plasma pipetted off and diluted 1:5 with specially prepared, particle free, 0.75% physiological saline solution, and examined under the ultramicroscope. Similar samples were received into heparin (1/3 mgm. per cc.), potassium oxalate (2 mgm. per cc.) and sodium citrate (2.5 mgm. per cc.). The blood was also made non-coagulable by injection of heparin into the dorsal lymph sac 45 minutes previous to bleeding. The appearance of the various samples of plasma was then compared under the ultramicroscope with the normal plasma obtained without the use of any anticoagulant. Quantitative readings were also made of the relative refractiveness of the various plasmas, employing the photometer described by Hirschfelder and Wright.[3]

The use of heparin either *in vivo* or *in vitro* produced only small changes in the appearance of the plasma. With potassium oxalate the particles appeared smaller, greater in number, and of increased refractiveness. Sodium citrate brought about a very marked reduction in the apparent size of the particles, with corresponding increase in the number of particles visible, and increase in refractiveness. This change in the colloidal equilibrium of the plasma proteins with addition of sodium citrate may be of importance in connection with the use of sodium citrate as an anticoagulant in blood transfusions.

Similar experiments on mammalian and human plasma are in progress and will be reported at a later date.

4850

Effect of Novasurol Upon the Ultramicroscopic Appearance of Frog's Plasma.

RAYMOND N. BIETER AND HAROLD N. WRIGHT.
(Introduced by Arthur D. Hirschfelder.)

From the Department of Pharmacology, University of Minnesota.

In a previous paper it has been shown by Bieter[1] that dilute solutions of mercuric chloride injected via the ureter in frog's kidneys, inhibit the reabsorption of water and phenolsulphonephthalein. Unpublished observations in this laboratory indicate that this action might also be shared by novasurol.

[3] Hirschfelder, A. D., and Wright, H. N., PROC. SOC. EXP. BIOL. AND MED., 1930, xxvii, 547.

[1] Bieter, Raymond N., *Am. J. Physiol.*, 1930, in press.

Attempting to obtain further information upon the fundamental action of novasurol as a diuretic, the effects of this drug *in vitro* and *in vivo* have been studied on the appearance of frog's plasma under the ultramicroscope. The technique of the ultramicroscope studies used has been that of Wright and Hirschfelder.[2] The novasurol in 0.1% concentration was made up in 0.75% NaCl. This gives 0.01 mgm. per 0.01 cc. To 1 cc. of frog's plasma diluted with 4 cc. of 0.75% NaCl solution was then added 0.04 mgm. of novasurol.

This amount of novasurol when added to a sample of frog's plasma, which was collected without the use of an anticoagulant, produced the following changes in particles as compared to those described in the preceding paper by Wright and Bieter.[3] The number of particles was decreased, their average size was increased, and their refractiveness, as measured with the photometer of Hirschfelder and Wright,[4] was increased.

When the same amount of novasurol was added to a sample of frog's plasma prepared as above, but where coagulation was prevented by the use of heparin (10 mgm. via lymph sac or 1 mgm. in 0.1 cc. saline placed in the tube used to collect the blood) the picture observed under the ultramicroscope was essentially the same.

The action of novasurol on citrated and oxalated plasmas has again progressed in the same general direction. That is to say, the number of particles was decreased, their average size was increased and their refractiveness as measured with the photometer, was increased. The differences, however, between citrated or oxalated plasmas alone, as compared with the same to which novasurol was added (as above) was not as marked as between heparinized plasma and heparinized plasma plus novasurol.

It is interesting to note further that these effects come on slowly, requiring from 3 to 6 hours for the full effect of the novasurol. This time element compares favorably with the time required clinically for the optimum action of the drug.

Similar experiments on mammalian and human plasmas are in progress and will be reported at a later date.

---

[2] Wright, H. N., and Hirschfelder, A. D., PROC. SOC. EXP. BIOL. AND MED., 1929, xxvi, 790.

[3] Wright, H. N., and Bieter, R. N., *Ibid.*, 1930, xxvii, 550.

[4] Hirschfelder, A. D., and Wright, H. N., *Ibid.*, 1930, xxvii, 547.