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Effect of Maternal Medication during Pregnancy upon Behavioral Development of Offspring

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The present study was designed to investigate the effects of four centrally acting drugs (chlorpromazine 6.0 mg/kg, reserpine 0.1 mg/kg, meprobamate 60.0 mg/kg and phenobarbital 8.0 mg/kg) upon the behavioral development, especially development of intelligence, of the offspring of rats, to which these drugs were administered during four days of the early and late periods, from the 5th to 8th and from the 17th to 20th days, of pregnancy.

When the offsprings were 90 days old, the level of their general activity was measured by the revolving drum technique. In both the administration periods the activity level was significantly lower only in the meprobamate-treated group than that in the control group.

Intelligence of 120-day-old animals was tested by Hebb-Williams' enclosed field test. When the administration was done in the early period of gestation, the intelligence of the chlorpromazine-, meprobamate- or phenobarbital-treated groups was each significantly lower than that of the control group. But when the administration was done in the late period of pregnancy, only the meprobamate-treated group showed a significantly lower intelligence than the control group.

These findings were also supported by the results of the brain activity test by a Woodbury's electroshock apparatus.

From these results it is suggested that some of the centrally acting drugs have possibility of causing a retardation of mental development of offspring, and that the prenatal medication in the early period of gestation has a profounder effect than that in the late period.

With the advance of pharmacology, new drugs have recently been developed one after another, and not only physicians but also laymen have employed these drugs without scruple. Under these circumstances, the effects of the drug administration during pregnancy upon the fetus or offspring have raised a social problem.

Until recently, the effects of drug administration during pregnancy upon the fetus and infant have been primarily studied in the field of teratology. But since the studies on the placental function revealed that the placenta was not an effective barrier between the maternal and fetal circulations, and most of drugs administered to mothers in pregnancy passed comparatively easily into the fetal

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circulation, the effects of the drug administration during pregnancy have increasingly been studied in relation to neonatal jaundice and so on.[1,2] There are a few studies which were carried out to determine the effect of drugs on the behavioral development of the infant[3,4] but their effect on the development of intelligence has not yet been studied thoroughly.

Hebb and others[5] suggested that the chemical factors in uterine environment had some effects on the development of intelligence of offspring,. The present study attempted to investigate experimentally the effects of some centrally acting drugs on the behavioral development, especially on the development of intelligence of the offspring, by testing the effect of prenatal medication on their general activity, intelligence and brain activity.

Chlorpromazine, reserpine, meprobamate and phenobarbital were selected in the present study, because all these four drugs have been used in the obstetric clinics. The first three of these are tranquillizers and the last is a hypnotic.

METHODS

Observations were made on 100 male albino rats. They were offsprings of the mothers to which the drugs had been administered during pregnancy. The mothers were fifty female albino rats of Wistar strain that were about 120 days of age when they were mated. After copulation was ascertained by the presence of spermatozoa on the examination of the vaginal smear, the gravid rats were randomly assigned to four groups according to the kind of drugs, and to a control group. A half of each group was administered a drug (or distilled water in the case of control group) in the early period of gestation (during four days, from the 5th to 8th) and the other half was administered the same drug in the late period of gestation (during four days, from the 17th to 20th).

The drugs administered were chlorpromazine, reserpine, meprobamate and phenobarbital. All the samples were prepared from powder in distilled water at room temperature, except meprobamate which was dissolved in distilled water heated to 80°C and cooled at room temperature. The drug was administered subcutaneously to the dorsal areas. The procedure was the same as was used by Werboff.[4] Daily dosages (mg/kg) and concentrations (mg/ml) of the drugs were as follows:

Chlorpromazine:	6.0 mg/kg,	10.0 mg/ml
Reserpine:	0.1 mg/kg,	0.1 mg/ml
Meprobamate:	60.0 mg/kg,	50.0 mg/ml
Phenobarbital:	5.0 mg/kg,	10.0 mg/ml

The daily dosage of each drug was approximately five times as large as human daily one. It was divided into two equal doses and injected every 12 hours, at 7 a.m. and 7 p.m., for consecutive four days. The drug administration was made in the early and late periods of gestation, as mentioned above. The control group

was administered 1.0 ml of distilled water with the same procedure, every 0.5 ml twice a day.

Of 337 90-day-old offsprings, 10 males were randomly selected from each of a total of ten groups described above, thus making 100 rats in all. They had an average body-weight of 180 g, ranging from 150 to 220 g, on the day of the group assignment.

Measurement of general activity: In the offspring at 90 days of age, the general activity was measured by the method of the revolving drum technique. The apparatus used was an ordinary activity wheel 52 cm in diameter which was constructed to revolve only in one direction. One-fifth of revolution was counted as one point by the counter. The scored points for 15 minutes were recorded every day. The measurement was carried out for three consecutive days and the sum total of the scores were regarded as an indicator of the general activity.

Intelligence test: The measurement of intelligence was carried out on 120-day-old animals by Hebb-Williams' enclosed field test. The apparatus in the present experiments was the same as described by Hebb and Williams[6] except that the floor and walls of the apparatus were painted white and the floor was partitioned into twenty areas of 18 cm squares by yellow lines, for the purpose of easily recording the route of running. The test situations used consisted of six problems which were named by Hebb as I-A, I-D, I-F, I-G, I-H and I-L.

After preliminary training for ten days, the tests were carried out for three consecutive days. After 23-hour food deprivation the rat was subjected to examination at a rate of two problems a day, five trials for each problem. The intertrial interval was about ten seconds. After the daily tests the rat was returned to his home cage and fed for one hour.

The scoring in this test was as follows: the routes of running of the rat tested are recorded at every run. One point was given when the animal took a direct route without noticeable deviation. The total score of an animal in six problems was regarded as an indicator of intelligence.

Measurement of activity level of brain by electroshock seizure: For the purpose of investigating the activity level of the brain, the threshold stimulus (mA) for the minimum electroshock seizure was measured, and the pattern and duration of the maximum seizure was also studied in each subject. The minimum and the maximum seizures were produced by a stimulator which was desinged by Woodbury and Davenport.[7] The apparatus and techniques were the same as those described in detail by them.

At the beginning of these tests the subjects were 150 days old. In each subject the trial was repeated at two days' intervals until the threshold was

determined. The duration and pattern of the maximum seizure was determined once in each animal after the threshold determination.

RESULTS

1. The effect of the prenatal medication upon the general activity of offsprings

Table 1 shows the mean scores (converted to logarithm) of the activity test with a wheel for the drug-treated and control groups of which mothers had been treated during pregnancy, as described above. The statistical test between the mean scores of the drug-treated and control groups was performed with *t*-test. Only the meprobamate-treated groups with drug administration either in early period or in late period of gestation, had a slightly significant difference in the score, that is, the score was smaller than in the control group at 5 per cent level. There was, however, no significant difference among the other three drug-treated groups and their control ones.

TABLE 1. *Mean scores (converted to log.) of activity wheel test*

Group	Early period			Late period		
	N	M	SD	N	M	SD
Chlorpromazine	10	2.4	0.25	10	2.2	0.21
Reserpine	10	2.6	0.23	10	2.3	0.24
Meprobamate	10	2.4*	0.38	10	2.1*	0.11
Phenobarbital	10	2.4	0.37	10	2.3	0.15
Control	10	2.6	0.24	10	2.3	0.11

* Difference between drug-treated and control groups significant at 0.05 level
N: Number of subjects M: Mean value SD: Standard deviation

TABLE 2. *Mean scores of intelligence test*

Group	Early period			Late period		
	N	M	SD	N	M	SD
Chlorpromazine	10	8.9†	2.38	10	10.0	3.20
Reserpine	10	10.7	4.32	10	11.6	2.39
Meprobamate	10	9.4†	2.58	10	9.5*	1.50
Phenobarbital	10	8.3†	1.68	10	9.9	2.76
Control	10	12.6	3.38	10	11.3	1.64

* Difference between drug-treated and control groups significant at 0.05 level
† Difference between drug-treated and control groups significant at 0.01 level

2. The effect of the prenatal medication upon the intelligence of offsprings

Table 2 gives the mean scores on the enclosed field test on the test subjects. Inspection of the table reveals that the scores in each of drug-treated groups were smaller than those in the control group. The *t*-test was used for statistical analysis. In the groups of drug administration in the early period of pregnancy,

the scores in the chlorpromazine-, meprobamate- or phenobarbital-treated group were each significantly smaller than those in the control group.

But among the groups of drug administration in the late period, only the meprobamate-treated one showed a smaller intelligence score than the control.

3. The effect of the prenatal medication upon the brain activity of offsprings

a) The threshold of the minimum seizure

The values of the minimum seizure thresholds (mA) of the subjects are presented in Table 3. The method of statistical analysis was the same as that described above. In the subjects whose mothers had been treated in the early period of gestation, the minimum threshold values in the chlorpromazine-, meprobamate- and phenobarbital-treated groups were each significantly higher than those in the control groups at 1 per cent level. On the other hand, the minimum threshold value in the reserpine-treated group was 21.9 mA and this value was almost identical with 21.4 mA in the control group. Needless to say, the difference between these two groups was not significant.

On the other hand, the mean values of the threshold in the subjects of which mothers had been treated in the late period of gestation showed less difference than those in the groups with drug administration in the early period of pregnancy. But the values in the chlorpromazine- and meprobamate-treated groups were each significantly higher than those in their control groups.

TABLE 3. *Mean values of minimum seizure threshold* (mA)

Group	Early period			Late period		
	N	M	SD	N	M	SD
Chlorpromazine	10	23.2†	0.87	10	22.6†	0.17
Reserpine	10	21.9	0.71	10	21.6	0.22
Meprobamate	10	23.4†	0.72	10	22.6†	0.18
Phenobarbital	10	23.5†	0.39	10	21.8	0.14
Control	10	21.4	0.50	10	21.7	0.14

† Difference between drug-treated and control groups significant at 0.01 level

b) The duration and pattern of the maximum seizure

The maximum seizure pattern consists of the following three phases: tonic flexion, tonic extension and clonus.

Table 4 presents the mean duration (sec) of each of the three phases in the rats of which mothers were treated with the drugs during pregnancy. The statistical analysis was carried out by using the *t*-test. With the drug administration in the early period of gestation, the duration of the phase of tonic flexion in the meprobamte- and in the phenobarbital-treated groups was significantly shorter than that in the control groups. On the other hand, there was no significant

TABLE 4. *Mean values of maximum seizure duration (sec)*

	Group	N	Tonic flexion		Tonic extension		Clonus	
			M	SD	M	SD	M	SD
Early period	Chlorpromazine	10	2.0	0.42	7.4*	0.83	6.3	1.55
	Reserpine	10	2.4	0.74	6.4	1.28	8.4	2.17
	Meprobamate	10	1.7†	0.46	7.0	1.47	7.8	2.29
	Phenobarbital	10	1.8*	0.39	7.6†	0.97	8.8	2.88
	Control	10	2.5	0.72	6.5	0.81	7.8	1.91
Late period	Chlorpromazine	10	2.3	0.58	6.0	0.97	7.5	2.65
	Reserpine	10	2.2	0.48	6.6	2.49	6.6	2.39
	Meprobamate	10	2.1	0.22	7.6*	1.02	7.4	2.06
	Phenobarbital	10	2.3	0.50	7.1	3.18	7.0	3.19
	Control	10	2.3	0.43	6.4	0.78	7.4	1.87

* Difference between drug-treated and control groups significant at 0.05 level
† Difference between drug-treated and control groups significant at 0.01 level

difference between the groups treated with two other drugs and their control ones. The duration of the tonic extension phase in the chlorpromazine- and phenobarbital-treated groups was each significantly longer than that in the control groups. As regards the phase of clonus there was no significant difference between the drug-treated group and the control group, so far as four drugs have been treated.

But with the drug administration in the late period of pregnancy, there was no significant difference in duration of the tonic flexion phase between the drug-treated and the control groups. As regards the tonic extension phase, however, only the duration in the meprobabmate-treated group was slightly longer than that in the control group. As to the phase of clonus, there was no significant difference in its duration between the drug-treated and control groups.

DISCUSSION

It was the purpose of the present study to investigate the effect of the prenatal medication on the behavioral development, especially on the development of intelligence of the offspring.

The data of the intelligence test showed that when the drug was administered in the early period of gestation, the intelligence scores of each of the chlorpromazine-, meprobamate- and phenobarbital-treated groups were significantly lower than those of the control, but when the administration was done in the late period, only the meprobamate-treated group had a significantly lower score than the control group.

On the other hand, the data of the general activity which was measured by the revolving drum technique indicated that regardless of the administration period, only the meprobamate-treated group had a slightly significant, lower

activity level than the control group, and that the scores in the groups treated with the other drugs scarcely differed from those in their control groups. Accordingly, it can be said that the difference of the intelligence score does not imply the difference in the general activity level. But, it suggests rather that the prenatal medication has the possibility of producing the retardation of intelligence of the offspring.

The experiments on electroshock seizure showed that the thresholds for the minimum seizure in some drug-treated groups were significantly higher than those in the control group, and the maximum seizure in these drug-treated groups showed a tendency to have a shorter duration of tonic flexion, or longer duration of tonic extension. On the basis of the studies on the electroshock seizure,[8-11] the present results can be interpreted as indicating that the prenatal medication has the possibility of producing biochemical changes in the central nervous system of the offspring, which, in turn, decrease the brain activity. The results agree well with those obtained with the intelligence test: the group of decreased brain activity corresponds with that of lowered intelligence.

Thus, it was found that the prenatatal medication exerted a profound effect on the behavioral development, especially on the development of intelligence of the offpsring. The mechanism of these drug actions cannot be revealed by the present study alone, but it may be suggested that the prenatal medication produces a long-lasting or relatively permanent biochemical changes which cause retardation of mental development.

In the present study, it was found that the administration of the drugs in the early period of gestation had profounder effects than that in the late period: the drug administration in the early period of pregnancy had a more adverse effect than that of the late period. Similar findings have been obtained also in the clinical and experimental studies of malformations.[1,2] Thus, the period of pregnancy in which drug administration has been made seems to be a decisive factor.

Among the drugs used in the present study, meprobamate produced the most serious effect on intelligence and general activity. On the other hand, reserpine had no effect on either kind of behavior. From these facts it is suggested that among centrally acting drugs, there may be some difference in their effect on the behavioral development of the offspring. More precise experimental studies are required for elucidating the mechanism of specific drug actions.

The daily dosage of each drug used in the present experiments was relatively small, only five times of the human daily dosages. Nevertheless, some of the drugs produced a serious retardation of intelligence. This raises problems of clinical interest concerning the prenatal medication.

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