demonstrated increased contractions in strips of guinea pig ileum exposed to their patient's serum, but not to a saline extract of the patient's tumor, to normal human serum or to an endometrial extract. No concentration range was reported as having been explored for the substances which had no effect on their preparation.

Summary. Clinical observations in a patient with a non-Beta islet cell pancreatic adenocarcinoma demonstrated excessive fecal losses of fluid and electrolytes and excessive volumes of fluid from the distal ileum during continuous gastric aspiration. Intestinal transit time was normal. *In vitro* studies demonstrated the capacity of a distilled-water extract of an hepatic metastasis to inhibit the net mucosal-to-serosal transfer of fluid, sodium and chloride across everted gut sacs from the distal portion of the hamster small intestine. No effect of the tumor extract on *in vitro* intestinal motility was observed. These results suggest that the patient's diarrhea resulted from the tumor or its products inhibiting the net absorption of fluid and electrolytes in the patient's small intestine.

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Received June 17, 1966. P.S.E.B.M., 1966, v123.

Antiprolactin and Experimentally-Induced Hepatic Metastases.* (31489)

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Hypophysectomy has been previously observed in our laboratory to inhibit the development of artificially-induced hepatic metastases following intraportal injection of known numbers of Walker tumor cells in female Sprague-Dawley rats(1). This effect was markedly minimized in hypophysectomized rats by intraperitoneal injection of ovine prolactin but not somatotropin, thyrotropin, adrenocorticotropin or luteinizing and follicle stimulating hormones administered either singly or in combination(2). This information, as well as other experimental (3,4) and clinical(5,6) evidence suggesting that prolactin may play a role in the induction and dependence of some mammary tumors prompted us to investigate the effect of a physiologically active antiprolactin preparation on the growth of hepatic metastases in our model system.

Materials and methods. Twelve adult white rabbits received subcutaneous and intraperitoneal injections of 8 mg of ovine prolactin† suspended in Freund's complete adjuvant weekly for 3 weeks. An intravenous booster of 0.25 mg prolactin in saline was administered 3 weeks later. Hemagglutination titers determined according to the method of Levy and Sampliner(7) and agar-gel diffusion were performed on blood samples obtained prior to immunization and 10 days following the booster dose. Only sera from animals exhibiting a titer greater than 1:600 and distinct precipitin bands (Fig. 1) were utilized in the experiments. Antiprolactin (APS) and normal rabbit sera (NRS) were dispensed in small sterile containers and kept frozen until used.

* Supported by USPHS Grants CA-05716, CA-06670, HE-07822 and Am. Cancer Soc. Grant P-142.

† Endocrinology Study Section, Nat. Inst. Health.

TABLE I. Incidence and Size of Hepatic Metastases and Pituitary and Adrenal Weights of Intact and Hypophysectomized Rats Treated with Prolactin and/or Antiprolactin.

Group	#	Metastases % +	Metastases % 2+	Pituitary (mg)	Adrenal (mg)
Intact					
A. .5 ml NRS*	31	48	20	10.9	71 ± 9§
B. 1.0 ml NRS	15	47	15	11.1	65 ± 8
C. 25 mg prolactin	32	53	20	11.0	66 ± 9
D. .5 ml APS†	32	59	20	10.9	70 ±10
E. 1.0 ml APS	15	50	20	10.5	70 ± 9
F. Untreated	19	45	15	11.2	65 ± 8
G. " ‡	29	31	0	10.6	65 ±12
Hypophysectomy					
H. NRS	20	10	0		20 ± 4
I. .25 prolactin	18	33	17		16 ± 7
J. .25 prolactin + NRS	23	25	13		19 ± 3
K. .25 prolactin + APS	23	13	0		18 ± 3

* Normal rabbit serum. † Antiprolactin serum. ‡ Pair-fed to H. § Standard deviation.

Adult female Sprague-Dawley rats weighing 150-200 g were utilized in all experiments. These were housed in individual cages and allowed water and laboratory chow *ad libitum* unless otherwise indicated. As noted in Table I, groups of intact rats received intraperitoneal injections of either 0.5 ml of NRS, APS, or 0.25 mg prolactin dissolved in saline, for 1 week prior to and 2 weeks following tumor cell injection. NRS and APS were administered in the morning and prolactin in the afternoon. Preliminary investigation revealed this schedule of APS administration to result in moderate, but distinct, ductal and lobuloalveolar atrophy of the breast in intact, adult female Sprague-Dawley rats. Normal intact rats also received 1 ml of either APS or NRS. Hypophysectomized rats,‡ received in the laboratory 2 days following operation, were maintained as described previously(1) and groups were subjected to the same treatments as intact animals, except that some received both prolactin and APS. Normal intact rats fed *ad lib* or pair-fed to hypophysectomized rats receiving injections of NRS also were subjected to tumor cell injection.

Walker tumors utilized in this study have been propagated in our laboratory for the past 8 years. Each animal received an intraportal injection of 5000 tumor cells in 0.5 ml saline according to previously described methods(1).

Animals were sacrificed by exsanguination 2 weeks following tumor cell injection. The incidence, as well as size of hepatic and other visceral tumors, was estimated. Portions of tumor were fixed in formalin and processed for light microscopy in the conventional manner. Inguinal fat pads were fixed in Bouin's fluid and processed for the preparation of total amounts of breast tissue as described by Cowdry(8). Adrenals and pituitaries of intact rats were weighed. Only hypophysectomized rats without evident pituitary remnants were considered in the analyses.

Results. As indicated in Table I, the administration of prolactin to hypophysec-

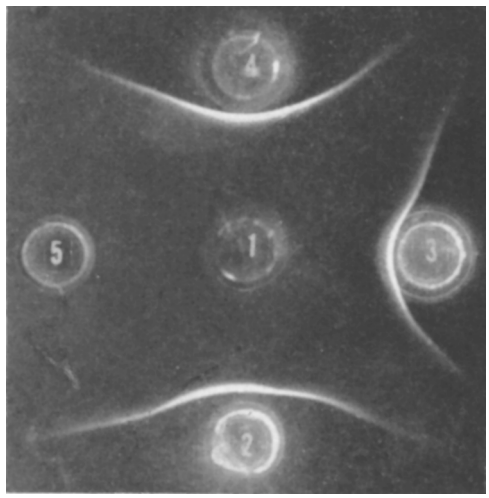

FIG. 1. Precipitin bands following double diffusion in agar gel of prolactin (1) and pooled antisera (2,3,4). No reaction is present with pooled sera obtained prior to immunization(5).

‡ Hormone Assay Laboratories, Inc., Chicago, Ill.

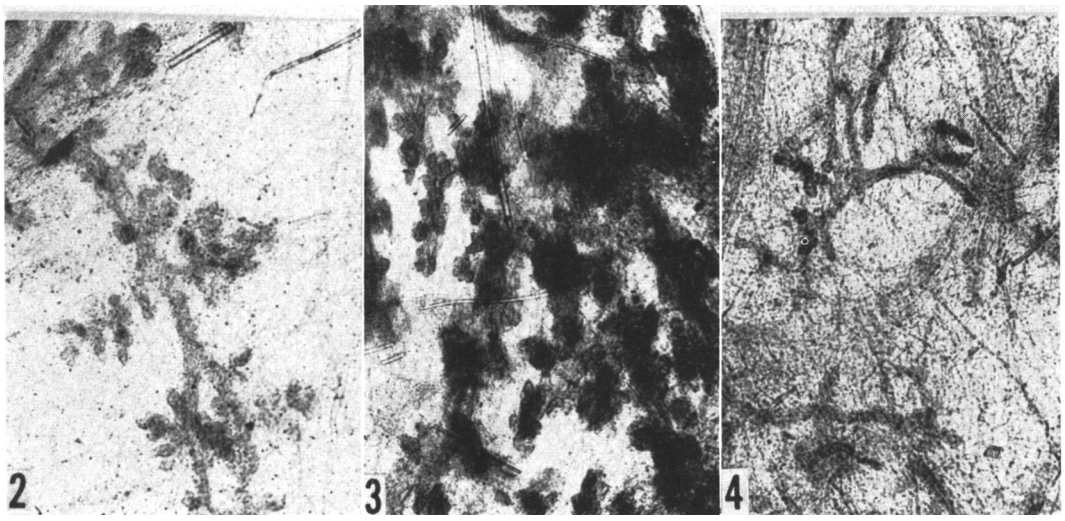

FIG. 2. Appearance of mammary ducts and lobules of intact rat treated with normal rabbit serum. × 25.

FIG. 3. Mammary lobule-alveolar hyperplasia and widening of ducts in prolactin-treated. × 25.

FIG. 4. Narrowing of mammary ducts and atrophy of lobules in rat treated with antiprolactin. × 25.

tomized rats receiving an intraportal injection of Walker tumor cells resulted in an approximately 3-fold increase in incidence and larger size of hepatic metastases than was observed in other hypophysectomized rats not receiving this hormone. This incidence of hepatic metastases was comparable to that observed in pair-fed intact controls. On the other hand, prolactin failed to exhibit any effect on tumor behavior in intact normal rats, although the dose employed was sufficient to induce lobulo-alveolar mammary hyperplasia (Fig. 2, 3). Similarly, APS failed to influence hepatic metastases in intact rats, although the dosage utilized was sufficient and in some animals greater than that required to induce ductal and lobulo-alveolar mammary atrophy (Fig. 4). On the other hand, hypophysectomized rats treated with both prolactin and APS exhibited fewer metastases than rats treated with prolactin and NRS or prolactin only. Mammary tissue from hypophysectomized rats receiving both APS and prolactin exhibited atrophic changes comparable to those in hypophysectomized members receiving NRS.

Administration of APS, NRS or prolactin failed to influence pituitary or adrenal weights of intact rats and the latter of hypophysectomized animals (Table I). None of the modalities influenced the histologic appearance of the Walker tumor or incidence of metastases in other visceral sites; these latter being less than 15% in all situations studied.

Comment. These results confirm our previous observations that prolactin increases the incidence and size of hepatic metastases following intraportal injection of Walker tumor cells in hypophysectomized, but not intact rats(2). The mechanism of this effect in the former, and the reasons for its lack of influence in intact animals, is not clear, but it does not appear related to prolactin dependence of the Walker tumor or other endocrine influences mediated by this hormone, although the Walker tumor was originally considered to be derived from mammary gland(2). No consistent relationship between the occurrence of metastases and lobulo-alveolar mammary hyperplasia induced by prolactin was evident in this and the previous study(2). Dao(4) has also failed to note any consistent relationship between mammary changes and the occurence of breast cancer in rats with isografts of mammotropin-producing pituitary tumors. A similar dichotomy between mammary response to antiprolactin, characterized by lobulo-alveolar atrophy, and

tumor growth was noted in this study. The failure of antiprolactin to influence metastases may represent a quantitative problem. This view is suggested by noting a lesser incidence of metastases in hypophysectomized rats treated with both prolactin and antiprolactin than in those receiving only the former. It seems unlikely that this effect resulted from a neutralization of prolactin at the injection site by antiprolactin since these agents were administered at different time intervals. It is apparent that an effective tumor-inhibiting dose of antiprolactin in intact rats would be at least greater than twice the dose necessary to induce mammary atrophy.

Summary. Neither prolactin nor antiprolactin affected the incidence or size of hepatic metastases in intact rats following the intraportal injection of Walker tumor cells. These failures were unrelated to the mammary changes induced by this hormone or its antibody. On the other hand, antiprolactin moderately inhibited the tumor growth-promoting effect of prolactin in hypophysectomized rats. This suggests that one factor concerned with the failure noted in intact members may be related to the quantity of antibody required to inhibit tumor growth when endogenous prolactin secretion is maintained.

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Received June 22, 1966. P.S.E.B.M., 1966, v123.

Effect of Tetracycline on Intestinal Absorption of Various Nutrients By the Rat.* (31490)

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Tetracycline administered to rats has been found to decrease incorporation of amino acids into the proteins of many tissues including those of the gastrointestinal tract(1). Other inhibitors of protein synthesis such as puromycin and acetoxycycloheximide(2) have been reported to decrease fat absorption in rats. This study was undertaken in order to determine the effect of tetracycline on the absorption of fat, fatty acid, and the water-soluble nutrients, ferrous ion, D-xylose, and vitamin B_{12}.

Materials and methods. Adult rats were given tetracycline HCl (TC) i.m. at 250 mg/kg body weight in 2 injections at an interval of 16 hours. Controls received an equal volume of saline adjusted to pH 2 with N/10 HCl. A single dose of each test substance (in the amount and solvent stated for each experiment) was given by stomach tube 2 hours after the second TC injection. The experiment was terminated at the time indicated in the pertinent table by ether anesthesia with blood obtained by cardiac puncture. Various organs were removed and dissolved in aqueous 30% KOH. Unless stated otherwise, the entire G.I. tract was removed and dissolved with its luminal contents. Radioactivity was measured in a well-type scintillation counter and xylose by a colorimetric method(3). When ^{131}I-con-

* This investigation was supported by Grant AM 06588 from Nat. Inst. of Arthritis & Metab. Dis., USPHS.