never functioned in the CB birds, one would be forced to conclude that other sites in the chicken are capable of conditioning or supplying immunocompetent cells. The delay in immune response observed in CB birds may be a reflection of the time necessary for these other sites to attain the level of development found in the bursa of neonatal chicks. The dependence of normal immune response on bursal development has been demonstrated by producing bursa regression in neonatal chicks which was followed by bursa regeneration and a delay in normal antibody production (18). Also, a cellular adaptation may have occurred in our CB birds. That is, certain cells in the chicken may have the potential for an immune commitment but only develop in the altered environment produced by bursectomy.

Summary. Feeding antibiotics to chemically bursectomized (CB) birds has demonstrated that immunological incompetence in CB birds is a direct result of embryonic bursectomy and not secondary to growth rate or factors that increase mortality. Mature CB birds appear to produce normal amounts of agglutinin to sheep red blood cells, but are still deficient precipitin producers. Apparently, the chicken is capable of adapting immunologically to the altered internal environment produced by embryonic bursectomy.

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Obesity in Force-Fed, Hypophysectomized Rats Bearing Hypothalamic Lesions* (32869)

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Because of the prevalence of the belief that some persons gain weight and become obese even though they do not overeat, there has been continued search for some disturbance other than overeating among obese patients. Obesity produced surgically by bilateral, electrolytic lesions in the ventromedial nuclei of the hypothalamus of experimental animals has been one of the most extensively studied obesities. Adult rats bearing such lesions when fed *ad libitum* eat more food than their controls, and the extra food intake of the lesioned

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TABLE I. Body Weight, Carcass Fat, and Dry Residue Weights of 18 Hypex Rats.[a]

Treatment	No.	Body wt. (gm) Initial	Final	Gain	Carcass fat wt. (gm)	Carcass dry residue wt. (gm)	Fat as % of wet wt.[b]
Control	8	179 ± 9.7[c]	191 ± 5.7	12.0 ± 6.9	35.8 ± 2.8	41.3 ± 1.2	19.7 ± 1.1
Lesioned	10	182 ± 6.0	219 ± 11.0	37.0 ± 13.5	50.9 ± 6.6	44.2 ± 1.9	24.1 ± 2.1
$p < 0.05$		No	Yes	Yes	Yes	Yes	Yes

[a] These 18 rats were hypophysectomized when they were 50 days old weighing 200 gm. They were given 10–13 ml of diet per meal with a total of 1160 kcal of food in 31 days, and were killed on day 32 after hypothalamic operations.

[b] Wet wt. is the final body wt. less the wts. of the brain and the gastrointestinal contents.

[c] ± SD.

rats can account for most of their extra weight gain. These findings led Brobeck to the conclusion that this obesity is mainly the result of hyperphagia (meaning overeating) (1). Recently Han and Liu showed that weanling rats with ventromedial hypothalamic lesions involving the median eminence (the VMM lesions) accumulated more body fat than their sham operated controls when both groups were force-fed equal amounts of food throughout the postoperative period (2). Since their lesioned rats also grew less in length, they believed that the VMM lesions have simultaneously caused hyperphagia and a growth hormone insufficiency, and that the latter dwarfed the rats. On equal amounts of food intake, the dwarfed rats requiring less nutriment for growth were receiving relatively more food than their normal controls. The obesity of the force-fed weanling rats with lesions was interpreted as the result of the dual effects of the VMM lesions.

Now I have observed that hypophysectomized (hypex) rats with VMM lesions accumulated more body fat than their hypex controls when both groups were force-fed equal amounts of food. Since in the present studies no overeating was allowed and the pituitary hormones were totally excluded, the results indicate that the VMM lesions have caused some other disturbance(s) which contributed to the development of this surgically produced hypothalamic obesity.

Materials and Methods. Three batches of male hypex rats from the Hormone Assay Laboratory of Chicago were studied. They were hypophysectomized at ages and weights as indicated in Tables I and II, and were kept by the supplier for 10 postoperative days before being sent to this laboratory. After their arrival, individual body weight was measured once every 2–3 days for 2 weeks. Those rats which gained more than 10 gm of body weight in 2 weeks were considered incompletely hypophysectomized and were discarded. The other rats were then given either the VMM lesions or a sham operation under sodium hexobarbital anesthesia (4 mg/100 gm of body wt. i.p.). The detailed methods for placing lesions and sham operations were previously described (3). One injection of hydrocortisone acetate (2.5 mg i.p.) was given to each rat immediately after the surgery.

Forced feeding was done at 4 p.m. and again at 9 p.m. that evening after all rats had recovered from anesthesia. Thereafter all rats were force-fed 3 or 4 times daily until autopsy. Care was taken that rats from the same batch always received equal amounts of diet at each meal.

The diet was prepared as follows: contents of two fresh, medium-sized eggs, contents of a 13-oz can of evaporated milk (Carnation), 75 gm of cane sugar, 10 ml of vitamin B concentrate, and 15 ml of Kaopectate (Upjohn), with water added to bring the total volume to 1000 ml. All these were mixed thoroughly in an electric blender. The diet was calculated to be approximately 11.3% CHO, 4.2% fat, and 4.0% protein by weight. The meal size, total amount given, and the duration of forced feeding to each of the three batches of rats are indicated in Tables I and II.

During autopsy, the completeness of hypophysectomy was checked by examining the sella turcica and the base of the brain for

TABLE II. Body Weight, Carcass Fat, and Dry Residue Weights of the Second and Third Batches of Hypex Rats.

Batch no.	Treatment	No.	Body weight (gm) Initial	Final	Gain	Carcass fat wt. (gm)	Carcass dry residue wt. (gm)	Fat as % of wet wt.
2[a]	Control	5	112 ± 7.2	127 ± 4.0	15.0 ± 7.9	14.4 ± 1.4	28.4 ± 1.7	12.2 ± 1.2
	Lesion	4	106 ± 10.0	130 ± 8.5	24.0 ± 0.9	19.4 ± 2.7	28.0 ± 1.4	15.9 ± 1.6
	$p < 0.05$		No	No	No	Yes	No	Yes
3[b]	Control	5	143 ± 8.7	153 ± 12.2	10.0 ± 8.3	20.5 ± 4.8	35.0 ± 1.9	13.4 ± 2.5
	Lesion	6	134 ± 7.4	170 ± 10.3	36.0 ± 5.5	30.9 ± 3.0	36.9 ± 1.5	19.3 ± 0.9
	$p < 0.05$		No	Yes	Yes	Yes	No	Yes

[a] The second batch of 9 rats were hypophysectomized when they were 35 days old weighing 120 gm. They were given 6–12 ml of diet per meal with a total of 480 kcal of food in 17 days, and were killed on day 18 after hypothalamic operations.

[b] The third batch of 11 rats were hypophysectomized when they were 40 days old weighing 150 gm. They were given 7–13 ml of diet per meal with a total of 640 kcal of food in 19 days, and were killed on day 20 after hypothalamic operations.

remnants of pituitary tissue. The brain was removed and fixed in 10% formalin, and later examined to check the lesion site under a dissecting microscope. All rats whose data are included in this report had complete hypophysectomies and symmetrical VMM lesions. The gastrointestinal tract was removed, opened, rinsed, and blotted. The individual carcass less the brain and the content of the gastrointestinal tract was dried to constant weight in a tared beaker in an oven of 105°C. The carcass fat was then extracted with petroleum ether and the fat weight was determined using previously described methods (4).

Results. Table I presents the body weight, fat content, and dry residue weight of the carcasses of 8 sham operated and 10 lesioned rats force-fed for 31 days. It is clear that the lesioned rats had accumulated more fat and gained more weight than the controls. Table II presents the body weight, fat content, and dry residue weight of the carcasses of two other batches of rats which were hypophysectomized at younger ages and were force-fed for 17 or 19 days (see legends of Table II). Their data again showed that lesioned rats contained more body fat and tended to gain more weight.

Discussion. These findings demonstrate that obesity due to VMM lesions can develop in the absence of both overeating and pituitary involvement, and indicate that other factor(s) must have contributed to the development of this obesity. The exact nature of this other factor(s) is not clear. The various possible answers can be considered by using an energy balance equation:

Energy input = energy storage + energy output.

This equation predicts that in the case of forced feeding which controls the energy input, an increase in energy storage must be accompanied by a reduction of the energy output (heat loss plus work output). The possibilities, therefore, are (a) that the VMM lesions have caused a primary disturbance in fat metabolism favoring an increase in energy storage in the form of fat and the reduction in energy output is the secondary result; (b) that the VMM lesions have caused a primary reduction in energy output and the increase in energy storage is the secondary result; and (c) that the VMM lesions have produced both effects simultaneously. In addition to these three, the possibility that the VMM lesions may have caused alterations in the intestinal absorption of food materials should also be considered. All these, the reduction of energy output in the form of hypoactivity and a decreased oxygen consumption (5), the metabolic alterations favoring an increase in fat deposition (6), and the alterations in intestinal absorption (7), have been shown to exist among hypothalamic obese animals. Similar studies performed on force-fed hypex rats

perhaps can give more meaningful results uncomplicated by over-eating and hormonal factors, and can lead to a better understanding of this experimentally produced obesity.

Summary. These experiments demonstrate that hypothalamic obesity can develop in the absence of hyperphagia and of pituitary involvement, and establish that VMM lesions must have caused additional disturbance contributing to this surgically produced obesity.

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Increased Excretion of Fecal Bile Acids by an Oral Hydrophilic Colloid* (32870)

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In a previous paper (1) we reported that the addition of an oral hydrophilic colloid to the mixed or egg-supplemented diets of normal young male medical students lowered the serum cholesterol to a statistically significant extent. We now report additional studies on two normally active young male medical students which confirm the previous finding regarding the lowering of the serum cholesterol of subjects on a self-selected mixed diet and also we report a large concomitant increase in the excretion of fecal bile acids. On the other hand, fecal excretion of neutral sterols was unaffected by the oral hydrophilic colloid.

Materials and Methods. A preparation of hydrophilic colloid derived from the blond psyllium seed (Plantaga ovata-Forsk) available commercially as Metamucil,[1] was used as the bulk-increasing supplement and as supplied was mixed with an equal weight of dextrose. The mucilloid supplement consisted of 6.4 gm of Metamucil taken 3 times daily with meals, making a total of 9.6 gm of hydrophilic colloid ingested each day. All serum cholesterol values were obtained by the method of Zlatkis (2) and were carried out in duplicate. The experimental subjects were 2 normal male medical students (both age 22 years) on mixed self-selected diets. Both subjects maintained their customary activity and neither reported any change in weight during the study. Each subject obtained one 24-hour feces collection each Wednesday of the study. The specimens were stored under 95% alcohol at 4°C until analyzed. Throughout the study the subjects took 0.5 gm of Cr_2O_3 in gelatin capsules with each meal, 1.5 gm total/day. This inert indicator was used to correct for variations in the amount of feces excreted from day to day and also to correct for any incompleteness in fecal collections (3). No corrections for incomplete analytical recoveries of Cr_2O_3 were made in these

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