

ALTERATION OF RESISTANCE OF THE RAT TO TUBERCULOSIS
WHEN MAINTAINED ON AN ATHEROGENIC DIET*

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Serum cholesterol levels are known to be depressed in a variety of acute (1-5) and chronic (6-9) infectious diseases. Conflicting data, however, appear in the literature concerning the effect of cholesterol administration on the pathogenesis of such diseases. Intraperitoneal injection of cholesterol reportedly causes a decreased susceptibility to tuberculous infection in animals (10) and man (11). When given orally, on the other hand, little or no beneficial influence on tuberculous disease has been noted (12, 13), and in at least one study such treatment proved detrimental to the defense responses of the host (14).

Little recent work on this subject has been reported and a restudy of the problem appeared justified.

We chose to study the effect of feeding rations containing large amounts of cholesterol, thiouracil, and sodium cholate on the course of disease produced in the albino rat by *Mycobacterium tuberculosis*. Such rations have been shown to be capable of eventually inducing atherosclerotic vascular changes. The results described in this paper indicate that the rat, which is normally highly resistant to tuberculosis, becomes markedly susceptible to infection with *M. tuberculosis* when maintained on these rations.

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Materials and Methods

Organisms.—The H37Rv strain of *Mycobacterium tuberculosis* was obtained from the Standard Culture Depot of the National Tuberculosis Association and maintained at -20°C without transfer. Prior to use for infection of animals, an aliquot of this stock culture was transferred to 20 ml of Kirchner's medium containing 0.5 per cent bovine serum albumin (fraction V) and 0.05 per cent tween 80 (polyoxyethylene sorbitol mono-oleate) and incubated at 37°C until an optical density of approximately 0.200 was obtained. Kirchner's medium consisted of sodium phosphate, dibasic, 3.0 gm; potassium phosphate, monobasic, 4.0 gm; sodium citrate, 2.5 gm; asparagine, 5.0 gm; magnesium sulfate, 0.6 gm; ferric ammonium citrate, 0.05 gm; glycerol, 20.0 gm; H_2O , *q.s.* 1000 ml. A second transfer was made using 2.0 ml of the first culture to 20 ml of the medium without addition of bovine serum albumin. After 5 days' incubation, the culture was diluted to give an optical density of 0.230 to 0.250, so as to contain approximately 8×10^8 viable units per ml.

Animals.—Sprague-Dawley female albino rats weighing 90 to 100 gm were obtained from the Sunset Hills Farm, Lawrence, Kansas. The animals were randomly caged and maintained on Purina laboratory chow. Groups of 5 to 8 animals were housed in large cages with wire bottoms. Water was present at all times.

Diets.—A semisynthetic diet similar to that described by Fillios *et al.* (15) was utilized. The basal ration consisted of vitamin-free casein, 20 per cent; sucrose, 58 per cent; Wesson salt mixture, 4 per cent; alphacel, 7.6 per cent; vitamin mixture, 0.3 per cent; corn oil, 8 per cent; and cod liver oil, 2 per cent. The vitamin mixture, per 100 kg diet, consisted of alpha tocopherol, 10.0 gm; *p*-aminobenzoic acid, 50.0 gm; *i*-inositol, 100.0 gm; choline, 200.0 gm; thiamine, 0.5 gm; riboflavin, 0.5 gm; pyridoxine, 0.25 gm; calcium pantothenate, 5.0 gm; niacin, 8.0 gm; folic acid, 0.025 gm; biotin, 0.02 gm; and menadione, 0.5 gm. In different diets, cholesterol, 5 per cent; sodium cholate, 1 per cent; and thiouracil, 0.3 per cent were substituted for equivalent quantities of alphacel. The vitamins, salt mixture, and casein of the diet were combined and thoroughly mixed in a ball mill. The remaining dry ingredients were blended with the former by hand mixing. Corn oil and cod liver oil were added and suspended into the diet by passage through a meat grinder. Sufficient water was added to make a thick paste of the diet. The final ration was stored at -20°C . Only sufficient diet was given at one time to provide the animals for 1 to 2 days. Control rations consisted of purina laboratory chow. Diets were fed *ad libitum*. Animals were maintained on the same diet until termination of the experiment.

Infection.—Rats were infected by intravenous injection (caudal vein) of 0.2 ml of the inoculum previously described. Dilution of the rat vein was accomplished by soaking the animal's tail in water at 60°C for approximately 1 minute.

Clinical-Pathological Observations.—Sections of organs fixed in formalin were stained with hematoxylin and eosin and with Ziehl-Neelsen acid-fast stain. Frozen sections were prepared and stained with turkey oil red for identification of lipid.

Hematological observations were made by standard techniques on blood obtained from the caudal vein.

Differential determination of fecal bacteria was performed on feces homogenized in saline with the aid of glass beads. Feces were stored at -20°C after collection until the time of analysis.

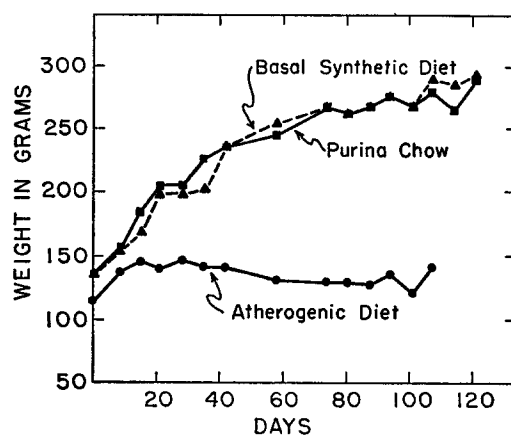
Analyses on 24 hour samples of urine were performed with the aid of Ames clinical reagent test kits (Ames Company, Inc., Elkhart, Indiana).

Cholesterol Determinations.—Serum cholesterol levels were determined by the method of Sperry and Webb (16). Cholesterol values of tissue were determined by a similar method on acetone-alcohol extracts of tissue saponified with alcoholic KOH. Prior to saponification the tissues were blotted on No. 1 Whatman filter paper to remove excess blood and fluid and dried to a constant weight at 100°C .

RESULTS

Resistance of Normal Rats to Injection of M. tuberculosis.—Preliminary experiments were performed to determine the resistance of normal rats to tuberculosis. Under the conditions of our experiments, female rats did not develop overt symptoms of disease for more than 60 days following intravenous injection of up to 1.0 ml of the culture of *M. tuberculosis*.

Decreased Resistance of Rats to Injection of M. tuberculosis when Maintained on Atherogenic Rations.—In the first experiment to be described, young female rats weighing 90 to 100 gm. were placed on commercial rations, on unaltered



TEXT-FIG. 1. Change of weight of rats fed different diets. Atherogenic diet consisted of basal synthetic diet supplemented with thiouracil, sodium cholate, and cholesterol.

basal synthetic rations, or on the synthetic ration supplemented with thiouracil, sodium cholate, and cholesterol. After being maintained on their respective diets for 64 days, each group of animals was divided into two comparable subgroups and continued on their respective diets. One of each of the subgroups was injected intravenously with 0.2 ml of the culture of *M. tuberculosis*.

Rats maintained on the unaltered synthetic ration gained weight throughout the experimental period in a manner similar to that of animals fed the commercial ration (Text-fig. 1). The physical appearance of animals in these two groups was similar throughout the experiment. On the other hand, animals maintained on the atherogenic ration (*i.e.*, containing thiouracil, sodium cholate, and cholesterol) failed to increase their weight after the first 2 weeks following initiation of the diet. The animals in this group manifested roughened hair and other signs of unthriftiness. This stunting of growth and lack of sleekness was determined to be caused by the dietary thiouracil.

The mortality data of this experiment are given in Table I. Rats maintained on either of the control rations were resistant to the lethal effects of the experi-

mental infection for at least 60 days following infection. Contrariwise, those animals maintained on the atherogenic rations were found to be markedly susceptible to the infection, all animals succumbing within 20 days following challenge.

Similar results were obtained in several other experiments of the same nature. Animals maintained on either the commercial ration or on the unaltered synthetic ration suffered no mortality during the course of any experiment. Again the majority of rats fed the atherogenic ration rapidly succumbed following infection with *M. tuberculosis*. Of 30 rats maintained on this diet in four experiments, 25 rats (84 per cent) died within 60 days following infection.

TABLE I

Susceptibility of Rats to Mycobacterium tuberculosis when Maintained on Atherogenic Rations

Diet	No. of rats	Day of death following infection*	Survived 60 days following infection
			<i>per cent</i>
Purina chow.....	8	None	100
Basal synthetic diet.....	8	None	100
Synthetic diet + cholesterol, thiouracil, sodium cholate.....	8	14, 14, 14, 15, 15, 16, 16, 19	0

* *M. tuberculosis*, strain H37Rv, injected intravenously on the 64th day following initiation of diets.

Twenty of these rats (67 per cent) died during the first 30 days following infection, while another 5 animals (17 per cent) died during the next 30 day period. In one experiment all animals (4) maintained on this ration survived the infection.

For maximum loss of resistance it was necessary to maintain the rats on atherogenic rations for 50 to 60 days of prefeeding. When the prefeeding period was shortened to 30 days the majority of animals survived the infection. When the dietary regimen was initiated at the time of infection only an occasional animal died.

Male rats maintained on the atherogenic ration under similar conditions also displayed an increased susceptibility to tuberculosis. A difference in the degree of susceptibility was noted with the majority of such animals surviving the first 60 days following infection, although succumbing within the next 30 to 45 days.

Pathological Observations.—In the gross, rats maintained on the atherogenic diet, whether infected or not, had marked fatty-type changes in the liver and increased accumulations of visible lymphoid tissue in the lung. Infected animals

could not be differentiated from uninfected animals in these respects. While little is known of the pathological significance of lung lymphoid tissue in rats, its increased presence in these animals could have led to an impaired cellular response to the infection. Splenic atrophy occurred in uninfected animals fed the atherogenic diet. Enlargement of the spleen occurred in infected animals maintained on either control or atherogenic rations. The degree of enlargement was relatively similar in both groups. No other changes were apparent in the gross in infected animals. Recognizable tuberculous lesions were not found.

Microscopic observation of stained tissue sections from animals injected with *M. tuberculosis* revealed that the loss of resistance to the infection occasioned by the atherogenic ration was paralleled by increased severity of pathological variation.

The appearance of lung sections from animals fed the unaltered synthetic ration and infected 14 days prior to sacrifice (Fig. 1) was characteristic of a well controlled granulomatous process and was similar to that described by other investigators (17). Most fields presented essentially unchanged architecture. In the affected areas, infiltration was patchy. The foci were composed predominantly of well circumscribed epithelioid cells. Acid-fast bacilli were found only in these foci. In sections of liver, areas of proliferation were found in the regions of portal spaces with some accumulation of epithelioid cells and a widespread infiltration of lymphoid elements. In the spleen, the reaction was characterized by numerous focal accumulations of cells of the epithelioid type. These occurred principally in the areas of the Malpighian bodies.

The microscopic appearance of lung sections from animals maintained on the atherogenic ration and infected for the same 14 day period was characterized by widespread, diffuse, granulomatous pneumonia, as well as interstitial pneumonitis, as evidenced by thickening of the alveolar walls (Fig. 2) Cellular infiltration of the parenchymal framework was somewhat patchy with a tendency for coalescence of considerable portions of these areas. Epithelioid-appearing elements were found, but no isolated granulomatous foci were apparent. Acid-fast bacilli were spread diffusely throughout the affected areas. Infiltration of lipid-staining material into the affected areas was noted, with this material accumulating predominantly in mononuclear cells. In sections of liver, considerable infiltration by cellular foci of epithelioid-appearing elements was observed. Many of these were somewhat rounded to polyhedral in shape and revealed a pale cytoplasm. In the spleen, the reaction was characterized by numerous focal accumulations of cells of the epithelioid type diffused widely throughout the spleen but with particular localization in the area of the Malpighian bodies.

Caseation necrosis was not observed in any of the lesions in either group of animals.

In the hearts of non-infected animals maintained on the atherogenic diet,

only minimal accumulations of lipid-staining material occurred. No lesions were observed microscopically in the coronary arteries. A slight amount of degenerative change was noted in the myocardium.

Cardiac tissue from infected and non-infected animals on the atherogenic ration was similar in microscopic appearance. In an occasional infected animal, however, small myocardial lesions were observed as evidenced by a relative increase in cellularity, with some loss of normal architectural detail. No evidence of occlusive vascular disease was noted in any of the animals. The primary cause of death was not considered to be of cardiac origin in these animals.

Clinical and Laboratory Observations.—The results of the several experiments described so far demonstrated that maintenance of rats on atherogenic rations increased the severity of subsequent tuberculous infection. During the course of these experiments several observations on the general well-being of uninfected animals were made which are worth noting.

There were no deaths among non-infected animals maintained on the different diets during the course of any experiment. In a separate experiment, rats maintained on the atherogenic ration for longer than 9 months experienced no apparent illnesses.

Rats on the atherogenic ration consumed only half as much diet as control animals. Since animals maintained on this diet were approximately one-half the size of control animals, there was no apparent difference in the food intake, per gram body weight, of the two groups of animals. Nevertheless, this decreased intake, while insufficient to cause death in uninfected animals, may have contributed to the severity of the tuberculous processes.

Urinalysis, performed weekly, failed to indicate any abnormality in renal function. Bacteriological analysis of the fecal excreta of the rats, performed weekly, did not reveal significant differences in the total numbers of, or general type of, organisms present. Organisms which are usually considered to be pathogenic were not found in the feces of any animal. Hematological determinations, made on blood from rats which had been maintained on the various diets for 6 weeks, revealed normal values for white blood cell count, differential count, and for the hematocrit.

The Response of Rats to Injection of M. tuberculosis when Maintained on Rations Containing Components of the Atherogenic Ration.—The effect of feeding rations containing thiouracil, sodium cholate, and cholesterol, singly or in paired combination, on the resistance of the rat to infection with *M. tuberculosis* was studied.

The procedures in these experiments were similar to those described for the first experiment. Young female rats were used. In the experiment illustrated in Table II, animals were maintained on rations containing the desired factors for 60 days. At this time the animals were injected with *M. tuberculosis*.

The outward appearance of the animals during the feeding period was similar

to that of the rats in the first experiment. Animals in groups which were not fed thiouracil developed normally, whereas animals maintained on any ration containing thiouracil became stunted in growth with roughened hair.

Rats maintained on diets supplemented with either cholesterol or sodium cholate alone revealed no loss of resistance. This was found to be true in several experiments. A similar absence of mortality occurred, in the majority of cases, in groups of animals whose rations were supplemented with thiouracil alone. In a single experiment, however, an appreciable mortality (4 out of 8) occurred in animals maintained on this ration.

TABLE II
Susceptibility of Rats to Mycobacterium tuberculosis when Maintained on Rations Containing Atherogenic Dietary Components

Diet	No. of rats	Day of death following infection*	Survived 60 days following infection <i>per cent</i>
Purina chow.....	8	None	100
Basal synthetic diet.....	8	None	100
Synthetic diet + cholesterol.....	8	None	100
Synthetic diet + sodium cholate.....	8	None	100
Synthetic diet + thiouracil.....	8	None	100
Synthetic diet + thiouracil, cholesterol.....	8	14, 20, 34	62
Synthetic diet + thiouracil, sodium cholate..	8	13, 23	75
Synthetic diet + cholesterol, sodium cholate..	8	36	88
Synthetic diet + cholesterol, thiouracil, sodium cholate.....	8	8, 8, 15, 17, 42, 42, 51	12

* *M. tuberculosis*, strain H37Rv, injected intravenously on the 60th day following initiation of diets.

A partial loss of resistance consistently occurred in animals whose diets were supplemented with thiouracil and cholesterol, thiouracil and sodium cholate, or cholesterol and sodium cholate. This loss of resistance was not sufficient to cause death in the majority of animals of any diet group and was never as pronounced as when all three factors were included in a single diet.

All non-infected animals in this experiment survived throughout the test period. In other experiments an occasional control animal died after being maintained on rations supplemented with thiouracil alone for periods greater than 4 months. These unexplained deaths occurred only in animals maintained on this diet.

Alteration of Serum and Liver Cholesterol Concentration in Rats Maintained on Atherogenic Rations.—Cholesterol concentrations of serum and liver were determined in paired, non-infected animals of the groups reported in Tables I

and II as well as in animals from other experiments of like nature. The results from a representative experiment are given in Table III. Serum cholesterol levels were found to be elevated in all groups maintained on diets containing cholesterol. An especially high level was present in the serum of animals on the atherogenic ration. Similar elevations of cholesterol occurred in liver tissue of animals on these diets.

It was apparent that the loss of resistance occasioned by the atherogenic diet could not be attributed solely to increased levels of stored cholesterol. On the

TABLE III
Effect of Feeding Atherogenic Dietary Components on the Serum and Liver Cholesterol Levels of the Rat

Additions to basal synthetic diet	Concentration of cholesterol*	
	Serum	Liver
	<i>mg per cent</i>	<i>mg/gm</i>
None.....	130 ± 10	7.8 ± 0.06
Cholesterol.....	307 ± 66‡	87.0 ± 24.0‡
Thiouracil.....	152 ± 33	5.7 ± 1.0
Sodium cholate.....	125 ± 20	7.9 ± 0.5
Cholesterol, sodium cholate.....	358 ± 54‡	183.0 ± 24.0‡
Cholesterol, thiouracil.....	254 ± 21‡	74.0 ± 8.0‡
Thiouracil, sodium cholate.....	176 ± 14‡	13.4 ± 2.1‡
Cholesterol, thiouracil, sodium cholate.....	1070 ± 379‡	146.0 ± 44.0‡

* Length of prefeeding period prior to sacrifice was 57 to 61 days. Values given are the average for 5 animals in each group.

‡ Refers to values significantly elevated ($p = < 0.05$) with respect to the cholesterol concentration of the group fed unaltered basal synthetic diet.

contrary, no apparent increase in susceptibility occurred in animals whose rations were supplemented with cholesterol alone or with sodium cholate and cholesterol although these rations caused a marked increase in the cholesterol levels of serum and liver.

DISCUSSION

The results described in this paper would indicate that maintaining rats on diets of an atherogenic nature increases their susceptibility to injection with *M. tuberculosis*.

Our results would not indicate that the loss of resistance occasioned by the atherogenic ration was attributable solely to increased stores of cholesterol. Nor could the resistance changes be explained solely by the alterations induced by the dietary thiouracil. Marked changes in resistance occurred only when all components of the atherogenic diet were present in a single diet. Such diets

probably cause drastic changes in the rat. The nature of the changes which were most intimately involved in resistance processes cannot be ascertained from the information on hand. It is of interest, in this respect, that of the numerous reported attempts to alter the susceptibility of rats to tuberculosis, only those causing physiological alterations have proven effective. These include thyroid-parathyroidectomy (18), pancreatectomy (19), hypophysectomy (20), alloxan diabetes (21), and cortisone treatment (22).

It is necessary to attempt to separate the effect of the diet on cardiovascular changes from its effect on resistance mechanisms. Fillios *et al.* (15), who maintained rats on a ration comparable to the atherogenic rations used in these experiments, found microscopically observable lipid in vascular tissue as early as 31 days after initiation of the dietary regimen. However, these investigators were unable to discern gross alteration in vascular tissue except in animals maintained on atherogenic rations for periods much greater than those used in the experiments reported here. In our experiments, alterations of cardiovascular tissue were not found to be sufficient to cause, *per se*, a marked alteration of life expectancy in either infected or non-infected animals. Careful autopsy of animals which succumbed during the experiments failed to indicate hemorrhagic infarction in the gross or other vascular changes which may have contributed significantly to the demise of the animals.

It cannot be concluded, however, that subtle alteration of vascular tissue does not play some part in the change in resistance to tuberculosis. It is known that after intravenous injection of tubercle bacilli, organisms can leave the blood stream by infarction of the capillary vessels of the host animal, and thereby enter the tissue proper (23). Under such conditions, it is apparent that even undiscernible alterations of such vessels can modify the initial distribution of the bacilli in the host.

SUMMARY

Albino rats maintained for 50 to 60 days on rations containing cholesterol, sodium cholate, and thiouracil were found to be highly susceptible to the lethal effects of infection with *Mycobacterium tuberculosis*. Maximum loss of resistance occurred only when all three components were present in a single ration. A lesser degree of susceptibility resulted from the deletion of one or more of the factors from the diet. Animals maintained on control rations never died of the infection.

Histopathological studies of stained sections of tissues of infected animals revealed that the reaction of the host to the bacillus was more extensive and less well contained in animals fed the complete atherogenic ration than in the controls.

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BIBLIOGRAPHY

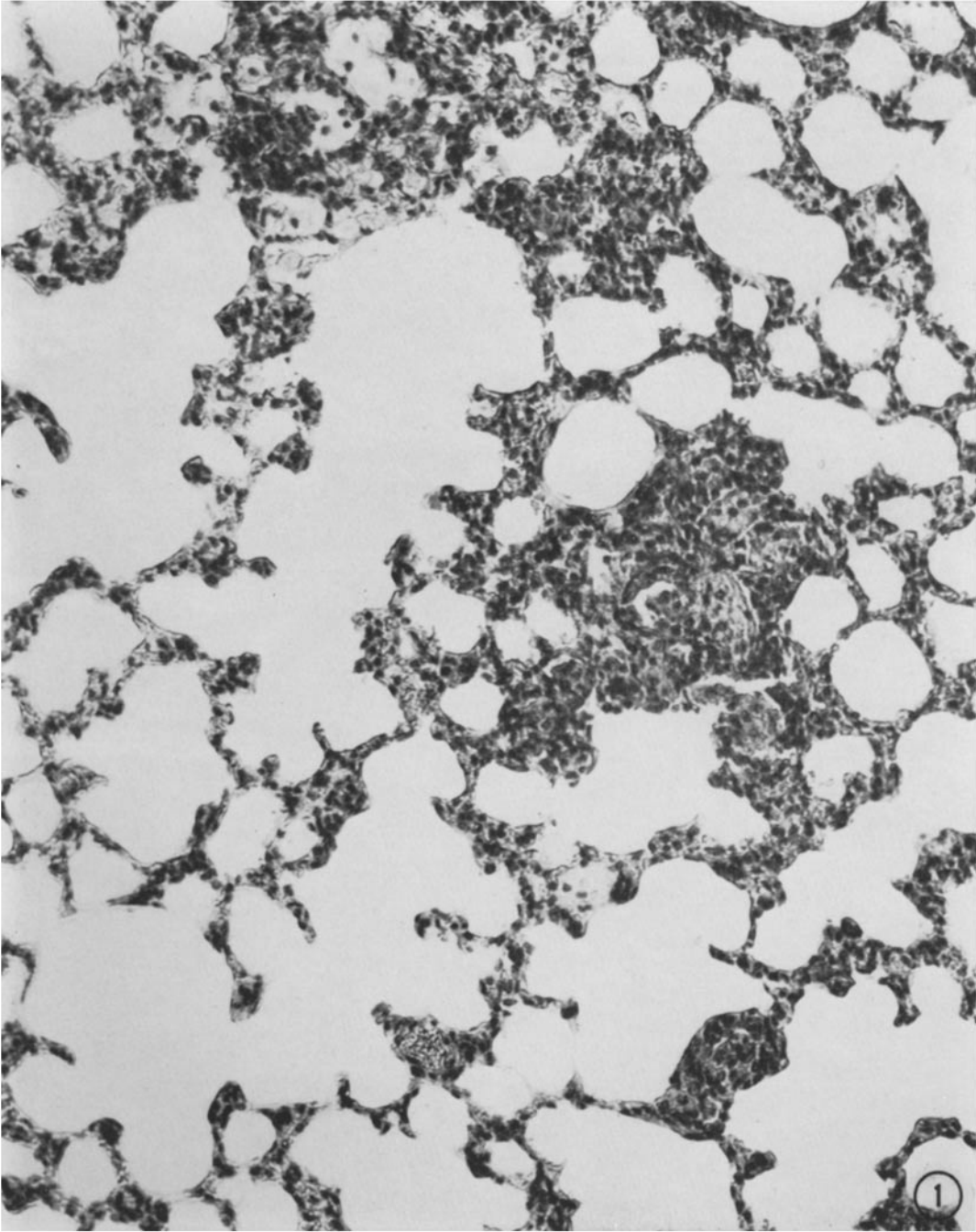
1. Denis, W., Cholesterol in human blood under pathological conditions, *J. Biol. Chem.*, 1917, **29**, 93.
2. Stoesser, A. V., and McQuarrie, I., Influence of acute infection and artificial fever on the plasma lipids, *Am. J. Dis. Child.*, 1935, **49**, 658.
3. Chauffard, A., Laroche, G., and Grigaut, A., Evolution de la cholestérinémie au cours des infections aiguës, *Semaine Med.*, 1911, **31**, 577.
4. Steiner, A., and Turner, K. B., Observations on the serum cholesterol in acute infections as recorded during and after pneumonia, *J. Clin. Inv.*, 1940, **19**, 373.
5. Millan Mendez Gil, A., La coleslerinemia en la tuberculosis pulmonar: su valor pronóstico, *Arch. Med. Exp.*, Madrid, 1953, **16**, 303.
6. Carnimeo, N., and De Sanctis, U., La colessterol-esterasi serica nella tubercolosi pulmonare, *Biochim. Appl.*, 1957, **4**, 432.
7. Henning, B. H., The lipoids of the blood in tuberculosis, *J. Biol. Chem.*, 1922, **53**, 167.
8. Boyd, T. C., and Roy, A. C., Notes on the cholesterol content of Indian blood in health and in leprosy, *Indian J. Med. Research*, 1928, **15**, 643.
9. King, S. E., and Brueger, M., Plasma cholesterol in tuberculosis and amyloid disease, *Ann. Int. Med.*, 1935, **8**, 1427.
10. Shope, R. H., The influence of cholesterol on experimental tuberculosis, *J. Exp. Med.*, 1928, **48**, 321.
11. Bersani, I., La coleslerina nella terapia della tubercolosi, *Tubercolosi*, 1923, **15**, 135.
12. Jaffé, R. H., and Levinson, S. A., The influence of hypercholesterinaemia on experimental tuberculosis of the rabbit, *Am. Rev. Tuberc.*, 1925, **11**, 217.
13. Levinson, S. A., Fat therapy in experimental tuberculosis of rabbits, *Am. Rev. Tuberc.*, 1931, **23**, 527.
14. Rix, E., and Schedtler, O., Der Einfluss von Cholesterin und Neutralfetten auf den Ablauf der experimentellen Tuberkulose, *Beitr. Klin. Tuberk.*, 1938, **91**, 493.
15. Fillios, L. C., Andrus, S. B., Mann, G. V., and Stare, F. J., Experimental production of gross atherosclerosis in the rat, *J. Exp. Med.*, 1956, **104**, 539.
16. Sperry, W. M., and Webb, M., A revision of the Schoenheimer-Sperry method for cholesterol determination, *J. Biol. Chem.*, 1950, **187**, 97.
17. Wessels, C. C., Tuberculosis in the rat. III. The correlation between the histological changes and the fate of living tubercle bacilli in the organs of the albino rat, *Am. Rev. Tuberc.*, 1941, **43**, 637.
18. Steinbach, M. M., Experimental tuberculosis in the albino rat: the comparative effects of avitaminosis, suprarenalectomy and thyroid-parathyroidectomy on experimental tuberculosis, *Am. Rev. Tuberc.*, 1932, **26**, 52.
19. Steinbach, M. M., and Duca, C. J., Experimental tuberculosis in hyperglycaemic albino rats, *Am. Rev. Tuberc.*, 1942, **46**, 304.
20. Steinbach, M. M., Duca, C. J., and Malomut, M. C., Experimental tuberculosis in hypophysectomized rats, *Am. Rev. Tuberc.*, 1944, **49**, 105.
21. Roche, P., Cummings, M. M., and Hudgins, P. C., Comparison of experimental

- tuberculosis in cortisone-treated and alloxan-diabetic albino rats, *Am. Rev. Tuberc.*, 1952, **65**, 603.
22. Cummings, M. M., Hudgins, P. C., Whorton, M. C., and Sheldon, W. H., The influence of cortisone and streptomycin on experimental tuberculosis in the albino rat, *Am. Rev. Tuberc.*, 1952, **65**, 396.
23. Mayer, E., Jackson, E. R., Whiteside, E. S., and Alverson, C., Experimental embolic pulmonary tuberculosis in mice, *Am. Rev. Tuberc.*, 1954, **69**, 419.

EXPLANATION OF PLATES

PLATE 115

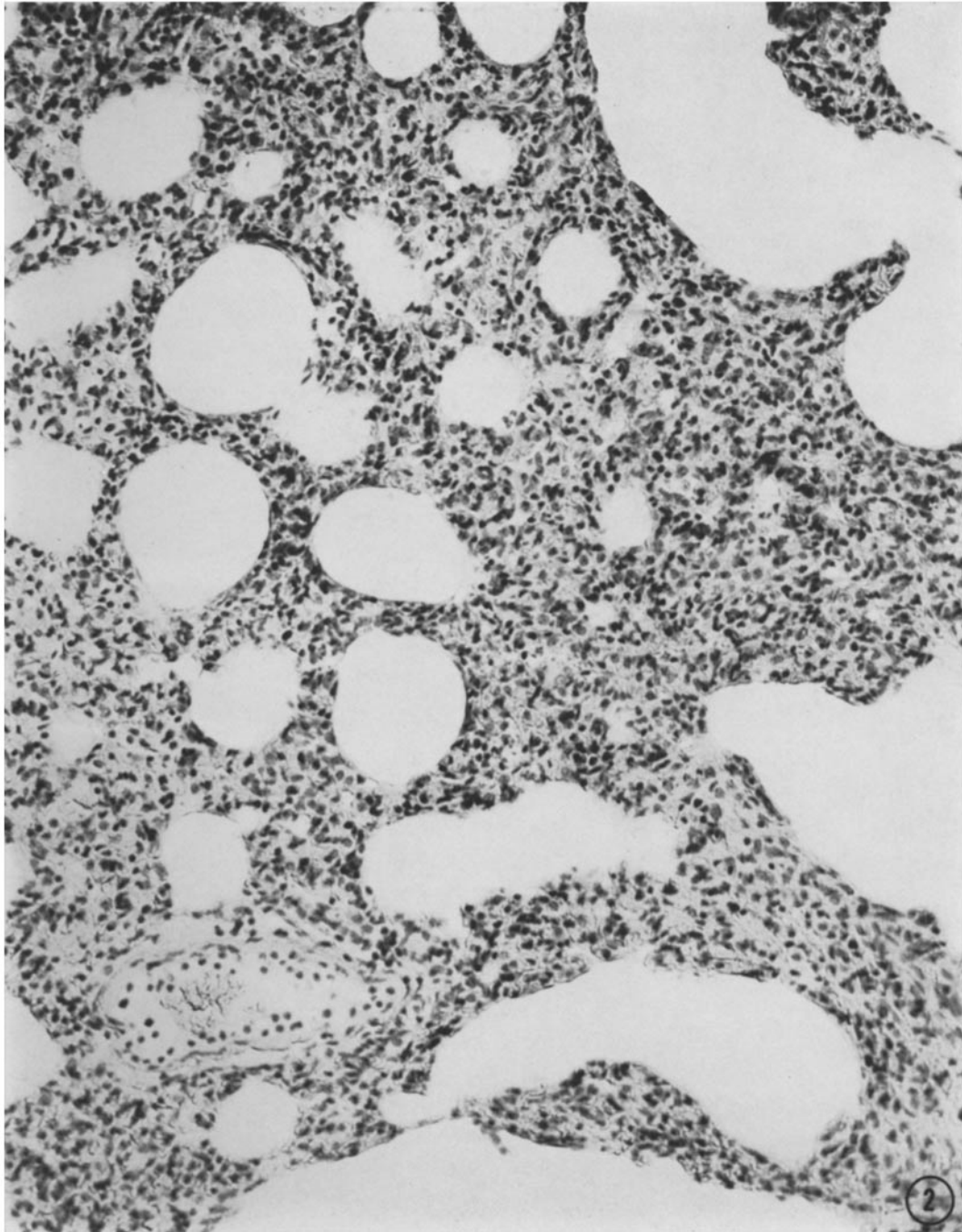
FIG. 1. Section of lung from rat fed unsupplemented synthetic rations and sacrificed 14 days after intravenous injection of *Mycobacterium tuberculosis*. Relatively unchanged architecture of lung tissue is shown. In the affected areas, there is infiltration with circumscribed foci composed primarily of epithelioid cells. Hematoxylin-eosin. $\times 300$.



(Costello *et al.*: Rat resistance to tuberculosis)

PLATE 116

FIG. 2. Section of lung from rat which was fed synthetic rations supplemented with thiouracil, sodium cholate, and cholesterol and which died 14 days after intravenous injection with *Mycobacterium tuberculosis*. Widespread, diffuse, granulomatous pneumonia, as well as interstitial pneumonitis is observed. No isolated granulomatous foci are present. Hematoxylin-eosin. $\times 300$.



(Costello *et al.*: Rat resistance to tuberculosis)