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Chromosome Breakage in Children Treated with LSD-25 and UML-491

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THE PROBLEM OF CHROMOSOMAL BREAKAGE reportedly found in humans using LSD-25 has attracted considerable research attention. Thus, Cohen et al. and Irwin and Egozcue have found increased occurrence of chromosomal breakage in humans exposed to LSD-25.[5,6] Skakkeback et al. have reported several breaks, gaps, and unidentifiable fragments in the meiotic chromosomes of mice treated with LSD-25.[9] However, several other workers could not find any increased incidence of chromosomal damage in humans treated with LSD-25.[3,7,10] Although many workers did not have an opportunity to study the incidence of abnormalities in the chromosomes of subjects before the administration of LSD-25, Abuzzahab et al. and Taylor et al. have studied human leucocyte mitotic chromosomes both before and after administration of LSD-25 to the same group of subjects.[1,11] These workers could not correlate effects of LSD-25 with human chromosomal breakage.

The question of the effect of LSD-25 is an extremely important one, not only because of the psychopharmacological effects of LSD-25, but also because of the possibilty that many drugs may act on chromosomes, with resultant effects on the genetic information, both in humans and animals, during extended periods of drug administration, as is the case in psychiatric patients. The present paper summarizes our findings of chromosomal breakage in psychiatric patients of the Children's Unit.

Materials and Methods

Fifteen patients were given LSD-25, on a treatment basis, as outlined in Table 1. Other subjects received UML-491, or a combination therapy consisting of both LSD-25 and UML-491. The average dose of LSD-25 for the whole population was 142.4 μg. per day, per patient. The number of days of LSD-25 administration, varied in this group from two days to 1366 days. All cases were treated from 1961 through 1965. Most of the cases, however, were treated more intensively from 1962 through 1964. Eight cases were on LSD-25 and UML-491. In this category the average daily dose of LSD-25 per patient was 97.4 μg., while the average dose of UML-491 per patient, per day was 4.54 mg. The duration of treatment in this group varied from 43 to 509 days. The number of patients who received UML-491 only, were nine. The average daily dose for the whole group was 6.86 mg. per day per patient, with the treatment ranging from 21 days to 243 days. Twenty-five controls (psychiatric child patients) did not receive LSD-25 or UML-491. Besides these patients, another subject (age 14 years) was also studied. This subject has never had any medication, is a well-rated student and a member of the general population and had a 4 per cent chromosome breakage. There were also seven adult

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nontreated, nonhospitalized employees studied. The ages of all the 58 child subjects ranged from seven to 15 years. The ages of the seven adults varied from 20 to 43 years.

The leukocyte mitotic chromosomes were studied according to standard methods.[13] The blood was drawn into a heparinized syringe; 10 ml. of blood being collected in 1 ml. of psysiological saline containing 10 mg. of heparin (specific activity 130 I.U. per milligram). Cultures were carried out using both plasma and whole blood. Although the cultures were not disturbed during incubation (the incubation lasted for 72 hrs. at 37° C), they were opened (under strictly sterile conditions) once a day to remove any excess CO_2 formed and to oxygenate the cultures more fully. While doing this, great care was taken not to disturb or shake the cultures. The final examination of the stained metaphase plates was carried out by workers who were not aware as to each patient's drug regimen. Examination of a minimum of 100 metaphase plates for each subject was conducted under an oil immersion lens, in a B&L Dynazoom binocular microscope, in order to determine chromosome number and chromosome breakage. Suspected chromosomal aberrations, including breaks, were further investigated by making photomicrographs of the metaphase plate.

Results

The quantity of LSD-25 administered per patient varied from 37.5 μg. to 165.8 μg. per day, per patient over a long period, extending in some cases, over three years. The psychiatric aspects of this therapy have been reviewed elsewhere,[2] In most cases treatment was stopped either in 1964 or in 1965. The chromosomal studies were done in 1966 through 1968. There was a total of 12 breaks out of 1500 metaphase plates in the subjects treated with LSD-25. The resulting average percentage of breaks in this group is 0.8 per cent. In no case was there more than one break per single metaphase plate in this group. In the subjects treated with UML-491, there was a total of five breaks for 900 metaphase plates, yielding an average of 0.56 per cent breakage. Again, no single metaphase plate had more than one break. In the subjects treated with both LSD-25 and UML-491, there was a total of seven breaks for 700 metaphase plates, yielding an average of one percent for chromosome breakage. This is not significantly higher than the per cent breakage in the controls or in persons treated with LSD-25 or UML-491 as shown by the Fisher t test. In the control group there were 2500 metaphase plates examined for 25 hospitalized children. There were 17 breaks out of these 2500 metaphase plates giving an average of 0.68 per cent breakage. The other nonhospitalized control child was 14 years old and out of 100 metaphase plates examined in the case of this subject there were four breaks as shown in Table 1. Including this subject, there are 26 control children with a total chromosomal breakage of 0.81 per cent. In the seven adults there were a total of four breaks for 700 metaphase plates. Thus, giving a 0.57 per cent chromosome breakage. The results in Table 1 show the average daily dosage of the drug and average number of days of treatment per subject. It also shows the number of persons who have no breaks or have a one to five percent breakage. To illustrate, out of seven subjects who received both LSD-25 and UML-491, there were three who had no breakage at all, two that had 1 per cent breakage, one that had 2 per cent breakage, and one that had 3 per cent breakage. As explained in the footnote to Table 1, there was also another patient who received both LSD-25 and UML-491 and had nine breaks out of 100 metaphase plates. The number of breaks in this case was thought to be to high to be included

with the rest of the cases and this patient was not shown in the results in Table 1. There were 26 child control subjects. Of these, 15 subjects had no breaks at all, six had 1 per cent breakage and three had 2 per cent breakage and one had 4 per cent and one had 5 per cent breakage. Thus, there were a total of 21 breaks for 2600 metaphase plates including an average of 0.81 per cent chromosome breakage.

Standard deviations were computed according to standard methods and presented in Table 1. The differences between the controls and the drug-treated groups, with respect to the frequency of chromosomal breakage, is not significant as shown by the application of the Fisher t test. These findings support our earlier report as to the lack of induced chromosomal breakage in psychiatric child patients treated with LSD-25 and/or UML-491.[3]

Table 1.—Effect of Treatment with LSD-25 and/or UML-491 on Human Chromosome Breakage

	Controls		LSD–25	UML	LSD and UML-491
	Adults	Children	Children	Children	Children
Number of subjects	7	25*	15	9	7†
Average daily dose of drug per subject	0	0	142.4 μg.	6.86 mg.	97.4 μg. and 4.54 mg.
Average number of days of treatment per subject	0	0	358	103	216 and 137
No. of subjects with					
No breaks	4	15	7	5	3
1 break	2	6	4	3	2
2 breaks	1	3	4	1	1
3 breaks	0	0	0	0	1
4 breaks	0	0	0	0	0
5 breaks	0	1	0	0	0
Per 100 metaphases					
Total breaks	4	17	12	5	7
Average % breaks	0.57	0.68	0.80	0.56	1.00
S. D.‡	±0.88	±1.00	±0.86	±0.73	±1.15

* Does not include one other (nontreated, nonhospitalized) child from general population. This child had four breaks out of 100 metaphase plates.

† Does not include one patient who had nine breaks. This patient is thought to be widely deviant.

‡ The large standard deviations denote the skewed distribution of breaks in the subjects.

Discussion

The question of damage to meiotic and mitotic chromosomes is an important one, especially in view of prolonged periods of drug treatment in psychiatirc patients. It is also important from an eugenic point of view. While some of the other reports have shown leukocyte chromosome damage in LSD-users, our studies have not indicated any significant damage. Our studies have been carried out on children who have received LSD-25 some two to four years prior to the chromosome studies. It is possible that the damaging effects of

LSD-25 on the leukocyte chromosome may have been rectified in these two to four years. This would signify that treatment with LSD-25 had no long lasting effects on leucocyte chromosomes.

LSD-25 is an extremely potent sympathomimetic agent.[8] It has the ability of increasing body temperature and metabolic rate.[12] It is possible that one of the effects of LSD-25 may lie in increasing the mitotic rate of leukocyte replication. When the rate of replication is increased, it is conceivable that the quality of the product of replication may be defective and give rise to the breakage in the chromosomes produced. This possibilty, however, must be investigated further.

In our studies, we have given the pure drug LSD-25 to the subjects. In many of the other studies reported, the LSD-users were obtained from general population, without knowledge as to the purity of the LSD used and/or other medications that the subject had been using. The LSD-user in the general population may already be partly deviant in his psycho-biology. He might have been using several drugs at different doses, for several months or even years. For this reason it becomes important to study the effects of LSD-25 in a population known to be free from other drugs and chromosomal aberrations before the use of LSD-25.

The breakages in our study are much lower than in those of other groups,[4,6] The question of laboratory techniques may be a factor here. A germane point is that the effect of LSD may some how be related to the percentage breaks that a given laboratory gets in its regular and routine work involving both drug treated and drug free groups. This can perhaps be best studied further by personnel from one laboratory working in another laboratory for a period of time.

Summary

The percentage of chromosome breakage was studied using 57 child psychiatric patients and eight non-hospitalized, non-drug treated volunteer controls. Out of this, there were 15 subjects who received LSD-25, nine who received UML-491 and eigh who received both the drugs. The per cent breakage in seven adults was 0.57 per cent and in 25 control children was 0.68. The per cent chromosome breakage in the children treated with LSD-25 and/or UML-491 ranged from 0.56 to 1 per cent. These differences, however, are not statistically significant. It is concluded that treatment with LSD-25 had no long lasting effects on human leukocyte chromosomes in child psychiatric patients.

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