rats which were fed iodine-deficient diets. Such bodies have not been observed in hamsters, mice, and monkeys maintained on similar diets.

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Morphological Changes in Rats Fed Navy Beans.* (30340)

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It has been reported that raw navy beans are toxic to rats when fed at a 10% protein level and that autoclaving the beans destroyed the toxic effect(1). The gross changes observed in rats fed raw navy beans were (a) growth inhibition, (b) reduced food consumption, (c) excessive flow of urine, (d) diarrhea, and ultimately (e) death. Booth *et al* (2) studied the morphological changes of heart, lungs, thyroid, testes, spleen, liver, pancreas, kidney, adrenals and intestine and found that all organs of the animals fed raw soybean meal were normal except the pancrease, which was hypertrophic. These authors suggested that the growth inhibition of rats fed raw soybean meal may be due to direct stimulation of the pancreas resulting in excessive flow of critical amino acids contained in the pancreatic enzymes, which are then excreted in the feces. However, studies made by Saxena *et al*(3) indicate that the growth depression in chicks fed raw soybean meal may be due to inability of the hypertrophic pancreas to supply the necessary digestive enzymes. This investigation was undertaken to determine the morphological changes associated with growth inhibition in rats fed raw navy beans.

Experimental. Composition of the basal diet and the details of experimentation have been described(4). Rats were divided into 5 groups of 6 each and the protein component of the diet fed each group was: (a) raw navy beans, (b) autoclaved navy beans, (c) raw navy beans plus 0.6% DL methionine, (d) autoclaved navy beans plus 0.6% DL methionine and (e) casein. The remainder of the diet was the same for all groups. The navy beans and casein were included in the diet to provide a level of 10% protein (N $\times$ 6.25). At the end of the experimental period of 20 days the rats were subjected to euthanasia and the selected organs were excised, blotted and weighed. Sections of tissues were fixed in 10% neutral formalin. Representative samples of each tissue were embedded in paraffin, sectioned at 6 μ and stained with hematoxylin and eosin. Special stains were used where needed.

Results and discussion. The results of the growth rates of the rats fed the diets were similar to those reported previously(4). For comparative purposes, Table I gives weight changes and food consumption of the rats fed raw navy beans, autoclaved navy beans and casein. In this experiment the rats fed the raw navy beans lost weight and consumed considerably less food compared to those fed the autoclaved bean diet. Although protein efficiency ratio value for autoclaved bean diet is greatly increased over that of raw bean diet, it is significantly less than that of the animals fed the casein diet.

Rats fed the casein or the autoclaved navy bean diets appeared to be normal. However, those fed the raw navy bean diet were emaciated, debilitated, had rough hair coats and were wet around the genitalia. There was paleness of the mucous membranes and the livers were yellowish in color. The abdominal

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TABLE I. Effect of Feeding Experimental Diets on Growth of Rats.

Protein source	Total change in wt (g)*	Total food intake (g)*	Protein efficiency ratio (PER)*
Casein	75.6	232	3.28 ± .12†
Raw navy beans	−12.8	84	−1.54 ± .10
Autoclaved navy beans‡	25.4	166	+1.58 ± .11

* Expressed as average of 6 rats for 20 days.
† Standard error of mean.
‡ Autoclaved at 121°C for 5 min.

cavity contained excess amounts of serous fluid.

The effects of the diets on the weights of different organs of the rats are given in Table II. The data indicate that the weight of heart and kidneys of rats fed raw navy beans is significantly increased ($P<0.05$) as compared to the weight of these organs in animals fed the casein diet. Phadke and Sohonie(5) also observed the similar differences in the weight of these organs of rats fed field bean diet. There were no significant differences in the weights of different organs of rats fed autoclaved bean diet as compared to those fed the casein diet.

The following tissues were examined microscopically: kidneys, liver, adrenal glands, stomach, duodenum, jejunum, ileum, pancreas, bone, skin, esophagus, trachea, and lymph nodes. The morphological changes observed in rats fed the raw navy bean diet were as follows:

Liver: (Fig. 1) Increased numbers of vacuoles were present in hepatic cells, and these were more prominent near the periphery but were distributed throughout the lobule. There was some derangement of liver cords and confluence of cytoplasm of adjacent cells. The nuclei were more densely packed, indicating a relative decrease in size, and the nuclei contained enlarged nucleoli. Fatty metamorphosis was marked in sections stained with oil red O. Similar findings were noted in rats fed the diets deficient in essential amino acids(6,7).

TABLE II. Effect of Feeding Experimental Diets on Weight of Different Organs of Rats. (Average weight of organs expressed in g/100 g of body wt.)

Organ	Casein	Raw navy beans	Autoclaved navy beans*
Liver	4.20 ± .06	3.84 ± .20	4.94 ± .20
Heart	.50 ± .001	.63† ± .001	.55‡ ± .002
Spleen	.27 ± .04	.18 ± .04	.28 ± .04
Kidney	.91 ± .02	1.40† ± .06	1.06‡ ± .17
Adrenals	.031± .005	.032 ± .005	.023 ± .001

± standard error of mean.
* Autoclaved at 121° for 5 min.
† t value is significant at 5% level when compared to casein group.
‡ t value is not significant at 5% level when compared to casein group.

Pancreas: (Fig. 2) According to micrometer measurements, acinar cells were reduced in size. The cytoplasm was almost devoid of secretory granules, occupied less area and uniformly stained deeply basophilic. Acinar arrangement and cell outline were indistinct.

Thyroid: (Fig. 3) Most of the follicles were small and contained little or no colloid, therefore rendering the thyroid gland more cellular per unit area.

Kidney: A few distal convoluted tubules and most of Henle's loops were dilated, with the lining epithelium pressed against the basement membrane in a thin layer.

Esophagus: There was a wide layer of keratin on the epithelial surface, and this partially filled the esophageal lumen. Foci of cellular remnants suggestive of active desquamation were present.

Skin: There was mild hyperkeratosis with excessive keratin attached to the free surface of the skin.

The morphological changes observed in rats fed autoclaved bean diet were:

Liver: Many of the hepatic cells had enlarged nuclei and nucleoli. Cellular outline was indistinct, especially in cells near the periphery of the hepatic lobule.

Kidney: Small hyaline casts partially filled the lumens of many collecting tubules.

Microscopic lesions were not observed in the tissues of rats fed the autoclaved navy bean diet plus methionine. The lesions in the tissues of rats fed the raw navy bean diet plus methionine were similar to those fed the

raw navy beans. Therefore, these details are not included in this report.

It appears that the differences in the weights of organs and the morphological changes observed in the tissues of rats fed raw navy beans are due mainly to a deficiency and/or unavailability of critical amino acids such as methionine. Bowman(8) has shown the presence of heat labile trypsin inhibitor in navy beans, the action of which would be expected to accentuate the deficiency of critical amino acids by inhibiting the action of trypsin. The beneficial effect of autoclaving the raw navy beans in inhibiting the morphological changes in rats could be due to the destruction of trypsin inhibitor and thereby to increase the availability of critical amino acids. It was pointed out in

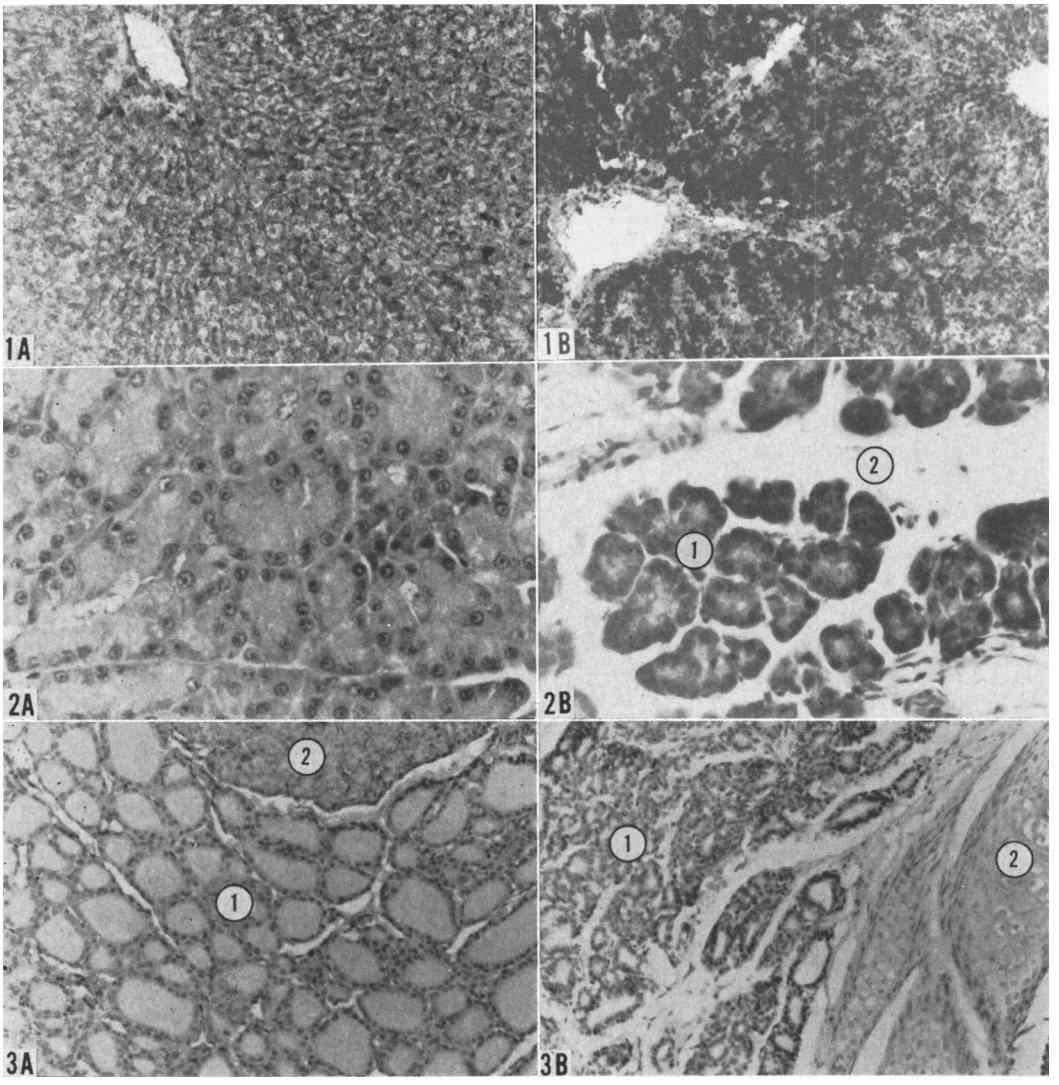

FIG. 1. A. Liver from a control rat fed a casein diet. Oil Red O. × 112. B. Liver from a rat fed a raw navy bean diet. Note fatty metamorphosis. Oil Red O. × 112.

FIG. 2. A. Pancreas from a control rat fed a casein diet. H. and E. × 281. B. Pancreas from a rat fed a raw navy bean diet. (1) atrophic acini, and (2) edema. H. and E. × 281.

FIG. 3. A. Thyroid gland from a control rat fed a casein diet. (1) thyroid follicles containing colloid, and (2) parathyroid gland. H. and E. × 112. B. Thyroid gland from a rat fed raw navy beans. (1) atrophic follicles containing little colloid, and (2) cartilage of trachea. H. and E. × 112.

a previous publication(9) that raw navy beans were poorly digested *in vitro* by trypsin as compared to autoclaved beans.

Haines and Lyman(10) proposed that the soybean trypsin inhibitor acts by stimulating the pancreas to secrete excessive amounts of enzymes, thus creating partial deficiencies of the most limiting amino acids in the animals fed soybean meal. It may be that the navy bean trypsin inhibitor as a factor present in raw navy beans also stimulates the production of pancreatic enzymes resulting in the loss of critical amino acids through feces. Another possibility of explaining the unavailability of critical amino acids is that there may be present a factor in raw navy beans which inhibits the production by the pancreas of the necessary digestive enzymes, thus making the intestinal proteolysis incomplete, as postulated by Saxena *et al*(3) in explaining the low nutritive value of raw soybeans. However, morphological changes observed in the pancreas of animals fed raw navy beans did not allow differentiation between these two mechanisms, because the condition in which the cytoplasm of the pancreas was devoid of secretory granules could arise from either mechanism of action described.

In the light of the above discussion, it can be concluded that the growth inhibition of rats fed raw navy beans could be partly due to the impairment in the availability of critical amino acids and partly due to the methionine deficiency. Moreover, this condition can be further aggravated by the low food intake resulting in the limited supply of amino acids in the animals fed raw navy beans.

Summary. An experimental diet of raw navy beans produced changes in rats which included: growth inhibition, reduced food consumption, pancreatic acinar atrophy, fatty metamorphosis of the liver, thyroid follicular atrophy, and hyperkeratosis of the esophagus and skin. Significant morphological changes were not observed in the organs of rats fed an autoclaved navy bean diet.

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Changes in Metal Distribution of the Avian Oviduct During the Ovulation Cycle.* (30341)

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The elucidation of the mechanisms of metal transport in biological systems has been of long and continued interest(1). Many systems have been studied for this purpose and among them the avian shell gland appears to be especially suited since in the domestic fowl this tissue produces in 20 hours a shell which contains about 2.0 g of calcium(2). Investigation of this highly amplified system of calcium transport may provide information which would be of general application to transport theory. In the present investiga-

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