

Menkin⁵ has shown that following repeated injections of a dilute solution of ferric chloride in rabbits the iron is deposited in large amounts within and about the tuberculous lesions and its presence tends to favor fibrosis and prolong the life of the animal.

Menkin administered 3 injections a week intravenously each of 6 cc. of a 0.25% solution of ferric chloride, during a period of 4 months. This represents a total amount of approximately 220 to 250 mg. of elemental iron for the entire experimental period. Though this is approximately the same quantity as that used in the experiments here reported, it is probable that the differences in the animals used may account for the difference in the results.†

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Relation of Cholane Nucleus to the Female Bitterling Test for Male Hormone.*

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After demonstrating that the female bitterling test was not a test for pregnancy,¹ we have stated² that the fraction of male urine containing the male hormones is responsible for the ovipositor elongating reaction. A positive reaction was taken as a lengthening of the ovipositor from a quiescent stage to a length equal to that of the anterior edge of the anal fin, *i. e.*, the ovipositor must reach the end of the fin.

Crystalline theelin and theelol were also tested and although they

⁵ Menkin, Valy, *J. Exp. Med.*, 1932, **55**, 101; 1934, **60**, 463.

† The literature⁶⁻¹⁰ on the therapeutic use of copper is reviewed in a previous article.³

⁶ Bevan, cited by Corper, H. L., DeWitt, L., and Wells, H. G., *J. Am. Med. Assn.*, 1913, **60**, 887.

⁷ Von Linden, G., *Beitr. Klin. Tuberk.*, 1912, **23**, 201.

⁸ Meissen, E., *Beitr. Klin. Tuberk.*, 1912, **23**, 215.

⁹ Strauss, A., *Beitr. Klin. Tuberk.*, 1912, **23**, 223.

¹⁰ Corper, H. J., *J. Infect. Dis.*, 1914, **15**, 518.

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¹ Kleiner, I. S., Weisman, A. I., and Barowsky, H., *J.A.M.A.*, 1935, **104**, 1318.

² Kleiner, I. S., Weisman, A. I., and Mishkind, D. I., *J.A.M.A.*, in press.

did not give positive reactions, they sometimes caused a slight lengthening of the ovipositor. We therefore felt it important to determine whether any other substance containing the cholane nucleus would produce this reaction in greater or lesser degree.

We have tested ergosterol† and cholesterol emulsions and solutions of sodium taurocholate according to our method.² Small doses of all these compounds produced no reactions. These doses were equivalent to, or several times greater than the doses of male hormone which produced pronounced reactions. Larger amounts of cholesterol, however, produced slight effects while the same amounts of sodium taurocholate were fairly active. For example, while approximately 1.7 mg. of the crude male hormone fraction produced a marked positive reaction when added to 4 liters of water containing 2 bitterlings, 40 mg. of cholesterol caused only slight effects. Doses of 8 mg. of sodium taurocholate caused no reaction, while 25 to 50 mg. gave a moderately strong reaction. Thirty animals were used. It is highly improbable that these weak reactions were due to contaminations of our "C.P." preparations with male hormone. This could be excluded only by the use of synthetic products.

Since bile salts occur in urine in appreciable amounts only in cases of obstructive jaundice, it is evident that ordinarily this factor would not interfere with the test. Moreover, it is doubtful whether even in extreme icterus sufficient amounts of bile acids are excreted to cause any reaction.

It appears that the cholane nucleus, *per se*, is probably not the sole causative factor in the production of the female bitterling (ovipositor) reaction.

† The ergosterol was generously supplied by Mead Johnson and Co.