10511

# Further Studies on Therapeutic Properties of Sulfapyridine* in Experimental Pneumococcus Infections.†

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An earlier report[1] showed that sulfapyridine (2-sulfanilamidopyridine) had a curative action when administered to mice infected with Type XXII pneumococci. This pneumococcus was one of 11 types which had been shown[2] to be refractory to sulfanilamide therapy. The greater effectiveness of sulfapyridine noted above, and a similar finding by Whitby[3] in Type I infections, suggested that this drug might have curative properties in infections with other types of pneumococci which were refractory to sulfanilamide therapy.

The present report concerns the therapeutic properties of sulfapyridine in experimental infections with pneumococci Types I, II, III, IV, V, VI A and B, VII, VIII, XI, XX, XXIV, XXVII and XXIX. These pneumococci include 10 of the 11 types which were refractory to sulfanilamide therapy and in addition Types I, V and VII which responded favorably.[2]

Groups of white mice were infected intraperitoneally with 100 to 1000 lethal doses of a given type pneumococcus.‡ Some of these mice served as untreated controls. The remainder received sulfapyridine 2 hours after infection and at repeated intervals thereafter according to one of the following schedules: *A*—20 mg sulfapyridine 2, 8, 14 and 20 hours after infection and every 24 hours thereafter for 5 days—least intensive therapy; *B*—20 mg of sulfapyridine

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1 Hilles, C., and Schmidt, L. H., PROC. SOC. EXP. BIOL. AND MED., 1939, **40**, 73.

2 Schmidt, L. H., and Hilles, C., in press.

3 Whitby, L. E. H., *Lancet*, 1938, **1**, 1210.

‡ For stock cultures of these pneumococci we are indebted to Miss Annabel Walter, Bureau of Laboratories, Department of Health, New York City, and to Dr. Mary Kirkbride, New York State Department of Health, Albany. All of the pneumococci used in this work were of highest virulence; 1 to 8 pneumococci constituted a lethal dose and this number of organisms was invariably contained in $10^{-8}$ or $10^{-9}$ cc of a blood broth culture incubated for 12 hours at 37°C. The technique of increasing the virulence and determining the lethal dose has been described elsewhere.[2]

every 6 hours for 4 days and then every 12 hours for 2 additional days—treatment used if *A* failed to cure 50% of the infected mice; *C*—20 mg of sulfapyridine every 6 hours for 7 days and then every 12 hours for 3 additional days—most intensive therapy, used if *B* failed to cure 50% of the infected mice. In all experiments the drug was administered orally as a 10% suspension in 10% acacia. At the conclusion of treatment cultures of tail blood were made. Mice were considered cured if they survived 30 days after infection and had negative heart blood cultures at the end of that time.

Table I summarizes the results in infections with the 13 types of pneumococci mentioned above and with Type XXII.[1] The experiments shown are those in which the best therapeutic response has been obtained. Other experiments with all of these 14 types of pneumococci, except I and VII, have shown that poorer results are obtained when mice receive less intensive treatment than that shown in the table; mice that received sulfapyridine always lived longer than untreated controls, but few of the treated mice recovered. For comparison Table I includes a summary of the results of sulfanilamide therapy in infections with the same strains of pneumococci (data from earlier experiments[2]).

Table I shows that sulfapyridine was most effective in Types I and VII infections; the least intensive of the various treatments (A) led to the recovery of all the infected mice. The drug was next most effective in infections with Types IV, V, VI A and B, XI, XXII, and XXIX, but in order to effect cures in 50% or more of the mice, intensive therapy (B) had to be maintained for 4 days. Types XX, XXIV, and XXVII infections were more refractory to treatment than any of the above, but 30% of the mice infected with Type XX and 50 and 85% of those infected with Types XXIV and XXVII, respectively, recovered when intensive sulfapyridine therapy (C) was continued for 7 days.

Sulfapyridine was least effective in infections with pneumococci Types II, III, and VIII.§ Although the most intensive therapy (C) prolonged life, it did not lead to the recovery of a significant number of animals. As judged by its life-prolonging action, the drug had a greater effect in Type II infections than in either Types III or VIII. Fourteen of the 20 mice, infected with Type II pneumococci and treated with sulfapyridine, survived the 10-day period of therapy;

§ It is worth noting that the preliminary clinical study of Flippin and his coworkers[4] indicated that Type III infections responded least well to sulfapyridine therapy.

[4] Flippin, H. F., Lockwood, J. S., Pepper, D. S., and Schwartz, L., *J. A. M. A.*, 1939, **112**, 529.

TABLE I.
Therapeutic Effectiveness of Sulfapyridine in Experimental Pneumococcus Infections.

| Type of pneumococcus | Infecting Dose: No. of lethal doses | Infecting Dose: cc of 12-hr culture | No. of mice in group | Sulfapyridine treatment | No. of Deaths Days After Infection: 1-5 | 6-10 | 11-15 | 16-30 | Survivors: No. | % | Results of Sulfanilamide Therapy† Avg surival time, Days |
|---|---|---|---|---|---|---|---|---|---|---|---|
| I | 100 | $10^{-7}$ | 24 | A* | 0 | 0 | 0 | 0 | 24 | 100 | (67 % of mice recovered) |
| | ,, | ,, | 20 | 0 | 20 | 0 | 0 | 0 | 0 | 0 | |
| II | 100 | $10^{-6}$ | 20 | C* | 1 | 5 | 13 | 0 | 1 | 5 | 9.4 |
| | ,, | ,, | 20 | 0 | 20 | 0 | 0 | 0 | 0 | 0 | |
| III | 100 | $10^{-6}$ | 20 | C* | 1 | 17 | 0 | 0 | 2 | 10 | 4.4 |
| | ,, | ,, | 20 | 0 | 20 | 0 | 0 | 0 | 0 | 0 | |
| IV | 1000 | $10^{-6}$ | 20 | B* | 1 | 7 | 0 | 0 | 12 | 60 | 9.8 |
| | ,, | ,, | 10 | 0 | 10 | 0 | 0 | 0 | 0 | 0 | |
| V | 100 | $10^{-6}$ | 20 | B* | 0 | 4 | 3 | 0 | 13 | 65 | (67 % of mice recovered) |
| | ,, | ,, | 10 | 0 | 10 | 0 | 0 | 0 | 0 | 0 | |
| VIA | 100 | $10^{-6}$ | 20 | B* | 1 | 4 | 0 | 0 | 15 | 75 | 11.7 |
| | ,, | ,, | 20 | 0 | 20 | 0 | 0 | 0 | 0 | 0 | |
| VIB | 100 | $10^{-7}$ | 20 | B* | 0 | 2 | 0 | 0 | 18 | 90 | 12.2 |
| | ,, | ,, | 20 | 0 | 20 | 0 | 0 | 0 | 0 | 0 | |
| VII | 1000 | $10^{-6}$ | 20 | A* | 0 | 0 | 0 | 0 | 20 | 100 | (92 % of mice recovered) |
| | ,, | ,, | 10 | 0 | 10 | 0 | 0 | 0 | 0 | 0 | |
| VIII | 100 | $10^{-6}$ | 20 | C* | 0 | 16 | 1 | 0 | 3 | 15 | 7.0 |
| | ,, | ,, | 20 | 0 | 20 | 0 | 0 | 0 | 0 | 0 | |
| XI | 100 | $10^{-7}$ | 20 | B* | 3 | 2 | 0 | 0 | 15 | 75 | 12.2 |
| | ,, | ,, | 20 | 0 | 20 | 0 | 0 | 0 | 0 | 0 | |
| XX | 1000 | $10^{-6}$ | 20 | C* | 0 | 4 | 10 | 0 | 6 | 30 | 9.2 |
| | ,, | ,, | 20 | 0 | 20 | 0 | 0 | 0 | 0 | 0 | |
| XXII | 100 | $10^{-6}$ | 20 | B* | 4 | 2 | 0 | 0 | 14 | 70 | 7.6 |
| | ,, | ,, | 20 | 0 | 20 | 0 | 0 | 0 | 0 | 0 | |
| XXIV | 1000 | $10^{-6}$ | 20 | C* | 1 | 3 | 6 | 0 | 10 | 50 | 11.4 |
| | ,, | ,, | 20 | 0 | 20 | 0 | 0 | 0 | 0 | 0 | |
| XXVII | 100 | $10^{-7}$ | 20 | C* | 0 | 2 | 1 | 0 | 17 | 85 | 7.3 |
| | ,, | ,, | 20 | 0 | 20 | 0 | 0 | 0 | 0 | 0 | |
| XXIX | 1000 | $10^{-6}$ | 20 | B* | 1 | 4 | 0 | 0 | 15 | 75 | 7.8 |
| | ,, | ,, | 10 | 0 | 10 | 0 | 0 | 0 | 0 | 0 | |

*A. 20 mg sulfapyridine 2, 8, 14 and 20 hours after infection and every 24 hours thereafter for 5 days.
B. 20 mg sulfapyridine 2 hours after infection and every 6 hours thereafter for 4 days and then every 12 hours for 2 days.
C. 20 mg sulfapyridine 2 hours after infection and every 6 hours thereafter for 7 days and then every 12 hours for 3 days.

†Results with most effective treatment: 10 mg sulfanilamide, subcutaneously, 2 hours after infection and every 6 hours thereafter for 8 days and then every 12 hours for 2 days.

these animals were apparently in good health at the conclusion of treatment; 13 of them had positive blood cultures, however, and died between the 11th and 16th days after infection.

Our observations in Type VIII infections do not support Whitby's conclusion[3] that sulfapyridine is especially effective in infections with this pneumococcus. The discrepancy might have resulted from the use of different strains of organisms; more probably it was due to Whitby having terminated his experiments (those with Type I excepted) 7 days after infection. Our experiments show that this period of observation is too short to warrant a conclusion of curative action unless supported by data on blood cultures; at least 80% of our mice infected with pneumococci Types II, III, and VIII survived 7 days, but very few have recovered.

Table I also shows that sulfapyridine is a more effective therapeutic agent than sulfanilamide in infections with all of the 14 types of pneumococci investigated. A similar observation had been made by others[3, 5] in Types I and II infections. It is interesting to note that infections with Types I and VII responded most readily to both sulfapyridine and sulfanilamide, and infections with Types III and VIII were the most difficult to treat. In view of the similarity in the relative effectiveness of the 2 drugs, one may predict that infections with the remaining 16 types of pneumococci will respond favorably to sulfapyridine therapy—since these types responded to sulfanilamide as did Types I and VII.[2]

The fact that sulfapyridine therapy was not equally effective in infections with all types of pneumococci seems especially important in so far as the clinical use of the drug is concerned. It suggests that infections with certain types of pneumococci may require much more intensive treatment than infections with other types, and that infections with some types may be refractory to sulfapyridine. If these suggestions are correct, it will be particularly important to evaluate the clinical usefulness of the drug on the basis of results obtained with each of the 30 types of pneumococci.

*Summary.* Experiments have shown that sulfapyridine has a curative action when administered to mice infected with pneumococci Types I, IV, V, VI A and B, VII, XI, XX, XXII, XXIV, XXVII, and XXIX. The drug has little curative action in infections with Types II, III, and VIII, although it does prolong life.

[5] Cooper, F. B., Gross, P., and Lewis, M., PROC. SOC. EXP. BIOL. AND MED., 1939, **40**, 87.