CORRELATION BETWEEN LEVEL OF SISTER CHROMATID EXCHANGES AND CHROMOSOMAL ABERRATIONS INDUCED BY CHEMICAL MUTAGENS *IN VIVO*

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KEY WORDS: chromosomal aberrations; mutagens; chromatid exchange.

For many chemical substances sister chromatid exchanges (SCE) are a more sensitive indicator of mutagenic effects than chromosomal aberrations [6]. In most cases, however, this has been shown for the action of mutagens *in vitro*. Yet it is not clear to what extent increased sensitivity of recording of SCE compared with chromosomal aberrations still applies *in vivo*. Experiments *in vivo* allow mutagenic action not only of original substances, but also of their metabolites, to be studied and allowance to be made for the pharmacokinetics of the compounds also. This allows a more realistic determination of mutagenic action in animals and man.

In this investigation the levels of SCE and chromosomal aberrations induced by five mutagens *in vivo* were compared on the basis of a study of their dose dependence.

EXPERIMENTAL METHOD

Dose dependences of five compounds with different types and numbers of alkylating groups were studied: mitomycin C, a bifunctional antitumor antibiotic, E-39, a quinone derivative with two ethylenimine groups; thiophosphamide and dipin, derivatives of aminophosphoric acid with three and four ethylenimine groups respectively, and degranol, a derivative of di-β-chloroethylamide, with two chloroethyl groups. The compounds were injected intraperitoneally into CBA mice 24 h before sacrifice. For each concentration, when chromosomal aberrations were studied, three mice were used in two or three repetitions, whereas when SCE were studied, one or two concentrations were tested. SCE were detected by the experimental scheme described previously [8]. Bone marrow preparations were obtained by the method in [2, 7]. To analyze SCE the preparations were stained by the method in [5]. To assess the number of SCE per cell 25 metaphases were analyzed in each case. To analyze chromosomal aberrations the preparations were stained with azure-eosin, 100 metaphases were studied in each repetition, and the fraction of aberrant metaphases, the total number of breaks, the fraction of chromatid breaks, and the number of exchanges per 100 cells were counted.

EXPERIMENTAL RESULTS

Dose dependences of SCE and chromosomal aberrations for the compounds tested are given in Figs. 1 and 2. For all substances, during induction of chromosomal aberrations larger doses were used than for induction of SCE, for both *in vivo* and *in vitro* [5] SCE proved to be a more sensitive indicator of mutagenic action. The dose dependence for SCE *in vivo* for thiophosphamide and E-39 is satisfactorily described by a linear function ($r^2 = 0.98$ and $r^2 = 0.94$ respectively). Since for all the other substances tested *in vitro* a linear relationship also is observed [5], the connection between the number of SCE induced *in vivo* per cell (S) with dose is described by the equation $S = B_0 + B_1 \cdot C$, where B_0 and B_1 are coefficients, C the concentration (in mg/ml); thiswas determined as numerically equal to the weight of the substance in milligrams per gram body weight [1]. For that reason, the injected dose can be

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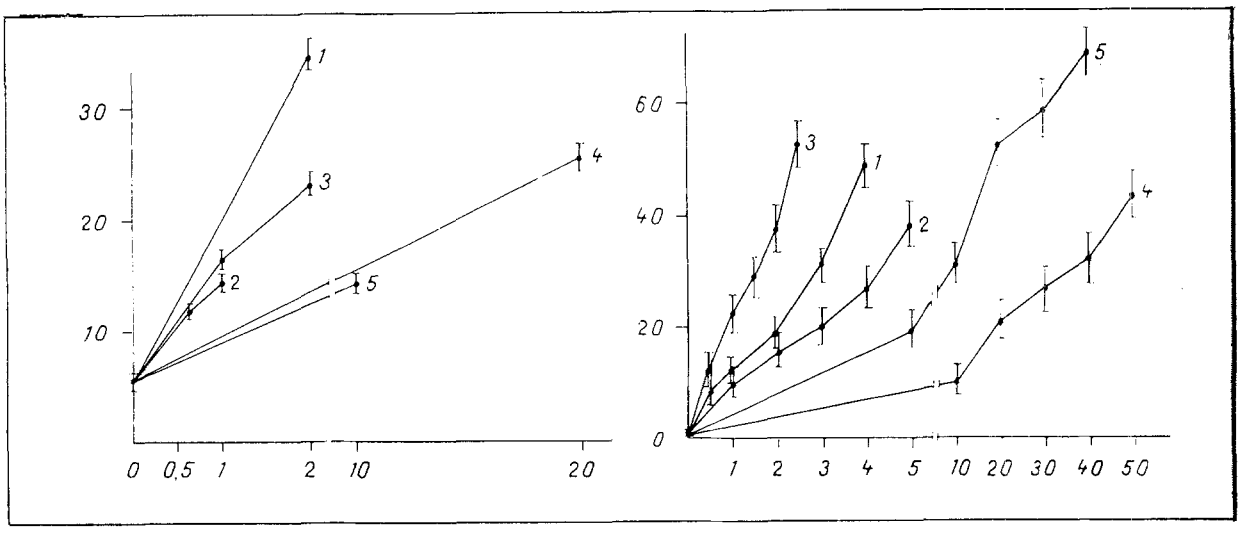

Fig. 1 Fig. 2

Fig. 1. Changes in number of SCE depending on concentration of substance. Abscissa, concentration (10^{-5} mole); ordinate, number of SCE per cell. 1) Mitomycin C, 2) E-39, 3) thiophosphamide, 4) degranol, 5) dipin.

Fig. 2. Dependence of fraction of aberrant metaphases on concentration of substance. Ordinate, fraction of aberrant metaphases (%). Remainder of legend as to Fig. 1.

TABLE 1. Comparative Effectiveness of Chemical Compounds in Inducing Chromosomal Aberrations and SCE

Substance	SCE, $B_1 \times 10^6$, mole^{-1}	Chromosomal aberrations $(K \pm m) \cdot 10^4$, mole^{-1}	B_1/K
Mitomycin C	4,87	10,2±0,89	48
E-39	2,95	5,11±0,75	58
Thiophosphamide	1,85	7,90±0,47	23,4
Degranol	0,37	0,68±0,06	55
Dipin	0,30	1,26±0,12	23,7

Legend. m) Standard error.

defined as concentration and expressed in moles. The change in the fraction of aberrant metaphases (ρ) depending on dose is described by the equation $\rho = (1 - e^{-(\alpha+KC)})^2$ where C is concentration, and α and K are coefficients. This equation satisfactorily describes the experimental data for all substances (level of correlation significant; $P > 0.05$). Values of the coefficients B_1 and K which determine the effectiveness of the substances for inducing SCE and chromosomal aberrations respectively are given in Table 1. The substances in the table are arranged in order of diminishing effectiveness of SCE formation. The order is different for chromosomal aberrations, although mitomycin C is the most effective as regards formation of chromosomal aberrations also. In the last column of the table, values are given for B_1/K, showing the relative effectiveness of SCE formation compared with chromosomal aberrations. It will be clear from Table 1 that effectiveness for SCE induction for all substances is 20-60 times higher than for induction of chromosomal aberrations. It must be pointed out that, according to their degree of effectiveness of SCE formation *in vivo*, the substances are arranged in the same order as for their action *in vitro* in human lymphocytes [5]. If, however, they are compared with respect to formation of chromosomal aberrations, the order of change of activity is mainly preserved, the exception being that thiophosphamide and degranol exchange places. This is evidently because different substances possess differential resistance when injected into the animal. In that case the duration of action on bone marrow cells will differ, and this in turn will cause the substances to act on different phases of the cell cycle. However, whereas in the case of SCE effectiveness is practically constant for all phases, except the last C_2-phase before fixation [3],

different phases of the cell cycle possess differential sensitivity to the formation of chromosomal aberrations [9].

SCE are thus both a more sensitive and a more stable indicator of the action of a mutagen *in vitro* and *in vivo* than chromosomal aberrations.

LITERATURE CITED

1. N. P. Bochkov, S. V. Stukalov, and A. N. Chebotarev, Byull. Éksp. Biol. Med., No. 3, 90 (1982).
2. A. M. Malashenko and N. I. Surkova, Genetika, No. 1, 71 (1974).
3. A. N. Chebotarev and T. G. Selezneva, Genetika, No. 10, 1941 (1979).
4. A. N. Chebotarev, T. G. Selezneva, and V. I. Platonova, Byull. Éksp. Biol. Med., No. 2, 242 (1978).
5. E. Gebhart, Hum. Genet., 58, 235 (1981).
6. S. B. Neal and G. S. Probst, Mutat. Res., 113, 33 (1983).
7. W. Vogel and T. Bauknecht, Nature, 260, 448 (1976).
8. V. S. Zhurkov and K. N. Yakovenko, Mutat. Res., 41, 107 (1976).

RADIOSENSITIVITY OF LYMPHOCYTE CHROMOSOMES IN DOWN'S SYNDOME

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Data on variability of individual radiosensitivity of human chromosomes are of considerable theoretical and practical interest. On the one hand, an increased frequency of radiation-induced chromosomal aberrations objectively reflects the defectiveness of the DNA repair system [7, 9], and it enables the efficacy of genetic control of this trait to be studied. On the other hand, even the comparatively small amount of information on changes in spontaneous and induced mutation of chromosomes in some hereditary human diseases is evidence that a tendency toward malignant tumors and to premature aging is causatively linked with DNA repair defects [4]. For example, in xeroderma pigmentosa, which is the most closely studied disease from the standpoint of repair defects, the likelihood of appearance of skin cancer is three orders of magnitude greater than the corresponding figure for a control population consisting of persons of the same age [10].

The discovery of a particular defect in the repair system of genetic material can thus be regarded as a criterion of belonging to an increased risk group with regard to these traits, and even now that is sufficient to justify the need for prophylactic measures and, in the near future, for special treatment also. Investigations carried out previously by the present writers [1, 2, 5] have shown that belonging to a high risk group can be determined sufficiently reliably by the use of lymphocytes cultured *in vitro*, by studying dose dependence on the yield of radiation-induced chromosomal aberrations over a wide range of doses.

The object of this investigation was an experimental analysis of dose—effect curves for the yield of chromosomal aberrations produced by the action of ^{60}Co γ-rays in lymphocytes from patients with various forms of Down's syndrome. The basis for the investigation

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