in comparison with non-exposed control; # ignificant differences in comparison with thermal power plant workers.

sis and the criteria for cytogenetic abnormalities conformed to the generally accepted recommendations [12, 13].

RESULTS AND DISCUSSION

We discovered that the frequency of blood lymphocytes with chromosomal damage in trained coal miners and thermal power plant workers was significant increased compared to non-exposed control (Table 1). The increase of frequency of the chromosomal breaks (chromosome and chromatid types) and the exchange type restructurings were reported. This fact demonstrates expressed mutagenic exposure to occupational factors to workers' genome. The most increased indicators were registered in coal miners contacted with the highest concentration of coal dust.

It is known that dust particles can accumulate molecules of benzo(a)pyrene, as well as α-particles of radon and transfer them for long distances, inpour into human organism during inhalation. In lung tissues, dust particles can induce of generation of reactive oxygen species (ROS) that initiate the breakage of DNA bases and desoxyribose, contribute to the formation of new covalent bonds (linkages) and other changing.

The average values of CAs obtained in our research in coal miners are similar to ones reported for workers from Turkey [4, 14].

The highest frequencies of CAs were described in drift miners ((5.76±0.51) %), drivers of extracted coal transport ((5.20±0.76) %), electricians ((5.26±1.15) %), stope miners ((4.29±0.525) %). These professions are connected with long underground work (during the entire shift). The average concentration of coal dust in miners' workplace reached to 180 mg/m^3 that was 45 times higher than the maximum permissible concentration.

Among thermal power plant workers, the highest frequency of cells with chromosomal alterations were registered in employees of the fuel and transport workshop ((4.19±0.19) % *vs.* (3.02±0.3) % – in employees of the repair workshops). Thus, the frequency of CAs in engine drivers was (4.55±0.24) %, whereas in mechanics – (3.70±0.32) % (p = 0.004).

CONCLUSION

The results obtained are the evidence of the expressed genotoxic exposure to occupational factors in workers contacting with coal dust and show the need of monitoring mutagenic and cancerogenic effects in coal industry workers.

Acknowledgement

This research was supported by the RCF grant No. 16-15-00034.

REFERENCES

1 Caballero-Gallardo K., Olivero-Verbel J., *Toxicol. Appl. Pharmacol.*, 294 (2016) 11.
DOI: 10.1016/j.taap.2016.01.009
2 León G., Pérez L. E., Linares J. C., Hartmann A., Quintana M., *Mutat. Res.*, 630 (2007) 42.
3 Cabarcas-Montalvo M., Olivero-Verbel J., Corrales-Aldana H., *Sci. Total Environ.*, 416 (2012) 208.
4 Donbak L., Rencuzogullar E., Yavuz A., Topaktas M., *Genetic Toxicol. and Environ. Mutagen.*, 588 (2005) 82.
5 Santa Maria S. R., Arana M., Ramirez O., *Genetics and Molecular Biology*, 30, 4 (2007) 1135.
6 Kumar A., Yadav A., Giri S. K., Dev K., Gautam S. K., Gupta R. and Aggarwal N., *Environ. Toxicol. Pharmacol.*, 32 (2011) 128.
7 Rohr P., Kvitko K., Silva F. R., Menezes A. P., Porto C., Sarmento M., *Mutat. Res.*, 758 (2013) 23.
8 Kvitko K., Bandinelli E., Henriques J. A., Heuser V. D., Rohr P., da Silva F. R., *Genetics and Molecular Biology*, 35, 4 (2012) 1060.
9 León-Mejía G., Quintana M., Debastiani R., Dias J., Espitia-Pérez L., *Ecotoxicol. Environ. Saf.*, 107 (2014) 133.
10 Mun S. A., Glushkov A. N., *Gig. Sanit.*, 2 (2014) 37.
11 Hungerford P. A., *Stain Technol.*, 40 (1965) 333.
12 Brogger A., Hagmar L., Hansteen I. L., *Cancer Genet. Cytogenet.*, 45 (1990) 85.
13 Savage J. R., *J. Med. Genet.*, 13 (1976) 103.
14 Celic M., Donbak L., Unal F., Yuzbasioglu D., Aksoy H., Yilmaz S., *Mutation Res.*, 627 (2007)158.

UDC 575.224. 23:613.6:622

DOI: 10.15372/KhUR20160414

Генотоксические эффекты воздействия угольной пыли на рабочих Кузбасса

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Аннотация

Изучены повреждения ДНК в лимфоцитах периферической крови у рабочих угольных предприятий Кузбасса и у мужчин, профессионально не контактирующих с угольной пылью. В исследование включен 501 человек (в том числе 137 шахтеров со средним стажем работы под землей 28 лет, 104 рабочих теплоэлектростанции, работающей на угле, со средним стажем 25 лет). Средняя концентрация пыли на рабочем месте шахтеров достигала 190 мг/м^3, а у рабочих теплоэлектростанции варьировала от 11 до 23 мг/м^3. Для контроля обследованы 260 не экспонированных мужчин близкого возраста, проживающих в той же местности. Образцы крови забирались из локтевой вены. Культивирование клеток, подготовка препаратов, оценка хромосомных аберраций проводились сходным образом во всех группах. Установлено, что частота клеток с хромосомными аберрациями у шахтеров и рабочих теплоэлектростанций статистически значимо превышает контроль. Наблюдалось повышение частоты хромосомных разрывов (хромосомного и хроматидного типов) и хромосомных обменов. Полученные результаты свидетельствуют о выраженной генотоксической нагрузке у рабочих, контактирующих с угольной пылью.

Ключевые слова: угольная пыль, хромосомные аберрации, шахтеры, теплоэлектростанции