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## Blood Changes of Normal Dogs During Chronic Blood Volume Expansion with Dextran.* (19379)

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Blood changes occurring during long continued administration of dextran have been studied in normal dogs.

*Methods.* Three litter mate mongrel bitches weighing 13-16 kg were fed equal parts dog chow and horse meat in amounts adjusted to maintain body weight. They received intravenously 10, 15 and 20 ml dextran solution (Macrodex†) per kg respectively, 3 times weekly for 13 weeks. This amounted to approximately 300, 500 and 600 g dextran respectively. A control dog from the same litter received 10 ml/kg physiological saline. Preceding the injections 8 ml of blood were withdrawn without stasis and heparinized. Hematocrit values were determined in Wintrobe tubes spun 30 min at 1050 G. Estimations of relative viscosity were made within 20 min of withdrawing the sample in an Ostwald viscosimeter at 3 different pressures. Specific gravities of whole blood and of plasma were measured by the copper sulfate method. Values for the sedimentation of the cells after standing undisturbed for 60 min were obtained within 3 hrs of withdrawing the sample. Hemoglobin was converted to acid hematin and read on the Klett-Summerson photoelectric colorimeter. Mean arterial blood pressures were recorded at weekly intervals by direct puncture of a femoral artery. Estimations of glomerular filtration rate (creatinine), effective renal plasma flow and maximal tubular transfer (p-amino hippurate) were made(1) on 2 dogs receiving dextran and on the control before and after 7 to 12 wks of injections.

*Results.* The repeated injections of dextran were well tolerated. Body weights, after an initial fall during the control period, were maintained with an increase in food intake of 25 g (10-12.5% of the total diet). Mean arterial blood pressures varied within normal limits. Plasma specific gravity was 1.022 to 1.024 in all dogs throughout the experiment. The renal function tests revealed no significant variations from the control. In agreement with the report of Thorsen(2) no significant pathological changes were found by microscopic examination‡ of the tissues of the dog that received 10 ml/kg Macrodex. The tissues of this dog were fixed in aqueous formalin and therefore were not suitable for demonstrating possible dextran deposits.

Average weekly changes in the hematocrit percentage and relative viscosity are shown in Fig. 1, where the mean of the preinjection

TABLE I. Preinjection Mean Values Taken as 100% in Fig. 1 for Hematocrit Volume and Relative Viscosity.

| Subsequent treatment Sol. | Dose, ml/kg | Hematocrit vol, % | Relative viscosity |
|---|---|---|---|
| Saline | 10 | 41.4 | 4 |
| Dextran | 10 | 46.9 | 4.68 |
| | 15 | 48.8 | 4.88 |
| | 20 | 41.3 | 4.01 |

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† Solution used was the Swedish product "Macrodex," 6% dextran in 0.9% NaCl.

‡ We are indebted to Dr. Benjamin Highman of the Laboratory of Pathology and Pharmacology for this examination.

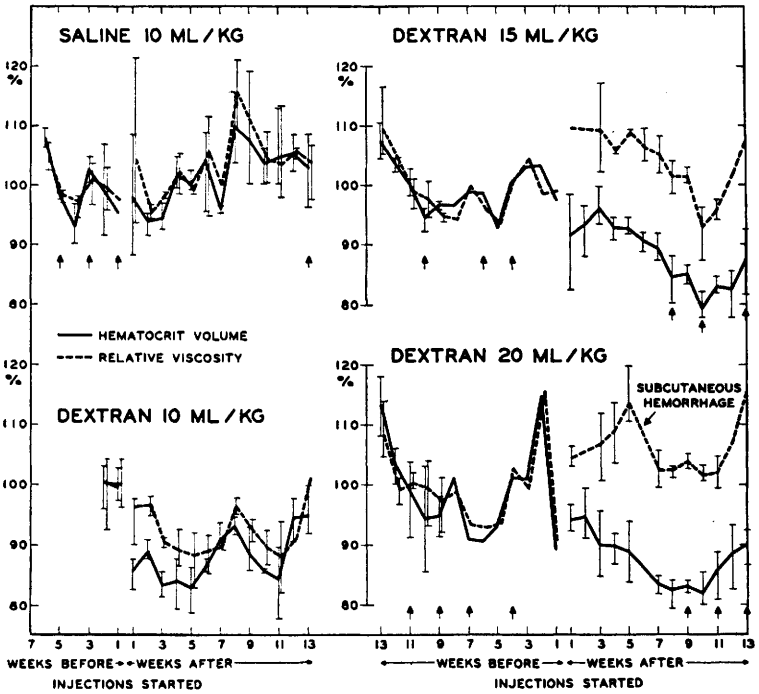


FIG. 1. Comparison of changes in hematocrit volume and relative viscosity in the control dog and in 3 dogs receiving Macrodex in varying amounts. The mean of the preinjection values equals 100%. Each point with bars represents the weekly average of 2 or 3 determinations and their range; absence of bars indicates only 1 determination was made that week. Arrows indicate renal clearance tests. Two additional renal clearances were made on the control dog during the 16th and 19th weeks of injection.

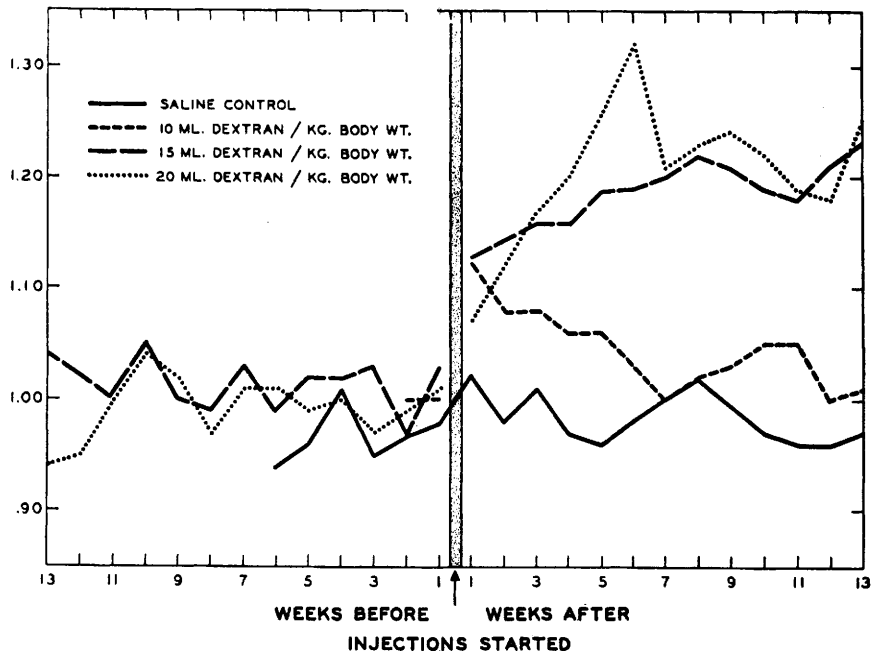


FIG. 2. Ratio of values shown in Fig. 1 for viscosity ($\times$ 10) and hematocrit volumes.

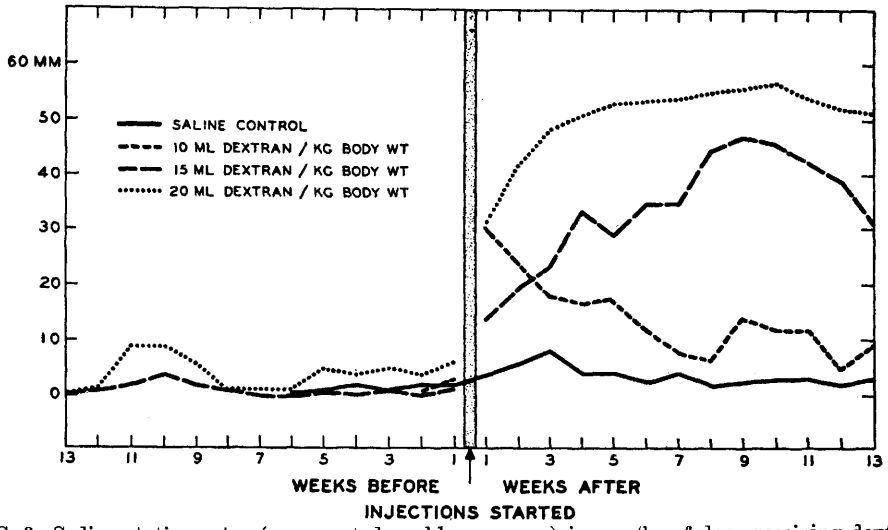


FIG. 3. Sedimentation rates (uncorrected weekly averages) in mm/hr of dogs receiving dextran and the control receiving saline.

values equals 100% (Table I). The extremely high values found in the 20 ml/kg dog for the single blood sample taken the second week before injections began were due to inadvertent dehydration. To facilitate comparison between dogs, use has been made of the ratio of these changes (Fig. 2). The mean weekly sedimentation rates uncorrected for red cell content are shown in Fig. 3. Specific gravity of whole blood and the hemoglobin values followed the hematocrit readings, so there was no change in the ratio of hemoglobin to mean cell volume.

*Discussion.* Whether or not the initial decrease in hematocrit percentage after starting the injections was due wholly to dilution is not indicated satisfactorily by the evidence presented. If dilution were the sole factor, the smallest dosage should have produced the least change in the ratio of cells to plasma. Actually, the largest initial decrease was with this dosage, while the smallest initial change occurred in the dog receiving most dextran. Hematocrit decreases attained subsequent to the initial values were consistent with the dosage administered. Whittaker and Winton (3) have shown the depressing effect upon blood viscosity of a reduction in hematocrit value. In spite of this decrease the relative viscosity of the blood of the 2 dogs receiving the largest injections was increased. This would indicate that injected dextran solution, by virtue of a relative viscosity approaching that of whole blood (Macrodex is slightly more than 3 times more viscous than water; plasma relative viscosity is 1.5)(3), compensates in part for the loss or dilution of erythrocytes while expanding the circulating volume.

The greatest increase in sedimentation rate with the largest injections of dextran confirms previous reports(4). Since the rates are uncorrected for numbers of erythrocytes, these changes are partially due to the hemodilution. To evaluate the importance of this factor, normal dog bloods were diluted with their own plasmas or with Macrodex to hematocrit readings approximating 90, 80 or 70% of the original value. Sedimentation rates in the plasma-diluted bloods averaged 67 to 74% of the rates found in the bloods diluted with Macrodex. It has been shown (5) that the tendency to produce erythrocyte aggregation and hence increase sedimentation is directly dependent on the molecular size of dextran above 60,000. The diminished sedimentation reaction as the injections continued in the dog receiving 10 ml/kg may signify an enhanced capacity to form smaller molecules from the freshly injected material. Evidence of a diminished reaction to repeated injections has been reported in rats(6).

The gradual disappearance of dextran from the blood stream after injection is generally

accepted as not being due to quantitative storage. The recent report(7) of increased glycosuria in phlorizinized starved dogs after dextran administration indicates that metabolism to glucose is one avenue of disposal. There is evidence in mice of transfer to exhaled $CO_2$(8). Partial degradation to smaller molecules capable of passing the renal glomerulus constitutes another possibility.

*Summary.* Except for a decrease in hematocrit volume, intravenous administration of dextran continued for 13 weeks produced in 3 normal, nonshocked dogs no demonstrable harmful effects. Blood viscosity was maintained at or above control levels when the injections were 15 to 20 ml/kg body weight. The dog receiving 10 ml/kg exhibited a tendency to adjust to the foreign polysaccharide as evidenced by a return towards normal hematocrit values and sedimentation rates in spite of continued dextran.

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## Sarcomas Induced in Rodents by Imbedding Various Plastic Films.* (19380)

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In 1948 we reported that sarcomas of various types could be induced in albino rats in a significant percentage of cases by imbedding cellophane film subcutaneously in the anterior abdominal wall, or by wrapping it around one kidney(1). Since then the experiments have been extended by imbedding cellophane film in albino and black mice, by testing alcohol-extracted cellophane, and by employing various other plastic films unrelated chemically to cellophane. In addition several other materials were imbedded in an effort to learn the nature of the carcinogenic agent or mechanism involved in the procedure. The results of these new experiments are herewith reported.

*Method.* The method previously described (1) was used, except that the film or other material was always inserted in the anterior abdominal wall, and no further experiments were attempted with wrapping the film around the kidney. The experiments were conducted over a period of 4 years. Some overlapped. The food cages, handling and room temperature were essentially the same. The rats were all albinos, of the Wistar strain. Films were usually sterilized by immersion in 80% alcohol and then washed in sterile saline solution before imbedding. Occasionally sterilization of the plastic film was accomplished by autoclaving, or by benzalkonium chloride ("Zephiran") 1:1000 or weaker. As malignant tumors arose after any one of these methods of sterilization, it is improbable that the particular technic of sterilization played a role in the result. The malignancy of the tumors produced was established by histologic examination and usually also by transplantability, as previously described(1). Experiments lasted 1-2 years. In calculating the percentage of positive results, *i.e.,* production of tumors, the figures were based on the number of animals that survived the minimum period neces-

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