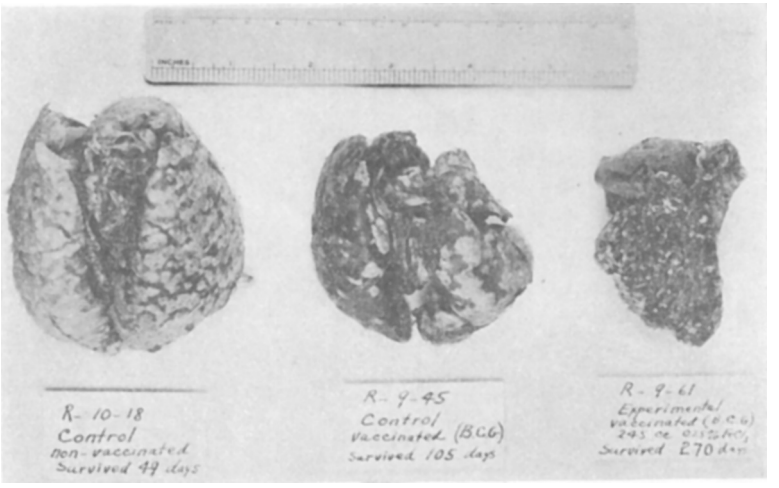

over 100 rabbits as summarized in Table II. The data show that dilute ferric chloride intravenously administered definitely retards the progress of tuberculosis in experimentally infected rabbits. The observations suggest possible clinical application which therefore the writer, with the permission of Dr. Donald S. King, has started at Channing Home, Boston. Several patients with far advanced tuberculosis received repeated intravenous injections of 0.0625% of isotonic ferric chloride for a few months. These injections proved completely harmless as far as noting any generalized deleterious effects. Small palpable thickenings occurred occasionally in injected arm veins. These invariably subsided within several days. The iron salt was evidently well tolerated in the concentration used.

8584 C

Fibrinolytic Activity of Beta Hemolytic Streptococci from Cow's Milk.

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One of the many unsettled problems concerning mastitis is its relationship to the organism designated as *S. mastitidis*. While the majority of workers agree that this relationship must be a very significant one, any new approach to the study of this subject should be of interest.

Tillet and Garner[1] found that many beta hemolytic streptococci from human diseases were capable of causing specific lysis of human plasma clots. Negative fibrinolytic tests were observed with other bacterial species from human sources, and strains which would actively lyse human plasma clots would not cause the dissolution of rabbit fibrin. Later Tillet[2] stated, "There is a relationship between the infectivity of beta hemolytic streptococci for man and the fibrinolytic property of the cultures."

Madison[3] confirmed the work of Tillet and Garner and[4] further tested strains from horse, rabbit, man, and hog. The lytic action of each strain was determined on fibrin from horse, hog, cow, rabbit, and man, but he did not test organisms from the cow on bovine plasma or fibrin. Thus, no conclusions could be drawn respecting the applicability of these tests to the study of *S. mastitidis*.

If the organism usually associated with infectious mastitis in the cow is able to lyse bovine fibrin, then fibrinolysis may offer a useful method of studying the rôle of this organism in mastitis.

Samples of milk were carefully taken from individual quarters: (a) udders were washed with 500 ppm. chlorine-solution; (b) first streams were discarded; (c) milk was drawn into sterile tubes, taken to the laboratory and held at 4°C. until plates could be made. Usually only 3 to 4 hours elapsed from the time of sampling to the time of plating, but in a very few cases the samples stood for 15 hours. Dilutions of 1/10 and 1/100 were plated on citrated horse-blood-agar, incubated for 48 hours at 37°C., when typical colonies were fished to blood-agar-slants.

Leukocytes were counted; unless an average of one per field (300,000 per ml.) was encountered no figures were tabulated. Chlorides were determined by the method of Hammer and Bailey,[5] and pH groupings were made according to the chart of Plastridge and Anderson,[6] using Brom Thymol Blue as the indicator.

If the leukocyte count exceeded 300,000 the milk was considered suspicious, while a count greater than 1,000,000 was tabulated as positive. A chloride content above 0.150% was looked on as either a doubtful or positive test for mastitis.

Fibrinolytic tests were made by the method of Tillet and Gar-

1 Tillet, W. S., and Garner, R. L., *J. Exp. Med.*, 1933, **58**, 485.

2 Tillet, W. S., *J. Bact.*, 1935, **29**, 111.

3 Madison, R. R., PROC. SOC. EXP. BIOL. AND MED., 1934, **31**, 1018.

4 Madison, R. R., PROC. SOC. EXP. BIOL. AND MED., 1934, **32**, 444.

5 Hammer, B. W., and Bailey, D. E., Iowa Agr. Exp. Sta. Res. Bul. 41, 1917.

6 Plastridge, W. N., and Anderson, E. O., Storrs Agr. Exp. Sta. Bul. 184, 1933.

TABLE 1.

Fibrinolytic Activity of Organisms Isolated from Bovine Udders.

Tests on Milk—Source of Culture								Reactions of Pure Culture					
								Fibrinolysis of bovine plasma					
Culture No.	Leukocyte count/ml.	Beta hemolytic count	Brom thymol blue grouping	Chlorides %	History of samples	Organism isolated	pH in dextrose broth	½ hr.	3 hr.	7 hr.	8½ hr.	13 hr.	24 hr.
1, 2, 3, 5	—	300	1	.132	A-1	Strep.	4.6	—	—	—	—	—	—
4	—	300	1	.132	A-1	”	4.6	—	—	—	+	+	+
6	—	20	1	.122	B-1	”	4.6	—	—	—	—	—	—
7	—	20	1	.122	B-1	”	4.6	—	—	+	+	+	2+
8	15,000,000	10	4	.270	C-1	Staph.	Above 6.0	—	—	—	—	—	—
9	300,000	40	3	.133	C-1	”	5.8	—	—	—	—	—	—
10	300,000	40	3	.133	D-1	”	5.8	—	—	—	—	—	—
11	6,000,000	300	4	.238	C-2	Strep.	4.6	—	—	—	—	—	—
12	6,000,000	300	4	.238	C-2	”	4.6	—	+	2+	3+	3+	3+
13	—	200	1	.151	D-2	”	4.6	—	—	—	—	—	—
14	—	200	1	.151	D-2	”	4.6	—	—	+	+	2+	2+
15	—	10	1	.126	A-2	Staph.	Above 6.0	—	—	—	—	—	—
16	600,000	100	1	.100	D-3	Coccus	4.6	—	—	—	—	—	—
17, 18, 20	—	70	1	.148	B-2	Strep.	4.6	—	—	—	—	—	—
19	—	70	1	.148	B-2	”	4.6	—	—	—	—	—	+
21	8,400,000	1,000	4	.123	C-3	”	4.6	—	—	—	—	—	±
22	8,400,000	1,000	4	.123	C-3	”	4.6	—	—	—	—	—	±
23, 24	6,000,000	3,300	4	.299	C-4	”	Above 6.0	—	—		—	—	—
25	6,000,000	3,300	4	.299	C-4	”	4.6	—	+	2+	3+	3+	3+
26	—	30	1	.130	A-3	”	Above 6.0	—	—	—	±	±	2+
27	—	50	1	.125	B-3	Coccus	5.8	—	—	—	—	—	—
28	—	50	1	.125	B-3	Strep.	4.6	—	—	—	—	±	+
29	—	30	1	.138	E-1	Coccus	Above 6.0	—	—	—	—	—	—
30	—	10	1	.130	F-1	”	5.8	—	—	—	—	—	—
31	—	20	1	.134	B-4	Strep.	4.6	—	—	—	—	+	+

ner, using a pure culture. All of the cultures used were isolated from milk which had been subjected to the above-mentioned tests, and all cultures were tested on bovine plasma.

The table giving the results of this work is divided into two parts: (a) that referring to the milk itself, its grouping and history; (b) that referring to the characteristics of the organisms isolated. Under "history," an arbitrary grouping of the milks was made as follows:

A. Normal milk*—negative 2-months' history.
B. Normal milk—mastitis in other quarter or quarters.
C. Mastitic milk—chronic type of mastitis.
D. Suspected of mastitis.
E. Normal milk—one or more other quarters suspected.
F. Normal milk—previously suspected.

Where the letters and numbers coincide the organisms are from the same milk sample.

While no autolytic test is listed in the table, we made this test and found it was negative.

Tests were made on 42 cows. In many cases more than one sample was taken from the same quarter. Samples which contained hemolytic streptococci were isolated from normal, suspected and mastitic milk. In regard to the normal samples (A-1, A-2) only one of these (A-1) contained hemolytic streptococci in appreciable numbers. However, cows listed in the table as suspected would probably have been listed as normal if we had not tested the samples from individual quarters, because an abnormal quarter might be masked by the other normal quarters, as measured by our non-specific tests, since, as can be seen from our arbitrary definition of normal, an equivocal test could quite easily be altered to conform to these standards.

Nine of the 23 strains of streptococci showed varying degrees of fibrinolysis of bovine plasma. Of these, 2 were from normal cows, 5 from cows having mastitis in one other quarter and 2 from animals with chronic mastitis.

Some of the strains were isolated from the same sample; however, the fibrinolytic activities of these strains sometimes differed. In one case only one of 5 strains, from the same source, was capable of causing the lysis of the plasma clot.

The few organisms other than streptococci were not fibrinolytic.

*In this article the word "normal" is arbitrarily used to designate milk having not over 300,000 leukocytes per ml., giving a No. 1 B.T.B. grouping, and having a chloride test of 0.150% or less.

The fact that all strains of streptococci from the same source were not able to lyse the clot is interesting. If this test indicates virulence, we must admit that both virulent and avirulent types of beta hemolytic streptococci may coexist in the cow's udder. This point might be proven by further studies since in this work cows considered as mastitic were probably suffering from a chronic type of mastitis. In acute cases, one would expect a much higher percentage of fibrinolytic strains if there is a correlation between fibrinolysis and virulence.

The occurrence of lytic strains in cows which we class as normal does not deny the possibility that the test is of value as an indicator of virulence. Ayers and Mudge[7] isolated *S. mastitidis* from cows they believed to be normal. Plastridge and Anderson[8] do not believe these cows are normal, but that they merely temporarily fail to give the usual non-specific tests. These cows, however, may often harbor streptococci, which, possibly, may be virulent.

There is considerable evidence that *S. mastitidis* causes bovine mastitis; still there is the possibility that it is merely an opportunist. By analogy with properties of human streptococci, we do think there is some possibility that fibrinolysis of bovine plasma, by beta hemolytic streptococci from bovine udders, may be a measure of pathogenicity.

Conclusions: 1. Some beta hemolytic streptococci from milk are capable of causing the lysis of clotted bovine plasma. 2. Organisms from cows that are negative to non-specific tests for mastitis may produce this fibrinolysis.

8585 P

Growth Inhibitor in Liver Tissue.

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Liver has been known for a long time to contain substances which inhibit growth and migration of cells from explants. Walton[1] observed that extracts of liver had an inhibiting effect on tissue cul-

7 Ayers, S. H., and Mudge, C. S., *J. Infect. Dis.*, 1922, **31**, 40.
8 Plastridge, W. N. and Anderson, E. O., Storrs Agr. Exp. Sta. Bul. 195, 1934.
1 Walton, A. J., *J. Exp. Med.*, 1914, **20**, 554.