

despite the inability to maintain adequate nutrition, and in the face of intermittent, severe vomiting (1500 to 2500 cc every 3-4 days) and a sustained elevated temperature due to thrombosis of veins resulting from the methods used, this animal was maintained for 45 days, and during the last 35 days all fluids were administered intravenously.

Conclusions. The length of survival of

dogs having a low simple intestinal obstruction depends upon the ability to maintain fluid, electrolyte, and nutritional requirements. By parenteral alimentation with the methods used adequate nutritional requirements were not obtained, but in 1 animal in which these requirements were approached more closely the length of survival was 45 days.

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A Rapid Mouse Test for Laboratory Diagnosis of Tuberculosis.*

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Recent reports have shown that pigmented strains of mice (dba, C3H and C57 black) are more susceptible to tuberculosis than albino mice.^{1,2} We have confirmed these findings and have completed additional experiments in which albino and pigmented mice were inoculated intraperitoneally with tubercle bacilli suspended in various diluents. This was done with the hope of finding an enhancing agent for a rapid diagnostic test for tuberculosis.

This paper is a preliminary report on the use of gastric mucin for enhancing the virulence of the H37Rv and several recently isolated strains of mammalian tubercle bacilli for pigmented mice. Although there have been no previous reports to our knowledge on the inoculation of mice with mucin suspensions of tubercle bacilli, Mills and Colwell³ showed that guinea pigs inoculated sub-

cutaneously with small doses of tubercle bacilli suspended in mucin developed larger local lesions than controls inoculated with saline suspensions. These workers found no difference in pathogenicity when larger dosages were inoculated. More recently Pierce *et al.*² have shown that the infectivity of tubercle bacilli for mice is enhanced by egg yolk.

For the present studies CFW albino mice and C3H, dba and C57 black strains of pigmented mice weighing about 13 g were used. The virulent human type tubercle bacillus H37Rv or recently isolated strains from cases of tuberculosis were studied. Suspensions were prepared from 3-4-week-old cultures grown on Petragnini medium. The organisms were suspended in physiological saline and clumps were dispersed by means of glass beads. The suspension was adjusted to approximately 1 million organisms per cc by a nephelometer tube, and the count was checked with a Petroff-Hausser counting chamber. Mucin was prepared by suspending 5 grams of granular mucin (Type 1701-W, Wilson lab.) in 100 cc of distilled water, adjusted to pH 7.4 and autoclaved. Egg yolk was obtained aseptically and diluted with equal parts of physiological saline.

Results comparing the virulence of the H37Rv strain suspended in mucin, egg yolk

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¹ Long, E. R., and Vogt, A. B., *Am. Rev. Tuberculosis*, 1941, **44**, 196.

² Pierce, C., Dubos, R. J., and Middlebrook, G., *J. Exp. Med.*, 1947, **86**, 159.

³ Mills, M. A., and Colwell, C. A., *Am. Rev. Tuberculosis*, 1939, **40**, 109.

TABLE I.
Effect of Gastric Mucin, Egg Yolk, and Saline Diluents on Virulence of Tubercle Bacilli (H37Rv) for C3H and CFW (Albino) Mice.

Mouse strain	Diluent	No. of mice	No. with macroscopic lesions	% liver lesions	% peritoneal and lymph node lesions
C3H	Mucin	18	18	39	88
"	Egg yolk	18	10	40	60
"	Saline	16	0	0	0
CFW	Mucin	15	2	0	100
"	Saline	12	0	0	0

and saline for C3H and CFW (albino) mice are summarized in Table I. The mice were inoculated intraperitoneally with 0.5 cc of each mixture composed of equal parts of tubercle bacilli suspension and diluent. Each mouse was inoculated with approximately 500,000 bacilli. All animals were sacrificed 10 days after inoculation and autopsied for evidence of macroscopic lesions. Impression smears were routinely made of gross lesions and stained by the Ziehl-Neelsen technic. Lesions varied in size from 2 to 3 mm and usually contained large numbers of tubercle bacilli in impression smears. When present lesions were seen at the site of inoculation, liver, spleen or intestinal lymph nodes. A few tubercle bacilli were usually found in impression smears of the spleen or liver of mice inoculated with mucin suspensions, even if macroscopic lesions were not present in these organs. No gross lesions were seen in the lungs. Similar results were obtained using dba mice and recently isolated tubercle bacilli.

Since the above experiments were completed we have made several (14) isolations of tubercle bacilli from patients 10 to 15 days after inoculation by the use of pigmented mice (C3H or dba) and mucin. A few of the positive results in mice were obtained with microscopically negative sputum. The patient's sputum or feces were prepared in the usual manner, mixed with equal parts

of 5% mucin, and inoculated both subcutaneously and intraperitoneally (0.5 cc) into each of 4 or 5 mice. Tubercle bacilli were readily recovered from a number of the lesions planted in Petraghini's medium. Impression smears of the spleen, liver and peritoneal fluid were routinely made of all inoculated mice because acid-fast bacilli could sometimes be demonstrated although no macroscopic lesions were present. Comparative studies are now in progress in which routine specimens from patients are being inoculated into mice, guinea pigs and culture media simultaneously.

Summary. Gastric mucin significantly enhances the virulence of tubercle bacilli for pigmented strains (dba, C3H or C57 black) of mice. Grossly visible lesions containing large numbers of tubercle bacilli are usually present 10 days after intraperitoneal inoculation with the dosages and strains of tubercle bacilli studied. Several isolations of tubercle bacilli from patients have been made 10 to 15 days after inoculation by use of pigmented mice (dba or C3H) and mucin.

Addendum. Recently the number of isolations from patients has been increased by feeding pigmented mice inoculated with mucin the cornmeal-gelatin-butter diet of Dubos and Pierce.⁴

⁴ Dubos, R. J., and Pierce, C., *Am. Rev. Tuberculosis*, 1948, **57**, 287.