

As may be seen from Table I, a 2% suspension of zinc oxide, 3% suspension of zinc peroxide, and 4% solution of sodium perborate, all in physiological saline, inhibit gelatin liquefaction by the above strains of microorganisms far more effectively than gelatin liquefaction resulting from the action of fecal specimens in suitable dilutions. Similarly, these chemical compounds failed to inhibit gelatin liquefaction by a 1:50 saline solution of crystalline trypsin, whereas the soybean trypsin inhibitor completely inhibited this solution. The other compounds tested were less satisfactory for the differentiation between bacterial and pancreatic gelatin liquefying enzymes.

Discussion and summary. In order to render the x-ray film liquefaction test by

fecal specimens more informative it is highly desirable to be able to differentiate between gelatin liquefaction due to the activity of pancreatic trypsin and that due to the action of bacterial enzyme. Soybean trypsin inhibitor, which prevents the liquefaction of gelatin (x-ray film) by crystalline trypsin and trypsin in feces, fails to inhibit gelatin liquefaction by various bacteria, including *Proteus ammoniae*, *Proteus morganii*, and *Pseudomonas aeruginosa*. Suitable preparations of zinc oxide, zinc peroxide, and sodium perborate have no or only a slight effect on trypsin in feces but markedly inhibit the bacterial enzyme responsible for gelatin liquefaction.

Received April 27, 1950. P.S.E.B.M., 1950, v74.

Relative Efficacy upon *Pasteurella multocida* of Various Antibiotics Aureomycin, Terramycin, Bacitracin, and Polymyxin B. (17894)

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Recently a strain of *Pasteurella multocida* was isolated from an abscess at the nose of a boy 15 years of age. The infection developed subsequent to a plastic operation undertaken to correct a deformity resulting from "a kick of a horse to the nose". Since this pathogen could be demonstrated repeatedly over a period of 6 months and other disease-producing microorganisms were not recovered, the conclusions are drawn that the infection was caused by the pasteurella organism, was of animal origin, and had remained latent until activated by the operation. That human infections with this microorganism have been reported only infrequently is evident from a review of the world literature by Schipper(1), who collected but 39 authentic cases described between 1930 and 1947. Because of the lack of data on the effectiveness of some of the newer antibiotics upon this

microbe, a study was undertaken to determine the relative efficacy of 7 antibiotics upon 8 strains of *P. multocida*, including the one isolated from the above patient. The results are embodied in this report.

Materials and methods. The following strains of *P. multocida* were employed:

No.	Strain	Source
200	<i>P. avicida</i>	*
1	Bovine	†
2	Unknown	†
3	Canine	†
9656	<i>P. oviseptica</i>	‡
9658	" "	‡
10544	From sputum	‡
4392	" abscess	§

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† Iowa State College (Dr. Merchant).

‡ American Type Culture Collection.

§ Children's Hospital, Buffalo.

The strains were maintained on blood agar and were transplanted twice a week. Six antibiotics were employed. Penicillin-G, crvs-

1. Schipper, G. J., *Johns Hopkins Hosp. Bull.*, 1947, v81, 333.

TABLE I.
Minimal Growth Inhibitory Concentrations of Sulfadiazine and Various Antibiotics on *Pasteurella multocida*.

Drug	Strain							
	1	2	3	200	9656	9659	4392	10544
Sulfadiazine, mg %	.003-10	.01-10	.01-10	.01-10	.03-10	.03-10	.01-10	.03-10
Penicillin, units %	.03-1	.1-1	.01-1	.03-3	.1-1	.1-1	.3-1	.03-10
Streptomycin, μ g %	1-10	1-10	10-10	10-10	1-10	1-10	10-10	10-10
Aureomycin, μ g %	.01-1	.1-1	.1-3	.01-1	.25-1	.1-1	.01-0.1	.3-1
Chloromycetin, μ g %	.1-1	.3-1	.3-1	.01-1	.3-1	.3-1	1-10	.3-1
Bacitracin, units %	10-100	3-100	10-100	10-100	10-30	3-100	.3-30	10-100
Polymyxin, μ g %	.01-5	.1-3	.03-3	.03-1	.003-3	.0001-3	.1-10	.1-10
Terramycin, μ g %	.3-1	.3-1	.3-1	.3-1	.3-1	1-3	.1-1	.3-1

talline-potassium, dihydro-streptomycin hydrochloride, and bacitracin were procured from commercial sources. Aureomycin hydrochloride (intravenous) was made available through the courtesy of Dr. B. W. Carey of Lederle Laboratories; pure crystalline chloramphenicol (chloromycetin) was kindly supplied by Dr. J. P. Gray of Parke, Davis and Co.; polymyxin B sulfate ("Aerosporin") by Dr. D. S. Searle of Burroughs Wellcome & Co. and terramycin hydrochloride by Dr. Elliott R. Weyer of Chas. Pfizer & Co. The drugs were dissolved in various concentrations in brain heart infusion (volume 1.8 ml) and, together with broth used as control, seeded with 24 hours broth cultures of the above strains. The size of the inocula was 0.2 ml and contained either 1 to 100 million organisms per ml or 100 to 1,000 organisms per ml. The tubes were incubated at 37°C and the resulting growth was noted at regular intervals.

Results. The minimal amounts of the 7 antibiotics capable of suppressing visible growth for 24 hours of each of the 8 strains of *P. multocida* was determined and the results were summarized in Table I. At least 6 experiments were carried out for each drug concentration and for each strain. The first figure given for each drug concentration is the one obtained in tests using small inocula and the second figure the one observed with large inocula of the particular strain.

Discussion. Studies on the antibacterial activity of several antibiotics against *Pasteurella multocida* have been reported previously, but different technics and different strains were used in each investigation. It was

deemed of interest, therefore, to determine the relative efficacy of these antibiotics, using a single method and identical strains, and to explore the efficacy of 3 antibiotics, aureomycin, bacitracin, and terramycin whose antibacterial activity against this species has not been studied before. It is evident from Table I that aureomycin is highly effective against all 8 strains of *P. multocida*. The strain (No. 4392) isolated from the above mentioned patient was sent to Lederle Laboratories and proved to be the most susceptible gram-negative organism Dr. Dornbush (2) has encountered up to that time. Terramycin, which was described recently by Finlay and associates(3) as an antibiotic of potential clinical usefulness, also inhibits the growth of this species. *P. multocida* appears to be as highly or more susceptible to this antibiotic as the various bacterial species tested by Finlay *et al.* although it must be realized that different methods were employed in these investigations. Bacitracin was found to be relatively ineffectual against *P. multocida*, particularly when one calculates the amounts of drug necessary to inhibit growth. Based on the fact that 100 units of the preparation used represent approximately 2,000 μ g it is evident that, with the exception of one strain, from 70 to 2,000 μ g/ml was required to effect bacteriostasis. This finding is not surprising in view of the fact that

2. Dornbush, A. C., personal communication, 12/2/49.

3. Finlay, A. C., Hobby, G. L., P'an, S. Y., Regna, P. P., Routien, J. B., Seeley, D. B., Shull, G. M., Sobin, B. A., Solomons, I. A., Vinson, J. W., and Kane, J. H., *Science*, 1950, v111, 85.

bacitracin is largely effective against gram-positive microorganisms, but it is interesting to point out that penicillin, whose spectrum of antibacterial activity somewhat resembles that of bacitracin, was found to be highly effective against *P. multocida*, as may be seen from Table I. That penicillin inhibits growth of this species was previously reported by Schipper(1) and by Queen and Quortrop(4). In view of the fact that *P. multocida* is a gram-negative rod of the family *Parvobacteriaceae* which includes, among others, the members of the genus *Hemophilus* it is somewhat surprising to find that 8 strains of *P. multocida* are rather resistant to streptomycin, as may be seen from Table I. Coles(5) studied the sensitivity of 78 strains of *P. multocida* by the agar plate method and found that for complete inhibition all but seven strains required 1 μ g or more and that none of the strains was inhibited by less than 0.5 μ g of streptomycin.

The above reported experiments confirm the observation of McLean, Schwab, Hille-

gas and Schlingman(6) and of White, Alverson, Baker and Jackson(7), to the effect that both chloromycetin and polymyxin are effective against *P. multocida*. The minimal effective concentration of polymyxin and "Aerosporin", according to White and associates(7), was 1 μ g/ml. It may be pointed out that as little as 0.0001 to 0.03 μ g/ml was adequate to prevent growth of small inocula of five of the strains of *P. multocida* used in this study.

Summary. The bacteriostatic activity toward eight strains of *Pasteurella multocida* of aureomycin, terramycin, and bacitracin and the relative efficacy of four additional antibiotics, previously studied only singly, have been determined. It was found that the eight strains were very susceptible to penicillin, aureomycin, chloromycetin, terramycin and polymyxin B but rather resistant to bacitracin and streptomycin.

6. McLean, I. W., Jr., Schwab, J. L., Hillegas, A. B., and Schlingman, A. S., *J. Clin. Invest.*, 1949, v28, 953.

7. White, H. J., Alverson, C. M., Baker, M. J., and Jackson, E. R., *Ann. N. Y. Acad. Sci.*, 1949, v51, 879.

4. Queen, F. B., and Quortrop, E. R., *J. A. Vet. M. A.*, 1946, v108, 101.

5. Coles, E. H., Jr., *A. J. Vet. Res.*, 1948, v9, 132.

Received April 27, 1950. P.S.E.B.M., 1950, v74.

Eosinophil Response to Epinephrine and Nor-epinephrine. (17895)

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It has been demonstrated repeatedly that a variety of stress conditions(1), adrenocorticotrophic hormone, adrenal cortical hormone and epinephrine(2,3), produce a pronounced reduction in the number of circulating eosino-

phils. This response has been shown to depend on the stimulation of the adrenal cortex. The fall in circulating eosinophils is so constant and sensitive an indicator of oxycorticosteroid release that it has been incorporated into a standard test for adrenal cortical function(4). For this purpose, either ACTH or epinephrine has been used. Nor-epinephrine†

*Sponsored by Veterans Administration and published with approval of Chief Medical Director. The statements and conclusions published by the authors are a result of their own study and do not necessarily reflect opinion or policy of Veterans Administration.

1. Dalton, A. J., and Selye, H., *Folia Haemat.*, 1939, v62, 397.

2. Laragh, J. H., and Almy, T. P., *Proc. Soc. Exp. Biol. and Med.*, 1948, v69, 499.

3. Hills, A. G., Forsham, P. H., Finch, C. A., *Blood*, 1948, v3, 755.