# Pregnancy and Liver Regeneration in Partially Hepatectomized Rats. (24475)

LEON L. GERSHBEIN (Introduced by C. G. Van Arman)
*Biochemistry Research Lab., Dept. of Biology, Ill. Inst. of Technology, Chicago, Ill.*

Because of the rapid proliferative changes involved in both pregnancy and liver regeneration, a study was undertaken of the interrelationship between these 2 phenomena in partially hepatectomized rats. According to an earlier report by Paschkis, Cantarow and Stasney(1), rats which were partially hepatectomized on the eighth or seventeenth day of gestation and sacrificed on the twentieth day of pregnancy suffered severe fetal damage. The latter ranged up to complete resorption of all fetuses in several cases and "liver regeneration was identical in nonpregnant and pregnant animals."

*Methods.* Mature female rats of Sprague-Dawley strain were employed. Virgin animals were exposed to fertile males for 7 days, then these females and nonpregnant controls were partially hepatectomized under ether anesthesia according to the procedure of Higgins and Anderson(2). The excised portions were dried to constant weight in an oven at 100°C. The rats were maintained in individual cages and administered water and powdered Purina Fox Chow *ad. lib.* They were sacrificed (ether) after an interval of 10.5 days and the livers removed and dried. The uteri of the gravid animals were closely examined at necropsy for extent of pregnancy or resorption; the data from the "negatives" were discarded. As described previously, the amount of liver regenerated or the increment was calculated from dry weights by subtracting the amount of tissue removed at surgery multiplied by 0.46 from the weight of the entire liver at necropsy(3).

*Results.* Body weights and liver increments for groups of controls, pregnant rats and those showing fetal resorption as well as the corresponding Fisher *t* values are given in Table I. The gravid animals ranged mainly from middle to late pregnancy, several being close to term when sacrificed. Microscopic examination of representative small liver sections removed at necropsy from the pregnant rats showed no remarkable pathology over those from the controls (hematoxylin-eosin stained).

*Discussion.* A marked increase in extent of liver regeneration in partially hepatectomized rats was caused by pregnancy. Presumably, one or more factors elaborated systemically during pregnancy predispose to such increased regenerative activity. A smaller but statistically significant elevation was also apparent in animals displaying resorption of the fetuses. In this regard, the liver increments were quite high for 5 of the 11 rats which exhibited recent or active resorption as compared to the remainder where this process was almost complete. The extensive resorptive changes should not be construed as due to liver regeneration as such but to the extreme surgery involved, numerous other types of which can lead to such damage.

*Summary.* The extent of liver regeneration in partially hepatectomized rats is markedly elevated by pregnancy. A lower but significant increase over nonpregnant controls was also evident with those animals displaying fetal resorption, especially where the latter was in active progress.

TABLE I. Body Weight and Liver Increment Data for Partially Hepatectomized Control and Pregnant Rats.

| | No. of animals | Body wt, g* | | Liver increment, g* | t |
|---|---|---|---|---|---|
| | | Initial | At necropsy | | |
| Controls (nonpregnant) | 25 | 205 ± 3.2 | 207 ± 3.6 | 1.524 ± .054 | |
| Pregnant | 16 | 215 ± 4.0 | 232 ± 5.7 | 1.933 ± .067 | 4.75† |
| Rats displaying fetal resorption | 11 | 215 ± 3.5 | 219 ± 4.8 | 1.740 ± .099 | 2.06† |

* Stand. error follows each ± sign. † $P < .05$.

This study was aided by grant from Toni Division of Gillette Co.

1. Paschkis, K. E., Cantarow, A., Stasney, J., *Fed. Proc.*, 1956, v15, 141.

2. Higgins, G. M., Anderson, R. M., *Arch. Path.*, 1931, v12, 186.

3. Gershbein, L. L., Labow, J. A., *Am. J. Physiol.*, 1953, v173, 55.

Received August 25, 1958. P.S.E.B.M., 1958, v99.

## Failure of Deoxyribonucleic Acid to Effect Somatic Transformation in the Rat.* (24476)

THOMAS L. PERRY AND DOROTHY WALKER (Introduced by Richard W. Lippman)
*Division of Chemistry and Chemical Engineering, California Inst. of Technology*

It is well established that isolated deoxyribonucleic acid (DNA) can produce heritable changes in several species of bacteria. This was first demonstrated by Avery *et al.*(1), who found that DNA extracted from a smooth (encapsulated) strain of *Pneumococcus* would transform rough (unencapsulated) cells into smooth cells. The capsule of the newly modified cells was serologically the same as that of the strain from which the DNA had been derived, and the conversion could be brought about in a rough pneumococcal strain which had not been observed to mutate spontaneously. In addition, the transformed cells propagated indefinitely as a smooth strain without further treatment with DNA. The pneumococcal DNA preparation had performed 2 functions usually attributed to genes: it had induced a specific heritable property, and had initiated its own reduplication. Subsequently other investigators demonstrated ability of isolated DNA to effect transformations in other species of bacteria, *viz. Hemophilus influenzae, Meningococcus, Escherichia coli,* and the crown-gall organism *Agrobacterium*(2). Benoit *et al.*(3,4) recently have reported that somatic changes are produced in one strain of ducks by injection of DNA derived from another strain, and that the changes were transmitted to progeny of the treated birds. The present paper describes an attempt to produce a similar transformation in mammals. This work was stimulated by the prospect that a successful result might have application to the treatment of genetically determined diseases in man.

*Material and methods. Preparation of DNA.* DNA was isolated from thymuses and spleens of rats by a modication of the method used by Benoit *et al.* (personal communication) to prepare DNA from duck erythrocytes and testicular tissue.† Adult rats were killed, and the spleen and thymus were immediately removed and frozen over dry ice. All further steps in isolation of DNA were carried out in a cold room at 0°C or in a refrigerated centrifuge. The following solutions were used in the preparation of DNA: I. Versene-fluoride: 18.7 g disodium versenate and 1.85 g sodium fluoride in 1000 ml water, adjusted to pH 7.0 ± 0.1. II. Fluoride-chloride: 58.0 g sodium chloride, 1.85 g sodium fluoride, and 8.0 g disodium versenate in 1000 ml water, adjusted to pH 7.0 ± 0.2. III. Versene: 3.75 g disodium versenate in 1000 ml water, adjusted to pH 7.0 ± 0.2. IV. Phosphate buffer, 2.5 M, pH 8.4. V. Sodium chloride solution, 0.85%. 100 g of frozen rat spleen and thymus was minced through a meat grinder and homogenized with 300 ml versene-fluoride solution in a Waring Blender for 5 minutes. The suspension was then centrifuged at 2500 rpm for 30 minutes, the supernatant liquid was discarded, and the sedimented nuclear fragments were collected. Homogenization and centrifugation were repeated as above 5 times. The nuclear fragments then were suspended in 400 ml fluoride-chloride solution and stirred overnight.

* This work supported by Ford Fn.

† This modification was devised by Dr. Matthew Meselson, Calif. Inst. of Tech.