ticoids. Presumably, the mineralocorticoids plus the small amount of Na in the Lonalac, and the increased water intake, enabled the animals to maintain sufficient arterial pressure to ward off circulatory collapse and enough renal activity to prevent lethal accumulations of potassium during the 8-12 days of the experiment. If salt, water and DCA are withheld during the post-reaction period, both electro- and Metrazol-treated adrenalectomized dogs usually succumb within 24-36 hours unless glucocorticoids are administered(11,13).

The sharp but evanescent rise in plasma Na which is an invariable accompaniment of the electroconvulsive and Metrazol reaction in dogs, has been discussed previously(12).

Summary. Adrenalectomized dogs upon recovery from the severe convulsions following Metrazol injection can be maintained active and vigorous 8-12 days with low plasma volumes but with normal electrolyte patterns. This is accomplished by daily i.v. injections of desoxycorticosterone or aldosterone, a low Na diet and allowing access to water. The plasma volume falls immediately after Metrazol averaging 35.2% below preinjection levels and persists at low values. The mean arterial pressure slowly declines eventually necessitating administration of glucocorticoids. The plasma volume fall occurs so rapidly that fluid loss to the exterior must be ruled out as a cause for the low volumes. Shift of water into cells evidently does not take place since the plasma electrolytes, after an initial marked rise, rapidly decline to preinjection levels. The convulsant drug apparently induces intense vasoconstriction, evidently closing large areas of the microcirculation, trapping and pooling substantial quantities of blood at the vascular periphery. Presumably adrenalectomized, Metrazol treated dogs receiving adequate mineralocorticoids, a low Na diet and free access to water are unable to relieve the induced vasoconstriction and reestablish free blood flow in the small vessels of the circulatory tree in the absence of C_{11}oxysteroids.

1. Swingle, W. W., Parkins, W. M., Taylor, A. R., Hays, H. W., *Am. J. Physiol.*, 1936, v116, 438.

2. Cizek, L. J. Semple, R. E., Huang, K. C., Gregersen, M. I., *ibid.*, 1951, v164, 415.

3. Painter, E. E., Holmes, J. H., Gregersen, M. I., *ibid.*, 1948, v152, 66.

4. Holmes, J. H., Cizek, L. J., *ibid.*, 1951, v164, 407.

5. Gregersen, M. I., Stewart, J., *ibid.*, 1939, v125, 142.

6. Shu Chien, Gregersen, M. I., *Physical Techniques in Biological Research*, vIV, Academic Press, New York, 1962, 63.

7. Delgado, J. M., *Physiol. Revs.*, 1960, v40, 146.

8. Hoff, C., Kell, J. R., Carrol, M., *ibid.*, 1963, v43, 68.

9. Weil-Malherbe, H. J., *J. Mental Science*, 1955, v101, 156.

10. Montagu, J. D., *ibid.*, 1955, v101, 110.

11. Swingle, W. W., Da Vanzo, J., Crossfield, H. C., Glenister, D., Osbcrn, M., Rowen, R., Wagle, G., PROC. SOC. EXP. BIOL. AND MED., 1959, v100, 617.

12. Swingle, W. W., Swingle, A. J., *ibid.*, 1963, v112, 847.

13. Swingle, W. W., Da Vanzo, J., Glenister, D., Wagle, G., Osborn, M., Rowen, R. *ibid.* 1960, v104, 184.

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Chlorpromazine Effects on Pregnancy and Offspring in Mice.* (28505)

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Chlorpromazine has attained extensive clinical use as a tranquilizing agent for alleviating symptoms in emotionally disturbed subjects. Its reported psychotropic and antiemetic effects have also made it useful for controlling premenstrual tension and anxiety, nausea, and various complications of pregnancy(1,2). In view of its potentiation of a variety of anesthetic agents, chlorpromazine

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has also been used as premedication for anesthesia in obstetric patients(3). Although mode and site of action in the central nervous system have not been clarified, the tranquilizing properties of phenothiazines have been attributed to a depression of the reticular formation and sympathetic activity of the hypothalamus(4,5). However, in addition to the more direct effects on the nervous system, a number of studies dealing with drug effects on hypothalamic and adenohypophyseal interrelationships in reproductive physiology have indicated that chlorpromazine may also produce menstrual irregularities and delays in ovulation in human subjects(6), and inhibition of estrus and ovulation in experimental animals(7,8,9,10). A number of preliminary studies have also suggested delays in length of gestation as well as decrease in litter size and weight of offspring following chlorpromazine treatments during pregnancy in rats(11,12,13). Since reproductive cycles in spontaneously ovulating species are assumed to be in part under hypothalamic influence, chlorpromazine effects on reproductive cycles and gestation have been attributed to interference of gonadotropin secretion of the pituitary resulting from drug effects on the hypothalamus. However, reported chlorpromazine effects on reproductive cycles, length of gestation, and litter size and weight of offspring can also be affected by possible drug-induced maternal debility, weight loss, as well as possible direct drug effects on the fetus(14). The aims of this investigation were to obtain some quantitative data on the effects of chronic chlorpromazine treatments in mice during pregnancy on delays between mating and birth of litters, maternal debility evidenced by changes in weight and behavior after daily chlorpromazine treatment, as well as drug effects on litter size and weight of offspring at birth.

Method. Eighty inbred C57Bl/10 mice breeder pairs were selected from a colony, mated, and assigned to 4 groups. Twenty females received orally 4 mg/kg and 20 female mice received 16 mg/kg of chlorpromazine throughout pregnancy. The remaining 40 females were divided into 2 groups, received corresponding volumes of placebo orally, and served as controls for the 2 dose levels.

To obtain reliable fecundity, fertility, and incidence of pregnancy data, large numbers of breeder pairs were mated and screened for parity. The 80 breeder pairs for the study were then selected only after each breeder pair yielded 2 consecutive litters within a 60-day period after mating. This selection procedure also provided a record on duration of 2 consecutive pregnancies as well as data on litter size and weight so that breeder pair assignments to drug or placebo treatments would be based on random distributions of fertility, incidence of pregnancy, and duration of gestation. To preclude disturbance of the estrous cycle following mating, onset of pregnancy was not established by vaginal smears. Since the expected mean duration of one complete estrous cycle in unmated C57Bl/10 mice represents approximately 5-6 days, the 80 screened breeder pairs were mated and left undisturbed for one complete estrous cycle. To exclude drug-induced disturbances of the estrous cycle, oral drug or placebo treatments were started 6 days after mating and continued daily until the litters were born. Males were removed when weight increments indicated pregnancy in females. At the time of the third mating, all females were approximately 160 days of age.

To obtain behavioral measures of drug effects on pregnant mothers as index of maternal debility, all 80 females were tested each day in an open field test for exploration and activity level one hour after drug or placebo. Since the higher dose level resulted in frequent sedation in some mothers lasting 1 to 5 hours after drug administration, medians and the Mann-Whitney test were used to evaluate differences in maternal behavior between drug and placebo mothers during pregnancy. A statistical analysis of variance design was used to evaluate chlorpromazine effects on delays between mating and birth, drug-induced changes in maternal weight, and litter size and weight of drug and placebo offspring at birth.

Results. The effects of 4 and 16 mg/kg

TABLE I. Comparisons of 4 and 16 mg/kg Drug and Placebo Effects on Gestation and Litter Size and Weight.

Group	Days between mating and birth*	Mean litter size*	Mean litter wt*
Placebo	22.35	8.0	1.41
4 mg/kg drug	23.85	7.9	1.33
16 " "	28.80	5.8	1.17

* Group means.

chlorpromazine on delays between mating and birth, as well as effects on litter size and weight of drug and placebo offspring at birth are presented in Table I. There was a significant difference ($p = <.005$; $F = 8.76$; $df = 1,60$) in number of days from mating to birth between drug and placebo mothers. The 16 mg/kg dose level produced the greatest delay between mating and birth compared to the 2 placebo groups as well as the 4 mg/kg drug mothers. The drug mothers had a significantly lower number of offspring per litter ($p = <.05$; $F = 4.19$; $df = 1,76$) than the placebo groups. With the 2 dose levels combined, the mean litter weights of the drug mothers were also significantly lower ($p = <.001$; $F = 41.92$; $df = 1,60$) than the mean litter weights of the placebo mothers.

A comparison of female weight levels at the time of mating revealed no significant differences among drug and placebo females. To obtain measures of drug effects on maternal weight gains during pregnancy independent of possible drug effects on fetal size and weight, the sums of litter weights were subtracted from the mothers at birth and adjusted drug and placebo mother weight levels were compared. Although the drug mothers weighed less at birth of their litters than the placebo mothers, the maternal weight comparisons among the 4 groups were not statistically significant but highly suggestive. Fig. 1 indicates that incremental changes in weight in drug-treated mothers were less than those of placebo mothers during pregnancy.

No significant differences in behavior were observed between placebo mothers and the 4 mg/kg drug mothers one hour after oral drug or placebo treatments during the course of pregnancy. However, the 16 mg/kg oral dose level produced frequent sedation in some mothers lasting 1 to 5 hours after treatment, resulting in significant decrements ($p = < 0.001$) in activity levels as indicated by the open field test of exploration.

Discussion. Although phenothiazines have become useful clinically for controlling symptoms of emotional disturbances and nausea in human female subjects of child bearing age, only fragmentary experimental evidence is available on effects of psychotropic drugs on hypothalamic, pituitary, or ovarian functions, or possible indirect drug effects on reproductive physiology. Previous findings with animal subjects have suggested that chlorpromazine may effect endocrine changes associated with reproductive cycles, gestation, and fetal development by possible inhibition of gonadotropin secretion with alterations in release of luteotropic hormones of the adenohypophysis, as well as direct drug effects on the fetus through placental transfer(3,15).

The significant delays between mating and birth of the chlorpromazine-treated mothers under the experimental conditions of the present experiment suggest possible drug interference with fertilization or post-conception implantation and increases in length of gestation. These alternative possibilities can be clarified experimentally by separation of the males from the females following mating at the completion of one complete estrous

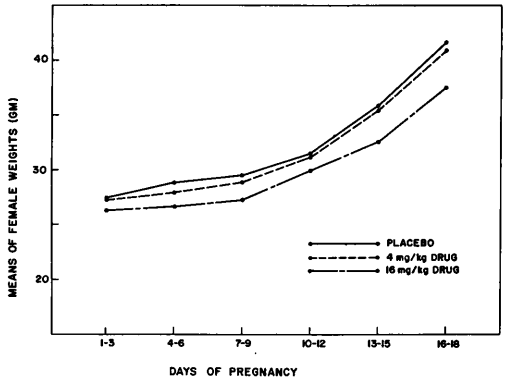

FIG. 1. Group means of incremental weight changes in 4 and 16 mg/kg chlorpromazine and placebo mothers during pregnancy combined into 3-day blocks.

cycle. The significantly lower number of offspring per litter of the chlorpromazine-treated mothers also suggest drug-induced abortions and resorptions of embryos. The significantly lower mean litter weights of the drug mothers could be attributed to lower incremental weight gains, maternal sedation and debility, as well as possible differences in maternal activity levels during pregnancy. In previous studies on chlorpromazine effects on gestation and parturition in the rat, chlorpromazine was also found to increase the length of gestation, produce abortions, and increase mortality in rat offspring during the first 24 hours after birth(11,13,16). These drug effects were most pronounced if chlorpromazine was administered during the early stages of pregnancy.

The additional implication of chlorpromazine and related phenothiazines in amenorrhea with delays in menstruation also suggest some caution in differential diagnosis of menstrual irregularities after repeated ataractic drug treatment(6). Since various species differences are apparent in reproductive cycles, and dosage levels in non-psychotic human subjects are generally lower for mild anxiety states. additional experimental studies are essential with human subjects as well as experimental animals to help clarify specific effects of psychotropic drugs on reproductive physiology.

Summary. The aims of this investigation were to obtain quantitative data on the effects of chronic chlorpromazine treatments in mice during pregnancy on delays between mating and birth of litters, maternal debility evidenced by changes in weight and behavior after daily chlorpromazine treatment, and drug effects on litter size and weight of offspring at birth. Eighty inbred C57Bl/10 mice breeder pairs were selected from a colony, mated, and assigned to 4 groups. Twenty females received orally 4 mg/kg and 20 female mice received 16 mg/kg of chlorpromazine throughout pregnancy. The remaining 40 females were divided into 2 groups, received corresponding volumes of placebo orally, and served as controls for the 2 dose levels. There was a significant difference in number of days from mating to birth between drug and placebo mothers. Drug mothers had a significantly lower number of offspring per litter than the placebo groups. Mean litter weights of the drug mothers were also significantly lower than the mean litter weights of the placebo mothers. Incremental changes in weight in drug-treated mothers were not significantly less than those of placebo mothers during pregnancy. Although no significant differences in behavior were observed between placebo mothers and the 4 mg/kg drug mothers, at the higher dose level highly significant differences were observed during pregnancy between drug and placebo mothers in activity levels as determined in an open field test for exploratory behavior.

1. Benaron, H. B. W., *Am. J. Obstet. and Gynec.*, 1955, v69, 776.
2. Sandberg, D. H., *Calif. Med.*, 1961, v94, 287.
3. Marx, G. F., *Anesthes.*, 1961, v22, 294.
4. Himwich, H. E., *J. Neuropsych.*, 1962, v3, 279.
5. Killam, E. K., *Pharmacol. Revs.*, 1962, v14, 175.
6. Whitelaw, M. J., *J.A.M.A.*, 1961, v175, 400.
7. Cranston, E. M., PROC. SOC. EXP. BIOL. AND MED., 1958, v98, 320.
8. Barraclough, C. A., Sawyer, C. H., *Endocrinol.*, 1959, v65, 563.
9. DeFeo, V. J., *Science*, 1956, v124, 725.
10. Purshottam, N., *Am. J. Obstet. and Gynec.*, 1962, v83, 1405.
11. Chambon, Y., *Ann. d'Endocrinol.*, 1955, v16, 912.
12. ———, *ibid.*, 1957, v18, 80.
13. Roizin, L., True, C., Knight, M., *Res. Publ. Assn. Nerv. Ment. Dis.*, 1959, v37, 285.
14. Clegg, M. T., Santolucito, J. A., Smith, J. D., Ganong, W. F., *Endocrinology*, 1958, v62, 790.
15. Franchi, G., Gianni, A. M., *Acta Anesth. Padova*, 1957, v8, 195.
16. Murphree, O. D., Monroe, B. L., Seager, L. D., *J. Neuropsych.*, 1962, v3, 295.

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