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Action of Urea, Indican, and Phenol on Red Cell Hemolysis.

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In chronic glomerulonephritis usually there is a progressive normocytic anemia which becomes manifest as renal insufficiency progresses and increases with the elevation of nitrogen retention.[1] There is, furthermore, no apparent lack of active blood-forming tissue, nor has there been any evidence presented to suggest that blood loss or increased hemolysis may play a rôle.[1, 2]

With the progress of renal insufficiency in glomerulonephritis there is an accretion of various chemical constituents in the blood plasma. The increase of certain of these, namely, urea and indican, is generally accepted, and certain studies on the blood phenol indicate that this substance likewise is increased.[3] It occurred to us that these retained substances in particular may play a rôle in the anemia of nephritis, and that they may cause hemolysis of such a degree as is not detectable by the ordinary tests usually employed for the determination of increased fragility.

Herrald and Pijoan[4] investigating the behavior of erythrocytes in a hemolytic system using standardized saponin as the lytic agent, demonstrated that the red cells of nephritic patients are more resistant than those of normal subjects to saponin hemolysis. However, they were unable to offer any adequate explanation for this phenomenon. Suspecting that indican, phenol and urea might play a rôle in hemolysis, we decided to investigate this problem using the method described by Herrald and Pijoan. Our plan was to compare the saponin hemolysis curves of the red cells of uremic patients with red cells from normal subjects, and then to add proportionally the same amounts in their equivalent proportion of urea, indican and phenol to red cells of normal subjects with and without saponin.

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2 Brown, G. E., and Roth, G. M., *Arch. Int. Med.*, 1922, **30**, 819.

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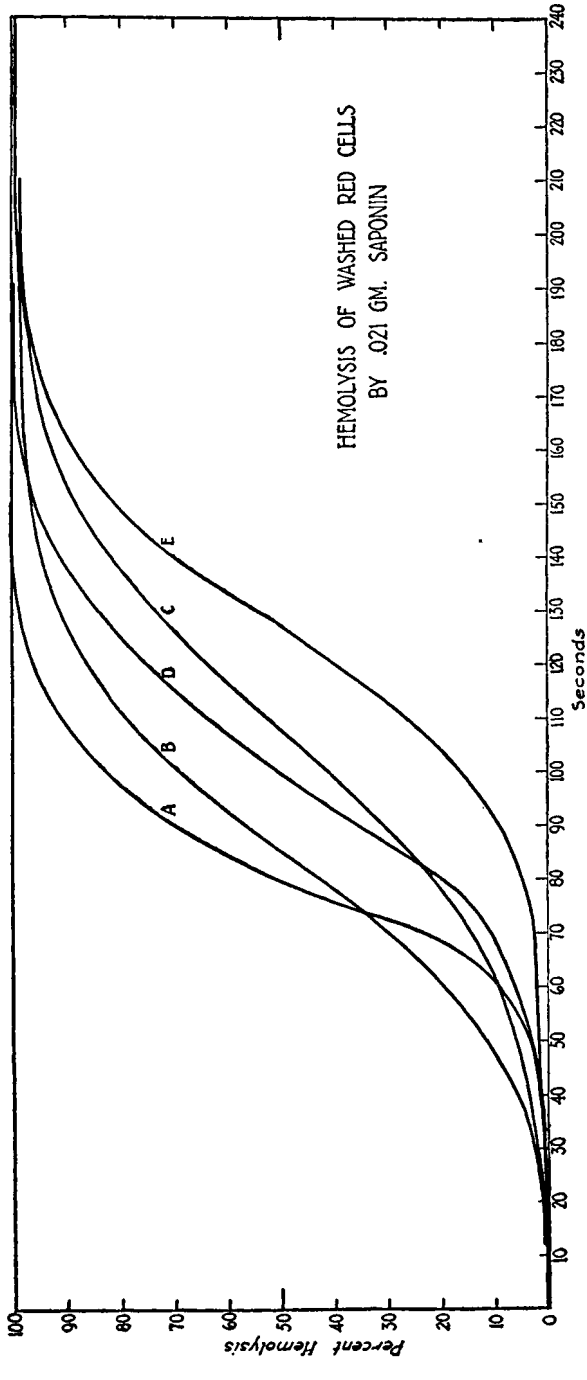

FIG. 1.

A. Hemolysis of washed normal red cells.
B. Hemolysis of washed red cells from a patient with chronic glomerulonephritis in uremia (Patient 1).
C. Hemolysis of washed red cells from a patient with chronic glomerulonephritis in uremia (Patient 2).
D. Addition to hemolytic system of normal washed red cells of equivalents in blood of 60 mg. % urea, 3 mg. % indican and 5 mg. % phenol.
E. Addition to hemolytic system of normal washed red cells of equivalents in blood of 120 mg. % urea, 3 mg. % indican and 5 mg. % phenol.

Red cells from uremic patients with the exception of those in the extreme terminal stage were found to hemolyze more slowly in saponin systems than normal cells, which is in agreement with the previous study.[4] In this investigation certain quantities of indican, urea and phenol were added individually and collectively to a suspension of known amounts of washed normal red cells in saline and were found to have no hemolytic activity whatsoever in their proportion found in the plasma. Furthermore, as can be seen from Fig. 1, urea, phenol and indican, added to systems of red cells in plasma, definitely inhibited saponin hemolysis and reproduced curves which could be compared to those obtained by Herrald and Pijoan for nephritic blood.

In light of these test tube experiments we are led to the belief that in the anemia of nephritis, which develops with the manifestation of renal insufficiency, there is no increased hemolysis of the red cells by urea, indican and phenol, and that these substances inhibit the action of a standard hemolysin.

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Effect of Urea Upon Growth of Fibroblasts from Cardiac Explants in Tissue Culture.

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It has been found recently that allantoin, a purine derivative occurring in maggots' secretions, stimulated the healing process in deep purulent wounds.[1] Another derivative, urea, was discovered to be more effective in bringing about the same results.[2] It was thought that, since urea may be obtained from allantoin by hydrolysis, the growth promoting effect of allantoin might be due to the urea and not to allantoin itself.

Fibroblasts from cardiac explants in tissue culture grew more abundantly in the presence of 0.5% allantoin than those in control cultures; although the growth increase was not sufficiently great to be of real significance.[3] It was deemed of interest to discover

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