higher than after the action of this dose of PB alone. However, the increase was not statistically significant.

Injection of PB, an inducer of the MFO system, thus caused an increase in the level of chromosomal aberrations induced by CP in rat bone marrow cells. Definite correlation could not be found between the activity of the MFO system and the cytogenetic activity of CP. Against the background of marked induction of the MFO system (PB in a dose of 80 mg/kg) and of its very mild activating effect (PB in a dose of 2 mg/kg) the increase in the cytogenetic effect of CP was equal. To explain this fact it can be postulated that administration of low doses of PB, which do not bring about any appreciable induction of the MFO system, "converts" it from a state of relative stability into a state of readiness to react, against the background of which the rate of metabolism of the different substrates is modified.

These results are in agreement with observations showing an increase in the teratogenicity of CP in rats pretreated with PB, and a reduction in the antitumor activity of CP as a result of inhibition of the MFO system [6, 10]. However, these results were not confirmed by other investigations [4, 5]. The contradictions thus arising may be attributed to the pharmacokinetics of CP and to differences in the experimental conditions. All these considerations point to a need for further experimental studies of the facts described above, including a study of the kinetics of CP associated with the action of MFO modifiers in different doses.

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GENETIC DIFFERENCES IN SENSITIVITY OF MICE TO THE IMMUNODEPRESSIVE ACTION OF ALKYLATING AGENTS

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KEY WORDS: cyclophosphamide; thiophosphamide; sarcolysin; immunodepression; genotype.

Regulation of the immune response of the organism by means of chemotherapeutic agents is a problem of great practical and theoretical interest. An urgent aspect of this problem is the study of the genetic basis of the immunodepressive action of different agents, for the end result of intervention, namely inhibition of the immune response. However, insufficient research of this kind has been undertaken so far. Previously [1] the writers studied the sensitivity of mice of different genotypes to the immunodepressive action of cyclophosphamide (CP), an alkylating agent belonging to the bis-β-chloroethylamine group. It was shown by the use of a model of the primary humoral immune response during immunization with sheep's red blood cells (SRBC) that DBA/2 mice are highly sensitive to the immunodepressive action of CP whereas BALB/c mice are relatively resistant.

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TABLE 1. Sensitivity of Mice of Different Lines to Immunodepressive Action of CP (mean values)

Line of mice	Dose of CP, mg/kg	No. of group	Number of mice in group	Number of AFC in spleen, % of control	P
BALB/c	50	1	13	0,05 (0,03—0,06)	$P_{1-4}>0,05$
	25	2	13	7,9 (5,8—10,8)	$P_{1-7}>0,05$
	12,5	3	13	55,1 (50—60,7	$P_{2-5}<0,001$
	50	4	13	0,06 (0,05—0,09)	$P_{2-8}<0,001$
C3H/Sn	25	5	13	1,8 (1,3—2,5)	$P_{3-6}<0,05$
	12,5	6	14	39,5 (28,7—54,5)	$P_{3-9}<0,001$
	50	7	12	0,03 (0,02—0,05)	$P_{4-7}<0,01$
DBA/2	25	8	14	0,26 (0,18—0,38)	$P_{5-8}<0,001$
	12,5	9	14	13,1 (8,4—20,5)	$P_{6-9}<0,001$

Legend. Here and in Tables 2 and 3, confidence intervals shown in parentheses.

TABLE 2. Sensitivity of Mice of Different Lines to Immunodepressive Action of Thiotepa (mean values)

Line of mice	Dose of thiotepa, mg/kg	No. of group	Number of mice in group	Number of AFC in spleen, % of control	P
BALB/c	16	1	22	0,08 (0,05—0,12)	$P_{1-4}>0,05$
	8	2	24	11,7 (8,4—16,2)	$P_{1-7}>0,05$
	4	3	23	51,5 (45,5—58,3)	$P_{2-5}<0,001$
	16	4	16	0,15 (0,13—0,27)	$P_{2-8}<0,001$
C3H/Sn	8	5	22	3,5 (2,2—5,5)	$P_{3-6}>0,05$
	4	6	24	44,1 (32,2—60,3)	$P_{3-9}<0,05$
	16	7	11	0,08 (0,04—0,13)	$P_{4-7}>0,05$
DBA/2	8	8	12	2,1 (1—4,5)	$P_{5-8}>0,05$
	4	9	13	34 (23,5—49,2)	$P_{6-9}>0,05$

In the investigation described below an attempt was made to discover whether relationships of the same kind exist between mice of other lines. It was also interesting to determine whether interlinear differences exist in mice in sensitivity to the immunodepressive action of sarcolysin (SL), which also is a bis-β-chloroethylamine in its chemical structure, and thiophosphamide (thiotepa), an alkylating agent of the ethylenimine group. These agents, unlike CP [2], act directly on immuno-competent target cells without any preliminary metabolic activation *in vivo*.

EXPERIMENTAL METHOD

Male mice weighing 18-25 g and belonging to the following inbred lines were used: AKR/ISto, BALB/cISto, CC57BR/MvRap, C3H/SnSto, C57BL/6ISto, DBA/2 ISto. CP (the Soviet preparation cyclophosphan), thiotepa (synthesized at the S. Ordzhonikdze All-Union Pharmaceutical Chemical Research Institute), and SL (synthesized in the Chemical Technological Laboratory of

TABLE 3. Sensitivity of Mice of Different Lines to Immunodepressive Action of SL (mean values)

Line of mice	Dose of SL, mg/kg	No. of group	Number of mice in group	Number of AFC in spleen, % of control	P
BALB/c	8	1	11	2,6 (1,5—4,4)	$P_{1-4}<0,01$
BALB/c	4	2	12	30,8 (26,9—35,2)	$P_{1-7}<0,001$
	2	3	12	58,3 (51,2—66,5)	$P_{2-5}=0,01$
	8	4	12	0,66 (0,32—1,37)	$P_{2-8}<0,001$
C3H/Sn	4	5	13	13,8 (8,3—22,9)	$P_{3-6}>0,05$
	2	6	12	44,8 (33,7—59,4)	$P_{3-9}<0,001$
	8	7	12	0,05 (0,03—0,07)	$P_{4-7}<0,001$
DBA/2	4	8	13	1,5 (1,1—2,4)	$P_{5-8}>0,001$
	2	9	14	18,2 (14,2—23,3)	$P_{6-9}<0,001$

the All-Union Oncologic Scientific Center, Academy of Medical Sciences of the USSR) were used as immunodepressants. SRBC was used as the antigen. The mice were immunized intravenously with SRBC in a dose of 5×10^8 cells, after which the immunodepressant was injected intraperitoneally. The interval between injection of the antigen and immunodepressant was 24 h in the case of CP, whereas thiotepa and SL was injected immediately after immunization. The number of antibody-forming cells (AFC) in the spleen of the mice was determined by Jerne's method [3] 4 days after injection of the antigen. Animals not treated with immunodepressants served as the control. The results were expressed as percentages of the control. Statistical analysis of the results carried out by Student's t test. Differences were considered to be significant at the $P \leqslant 0.05$ level.

EXPERIMENTAL RESULTS

In the first stage of the investigation sensitivity of mice of the AKR, C3H/Sn, C57BL/6, and CC57BR lines was compared with that of the highly resistant BALB/c line of mice. Only C3H/Sn mice were found to be significantly more sensitive to the action of CP than BALB/c mice. Mice of the other lines did not differ significantly as regards relative immunodepression from BALB/c mice.

Differences found between BALB/c and C3H/Sn mice still remained when the immunodepressive action of CP was studied simultaneously on BALB/c, C3H/Sn, and DBA/2 mice (Table 1). These differences were most marked when minimal and intermediate doses of the agent were used. It should be pointed out here that DBA/2 mice were significantly more sensitive than C3H/Sn mice.

In the next stage of the investigation the problem of whether the differences found in BALB/c, C3H/Sn, and DBA/2 mice were still present when thiotepa and SL were used as immunodepressants was studied. The results of these experiments are given in Tables 2 and 3.

A similar picture also was observed when the immunodepressive action of SL was studied, the only difference being that the differences were significant whatever dose of SL was used.

These experiments thus revealed distinct interlinear differences in the sensitivity of BALB/c, C3H/Sn, and DBA/2 mice to the immunodepressive action of alkylating agents belonging to different classes according to chemical structure. On the whole it can be concluded that DBA/2 and C3H/Sn mice are highly sensitive, whereas BALB/c mice are relatively resistant to the immunodepressive action of CP, thiotepa, and SL. Consequently, regardless of differences in structure of the immunodepressant studied and possible differences in the pharmacokinetic and pharmacodynamics of these agents in mice of the different lines studied, the degree of sensitivity to immunodepressive action is maintained. Differences found between mice of the various lines may be due mainly to the character of interaction of immunocompetent target cells with the immunodepressants.

The elucidation of these problems will be a task for future research.

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INTERPHASE CHROMATIN OF HUMAN CELLS WITH DIPLOID AND HAPLOID GENOME

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A previous analysis of formal denaturation of intracellular DNP by fluorescence microscopy and acridine orange staining in the writers' modification [4] showed that the structure of the interphase chromatin of normal human lymphocytes exhibits marked polymorphism under the influence of temperature: In about 40% of cases, irrespective of sex, the melting profiles of lymphocyte chromatin consist of a curve with six (or seven) maxima at definite temperatures; in 60% of the cases types of deviations, repeated regularly in different individuals, and whose character depended on sex, were found. The commonest type of deviation in men of the control group was absence of a maximum at 85°C [5].

In the present investigation the structural features of chromatin of human cells with equal quantities of genetic substrate, i.e., with a haploid set of chromosomes (and with the highest degree of condensation) were studied.

EXPERIMENTAL METHOD

The structure of chromatin of mature spermatozoa from 10 normal men aged from 25 to 40 years with normal spermatogenesis (the ejaculate was analyzed at the Family and Marriage Guidance Clinic, Sverdlovsk District Health Department) by comparative analysis of the melting curves of DNP from spermatozoa and lymphocytes of the same individual.

Changes in the structure of chromatin in response to temperature were tested relative to the quantity (F_{530}) of bound luminescent dye (acridine orange — AO). Tests were carried out on films of ejaculate stored in fixing solution (acetone:ethanol = 1:1) for not more than 72 h.

The intensity of luminescence of AO bound with DNA of chromatin from spermatozoa was measured on an MSP-0.5(P) microscope-photometer (from "Opton"). The excitation wavelength was λ = 365 nm. The intensity of luminescence was determined at λ = 530 nm. Thermal denaturation of chromatin in the spermatozoa was induced by Ringertz' method [9] in the writers' modification [4]. Values were read with an interval of 1-2°C.

As the control, melting profiles of lymphocyte chromatin from the same individual were tested in parallel experiments. The methods of taking blood, preparation of the films for spectrofluorometric analysis of the lymphocyte chromatin, the parameters of the apparatus, and calculation of the coefficient "α" were all described previously [6].

Melting profiles of chromatin from lymphocytes and spermatozoa of each individual were compared for all types on the melting curve with respect to mean intensity of fluorescence and the parallelness of the change in its intensity. Significance of differences were assessed by a special variant of two-factor dispersion analysis (F^2). The significance of differences in the mean level of the processes was determined as the significance of the

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