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Effect of Sulfapyridine (Dagenan) on *Brucella abortus* *in vitro* and *in vivo*.*

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In vitro. A virulent, aerobic strain of *Brucella abortus* was used in this experiment. Tryptose broth and Tryptose agar respectively were used for the *in vitro* experiment. Sulfapyridine was added to 100 cc of broth in flasks to give each of the following dilutions: 1:1,000; 1:10,000; and 1:100,000. Each dilution was made in triplicate. Three flasks of broth without sulfapyridine were left as controls. Sulfapyridine was added to the broth before autoclaving to obtain complete solution of the drug. All flasks of broth were inoculated with the same amount of dilute bacterial suspension. Petri plate counts made on each flask of broth at the beginning of the experiment averaged between 5,000 and 6,000 organisms per cubic centimeter. The inoculated broth was incubated at 37.5°C and Petri plate counts made in duplicate 10 minutes, 1, 2, 3, 5, and 7 days after addition of the organisms. The bacterial content of the inoculated broth after the various intervals of incubation was determined on dilutions of 1:100, 1:10,000 and 1:1,000,000 by placing 1 cc of each dilution in Petri plates in duplicate and adding 25 cc of Tryptose agar. The plates were incubated at 37.5°C for 96 hours and the number of colonies counted. Final counts were based on the averages of the three samples in each dilution group. The results of the final counts are shown in Table I.

TABLE I.
Number of *Brucella* Colonies per 1.0 cc of Broth After 96 Hours' Incubation at 37.5°C.

Conc. of sulfa- pyridine	Time after addition of <i>Brucella abortus</i>					
	10 min	1 day	2 days	3 days	5 days	7 days
1:1,000	5,000	36,800	64,400	34,500	1,900	230
1:10,000	5,300	163,400	123,600	103,100	3,700	250
1:100,000	5,900	180,000	320,000	2,900,000	12,300,000	28,600,000
Control	5,100	6,000,000	223,000,000	120,600,000	320,600,000	207,000,000

* Journal Article No. 372 n.s., Michigan Agricultural Experiment Station. The writers are indebted to Merek and Company, Inc., for the Dagenan used in these experiments.

The results of this experiment show that sulfapyridine has a definite bacteriostatic action on *Brucella abortus in vitro*. The decrease in the number of organisms is especially noticeable after 5 days in the 1:1,000 and 1:10,000 concentrations of sulfapyridine. After 7 days, there is no appreciable difference in the number of organisms in the 2 dilutions. In the 1:100,000 concentration, the decrease is less noticeable. The number of organisms in the control flasks did not vary appreciably after the second day of incubation.

In vivo. Eleven guinea pigs were used in this experiment. A newly isolated, anaerobic strain of *Brucella abortus* was suspended in sterile saline solution to a turbidity of I by the McFarland nephelometer and 0.5 cc injected subcutaneously into each guinea pig. All guinea pigs were weighed before injection. Six of the animals were divided into 3 groups of 2 each and 5 were left as controls. After an incubation period of 12 days the guinea pigs in Group I were given 6.7 mg at 11 a.m.; 6.7 mg at 1 p.m. and 3.4 mg at 3 p.m. of sulfapyridine for 10 consecutive days. Group II was treated in the same manner with the exception that the dosage was 50, 50, and 25 mg respectively. In Group III the dosage was 100, 100, and 50 mg. The sulfapyridine was suspended in water and administered orally.

The guinea pigs were re-weighed and killed 29 days after the initial dose of the drug was administered. The rapid agglutination test for brucellosis was conducted on the serum of all pigs. Duplicate cultures of the spleen, liver, and kidneys were made. All organs were examined for macroscopic lesions.

All 11 guinea pigs showed a positive reaction in a serum dilution of 1:400. *Brucella abortus* was isolated from one or more organs of all animals. Macroscopic lesions were present in the liver or spleen of all controls, and in one of the treated guinea pigs. There was a fairly uniform increase in the weight of all animals.

It is apparent that the per oral administration of sulfapyridine in large amounts over a period of 10 days had little, if any, effect on the course of *Brucella* infection.