Cytogenic Aspect of Distant Outcomes in Workers Occupationally Contacting with Dioxin-Containing Products

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1. Introduction

Indications of dioxin genotoxic danger takes an important place among distant outcomes of dioxin exposure. Mutagenic effect can show itself not only in the present population, but also in the following generations by transference of mutation to offsprings. Valuable information about cytogenetic effects can be obtained in the early periods by evaluation of mutability in somatic cells and in lymphocytes of the peripheral blood.

2. Materials and Methods

2.1. Study Group

The main study group consisted of 15 workers (2 women and 13 men) aged from 30 to 60 working at Chimprom Production Association first in contact with 2,4,5-T butyl ether then with 2,4-dichlorphenol. The control group included 37 healthy donors of the same age without any harmful occupational influence.

2.2. Analytical Methods

Venous blood was cultured by the widely used Hungerford method (1). Phytohemagglutininum ("Panax", Moscow) deluted to 5 ml by sterile water was used as a stimulant of lymphocyte proliferative activity. 0,2 ml of this mixture was added into a cultured flask. Mixtures of the same series were used for workers' and donors' blood samples culture.

For the chromosome aberration analysis the blood with nutrient medium was kept up at 37° for 48 hours since the phytohemagglutininum introduction, as the majority of cells was in the first mitosis during that time. Colchicinum with the final concentration of 0,5 mgk/ml was introduced into the culture 2 hours before fixation. After centrifugation and draining the cells were put to hypertonic treatment by 0,55% KCL solution at 37° for 10 minutes. After that the flasks were again centrifuged, drained and fixed by the cooled Kornua fixative (menthol /icy acetic acid in the proportion of 3:1), with the fixative being changed three times. Fully secured cellular suspension having

been obtained, chromatin body preparations were produced by dropping the cells onto clean, degreased and cooled microslides. Preparations were dried above the burner flame.

For chromosome aberration counting the preparations were dyed by Giemsa dye-staff water solution (0,1 ml of dye-stuff per 1 ml of distilled water) for 8-10 minutes. Chromatic aberration analysis was carried out by the standard light-microscopic preparation testing in 100-200 cells from each sample. Registration of chromatide and chromosome type aberration was carried out in accordance with WHO classification principles.

3. Results

Obtained data analysis showed that the mean value of chromatic aberration frequency exceeded the control value almost twice as much. However, the mean values of chromosome break frequency in both groups practically did not differ (table 1).

Table 1: Chimprom PA Workers' and Control Donors' Chromosome Aberration Frequency

Aberration name	Workers	Donors	P
Number of checked up people	15	37	
Number of analysed metaphases	1500	3700	
Medium frequency of metaphases with chromatic aberration	4,47± 2,79	2,9± 0,27	<0,05
Medium frequency of chromosome breaks	0,046± 0,028	0,04± 0,00	>0,05

Distribution of different type chromatic aberrations in the tested and control groups revealed that workers occupationally contacting with dioxin-containing products had both chromatic and chromosome aberrations (Table 2). Predominance of a single acentric fragments share in 57,4% and the true frequency increase of the chromosome aberrations respectively confirmed the participation of a chemical factor in the mutation origin, that influenced the G_1 and S phases of the cellular cycle. The chromosome aberrations were presented by pair fragments and had true distinction in 42,6% of people, single cases (2%) manifested chromosome changes.

Table 2: Distribution of Different Chromosome Aberration Types

Number of aberrations	Per 100 metaphases		% of overall number	
Aberration types	workers	controls	workers	controls
Overall number	4,53*	3,08	100	100
Chromatide aberrations:				
single acentric fragments	2,6±0,41*	1,55±0,2	57,4	50,5
chromatide exchanges	0,08	0,13	3,1	4,2
total	2,68	1,68	60,5	54,7
Chromosome aberrations				
pair fragments	1,93±0,35*	1,27±0,18	42,6	41,38
chromosome exchanges	0,06	0,12	2,0	3,9
total	1,99	1,39	44,6	45,3
N.B.: Differences are true	($p<0,05$)			

4. Conclusions

Thus, cytogenetic analysis results in the cohort study group permitted to establish chromosome aberration frequency increase in workers occupationally contacting with dioxin-containing products, that indicated a cytogenetic action on the subcellular level. Such an increased chromosome sensibility is an unfavourable prognostic sign as far as distant medical and biological outcomes are concerned, because the revealed mutation effect is evidence for the potential genetic activity. In support of this interpritation one can also consider the revealed by us tendency for somatic abortions in women with a past history of chloracne and sex disproportions of new-born children in this cohort group.

5. References

(1) Hungerford D.A. Leucocytes cultured from small inocula of whole blood and preparation of metaphase chromosomes by treatment with hypotonic KCL / Stain. Technol. -1965. Vol. 40. -P. 333-338.