action to estrogen assay in a material which contains several estrogens must entail prior separation of the different components. Several recommended fractionation technics fortunately meet this requirement.[6,7,18-20] It may be noted here that the nonsteroid estrogen, stilbestrol, does not produce fluorescence under the conditions of the test.

The fluorimetric method is simple, the reproducibility of the result is good, and the specificity of the test promises to be as good as or better than has been obtained with the previously described colorimetric methods. Further study of the analytical possibilities of this assay seems therefore to be desirable. A survey of the effect of phosphoric acid on a larger number of sterols and related substances with a view to elucitating the relation of sterol structure to fluorescent response has accordingly been undertaken. At the same time the application of the fluorimetric method to estrogen determination in blood and urine is being examined. The results will be reported elsewhere.

Summary. 1. A method for estimation of estrone, estradiol, and estriol in pure solution is based on the observation that these substances afford products with a green fluorescence when heated in phosphoric acid.

2. The fluorescent response of the urinary estrogens is over a wide range a linear function of their concentration. The error of the fluorimetric assay in suitable ranges of estrogen concentrations (estradiol, 0.5-2.5 γ; estrone, 1.0-5.0 γ; and estriol, 1.0-10.0 γ) does not exceed ±3%.

3. Sterols which do not contain a conjugated double bond, some naturally occurring nonsteroid aromatic substances, and many common constituents of biological material give no fluorescent response under the conditions of the test.

4. Estrogen fluorimetry appears to compare favorably as to sensitivity, specificity and simplicity with previously described methods of estrogen assay.

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Induction of Renal Glomerular Lesions by Urethane in Inbred Mice Susceptible to Spontaneous Glomerulonephritis.*

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Mice of the NH strain develop spontaneous glomerulonephritis. Histologically the glomeruli are similar to those of the human chronic disease.[1]

Dunn and Larsen[2] reported that urethane induced hyaline alterations in the glomeruli of mice of the Strong A strain, whereas similar changes were lacking in the glomeruli of other stocks of mice following the administration of this drug.

The work of Dunn and Larsen suggested to the authors that urethane might provoke interesting changes in the glomeruli of mice which are susceptible to the development of spontaneous glomerulonephritis. Twenty-one NH mice received 1 mg per g of body weight of urethane (anesthetic dose) in

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aqueous solution once each week by subcutaneous injection. The mice were 6 to 8 weeks of age when the experiment was begun.

Between 5 and 6 months after the series of injections was started 11 of the mice became grossly edematous. The plasma protein levels of these animals were depressed (values of less than 5.2 g%,[3] the lowest level obtained for the mice without edema), the urine contained albumin, and blood urea nitrogen was elevated (range of from 50 to 265 mg% as compared with a normal of 18-27 mg%).[4] In one case there was a gross hyperlipemia; the plasma cholesterol value was 430 mg% as compared with a normal for mice of approximately 100 mg%.[5] All the injected mice were anemic. The degree of nitrogen retention, but not of anemia, could be correlated with the extent of the pathologic alterations observed in the glomeruli.

The essential pathologic changes in the glomeruli were thickening of the capillary basement membrane (azo-carmine stain), capillary thrombosis, and closure of many capillary tufts resulting from the basement membrane alteration. The affected glomeruli were smaller than those observed in spontaneous glomerulonephritis of NH mice, more glomeruli were normal, and except in an occasional glomerulus there was nothing to suggest endothelial proliferation. Histologically the glomeruli resembled those of human lipoid nephrosis.[6] In one case most of the glomeruli were extensively infiltrated with polymorphonuclear leukocytes.

Of the 21 mice injected with urethane, either edema or histologic evidence of renal disease and nitrogen retention were observed in 17. One animal was not nephritic as judged by gross, histologic and biochemical evidence; 3 were without renal disease grossly and the kidneys were not studied histologically because of postmortem degeneration.

Spontaneous glomerulonephritis appears rarely before the age of 8 months in NH mice. Of 53 cases in a total of 199 controls only 5 appeared before 8 months of age. There is no doubt that the glomerular lesions were induced by urethane; the effect of urethane on the kidney was confined to the glomerulus.

Urethane is a capillary poison.[7-9] In NH mice the glomerular capillaries especially seem to be susceptible to its effects. The drug also induces lung tumor development in mice,[10] and most of the animals of this experiment developed multiple lung adenomas. It is likely that the anemia was not entirely the result of the renal disease. Urethane depresses the myeloid tissues of mice,[11] and, furthermore, the injected animals failed to gain in weight to the same extent as normal mice. These animals might belong in the same category as underfed mice, which are characteristically anemic.

Summary. Urethane induced glomerular lesions in NH mice, an inbred strain which is susceptible to the spontaneous development of glomerulonephritis. Mice with the urethane-induced glomerular alterations exhibited renal insufficiency, albuminuria, edema, and depressed plasma protein levels. One case evidenced hypercholesterolemia.

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