

monal effects. Being benign these tumors retain the capacity to respond to the hormones which affect the normal mammary gland.

Summary. The transplantability and rate of growth of a pure mammary fibroma were markedly enhanced by the presence of physiological levels of estrogens in the host rats. Physiological levels of androgens, however, had no demonstrable effect on this tumor. The tumor-stimulating effect of estrogen injections was greater in male castrates than in female castrates. With the onset of malignancy the connective tissue tumors lose their responsiveness to hormonal influences.

11169 P

Destructive Effects on Carcinoma of Colchicine Followed by Distilled Water.*

M. F. GUYER AND P. E. CLAUS.

From the Department of Zoology, University of Wisconsin, Madison.

While studying the enhanced effects of X-rays on transplanted Flexner-Jobling carcinoma following injections of colchicine into the host (paper in press), distilled water injected into the tumors some 15 hours after administration of the drug was used as a control for comparison with the effects of the irradiation. The results are summarized in Table I. Since colchicine alone has a retardative but only occasionally a lethal effect on some tumors¹ it is evident from the tabulation that the subsequent treatment with distilled water was unexpectedly destructive. F.-J. tumors injected with distilled water alone, in 47 rats, showed approximately the same percentage of recoveries as the spontaneous regressions and recoveries in our present strain of rats when the implants are left to grow for 3 weeks and only those regarded as "takes" which have increased to at least 1 cm in diameter; namely, between 3 and 4% (6 of 157 rats). Such 3-week growths were the only kind used in the present experi-

* This investigation was aided by support from the Brittingham Trust Fund and a Research Grant from Elizabeth Hopkins Johnson.

¹ Ludford, R. J., *Arch. f. exp. Zellforsch.*, 1936, **18**, 411; Clearkin, P. A., *J. Path. and Bact.*, 1937, **44**, 469; Lits, F. J., Kirschbaum, A., and Strong, L. C., *Am. J. Cancer*, 1938, **34**, 196; Guyer, M. F., and Claus, P. E., *Proc. Soc. Exp. Biol. and Med.*, 1939, **42**, 565.

TABLE I.

Experiments with Colchicine and Distilled Water.

Tumors all actively growing and ranging in diameter from 1 cm to 2.5 cm; colchicine dosage, 0.1 mg per 100 g of body weight; distilled water injected into tumors 15 hours after subcutaneous injection of colchicine.

Series	No. of rats	Time, end of	Healed	Dead	Unhealed
22	13	7th wk	10	3	0
23	15	12th wk	8	6	1
24	17	10th wk	10	3	4
27	58	8th wk	31	14	13

ments. If one takes beginning growths at the end of only 2 weeks as a criterion, then the spontaneous recovery rate is about 8% (actually 24 of 312 rats).

Rats bearing the rapidly growing cancers were injected subcutaneously with 0.1 mg of colchicine per 100 g of body weight. Fifteen hours later distilled water was injected directly into each tumor in as great quantity as it would hold. By the end of a week the surfaces of most of the growths had sloughed off and many of the cancers appeared to be healing. Some healed entirely after one treatment. Hosts bearing tumors which showed evidence of renewed growth at the end of 2 weeks were treated again as at first. This was repeated at 2-week intervals in all cases showing recurrence of tumor growth. In the 4 series of experiments (see Table I) a total of 103 cancer-bearing animals were used. Complete healing was secured in 59 cases (57.3%). At the conclusion of the experiments 18 tumors (17.5%) remained unhealed and showed no evidence of healing. Of the 26 animals which died, death in several cases, since it followed the water treatment within a few minutes, was due presumably to embolism resulting from injection of the tumors under sufficient pressure to force necrotic tissue into the blood stream. Inasmuch as the cancers were not advanced far enough to be lethal, the remaining deaths, lacking other explanation, were attributed to the unfavorable, warm-weather conditions which prevailed during the course of the experiments.

Just what happens to cancer cells under such treatment is not clear. That it is not merely a matter of pressure is indicated by the fact that similar tumors in rats not given a preliminary treatment of colchicine show no such degree of regression and disappearance following like injections of distilled water. The general histological and cytological picture seen in sections of carcinomas taken from rats 15 to 18 hours after subcutaneous injections of colchicine is strikingly like that observed in similar cancer cells following irradiation. The chromatin of the nuclear contents often becomes clumped into dense

masses which stain deeply with basic dyes. This may mean increased acidity and consequently an increased hydrophylic tendency on the part of the cell, particularly the nucleus. It is probable, however, that the cytoplasm is also involved inasmuch as certain of its constituents, namely the Golgi apparatus and the mitochondria are markedly modified within 5 hours after administration of the drug. Under such treatment the Golgi apparatus which ordinarily occurs in the carcinoma cell as a basket-like or looser thread-like mass at one side of the nucleus is dispersed into a cloud of fine droplets, and the usually thread-like mitochondria break up into small rods or granules. Whether the plasma membrane of the cell becomes more permeable to water is not yet known.

There is even noticeable change after some hours in the gross appearance of carcinomas in rats which have been given as high a dosage of colchicine as 0.2 mg per 100 g of body weight. The usual white, waxy appearance of the tumor is replaced by a decidedly reddish color, indicating considerable extravasation of blood. Experiments with various aqueous agents and with other types of tumor are in progress.

Summary. Following administration of colchicine to their hosts 15 hours earlier, Flexner-Jobling carcinomas were found in a majority of 103 cases to be rapidly destroyed by distilled water injected directly into their substance.

11170

A Note upon the Micro-Determination of Ammonia.*

M. F. MASON AND J. D. ROZZELL. (Introduced by C. S. Robinson.)

From the Department of Biochemistry, Vanderbilt University School of Medicine, Nashville.

Several excellent procedures are available for the micro-estimation of ammonia of which the methods of Conway and Byrne¹ and Gibbs and Kirk² are notable examples. In connection with studies of tissue respiration it became desirable to determine quantities of ammonia of the order of 5 γ to 50 γ present in the Ringer fluid in which various

* Aided by a grant to the Vanderbilt University School of Medicine from the Division of Medical Sciences of the Rockefeller Foundation.

¹ Conway, E. J., and Byrne, A., *Biochem. J.*, 1933, **27**, 419.

² Gibbs, G. E., and Kirk, P. L., *Mikrochemie*, 1935, **16**, 25.