skin reactions when employed in the undiluted state. No reaction was elicited when ground agar removed from different depths of the slant was tested.

To determine the optimal time for incubation of cultures, a series of 7 tubes was prepared by inoculating chick embryo with an infected chorio-allantoic membrane. One tube was immediately placed in the icebox, and the remaining 6 tubes were incubated for 3, 4, 5, 7, 9, and 11 days, respectively. Titration of all of these cultures, performed upon the same rabbit, showed the titer of 1:1,000 for the first tube, and of 1:10,000, 1:10,000, 1:10,000, 1:1,000, 1:0 and 1:0 for the remaining 6 cultures. The experiment showed that the multiplication of the virus took place during the first 5 days. Deterioration of potency observed on further incubation was due, in all probability, to the effect of incubator temperature. This is suggested by the fact that cultures grown for 5 days and then kept in the icebox showed no decrease in their titer during the first 3 weeks of storage.

Summary. The cultivation of the vaccine virus in agar-slant tissue cultures was shown to be possible both with chick and mouse embryo tissues. The former was found to be distinctly superior so far as the growing qualities of the tissue are concerned.

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Toxic Manifestations After Oral Administration of Sodium Sulfapyridine.

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In 2 previous papers[1, 2] we have reported the occurrence of uroliths consisting mainly of 2-(acetylsulfanilyl amino) pyridine, in rats, rabbits and monkeys following oral administration of sulfapyridine. Similar results were also published by Gross, Cooper and Lewis.[3] Marshall, *et al.*,[4] have shown that sulfapyridine in the form of its soluble sodium salt is more rapidly and completely absorbed than sulfapyridine, and have recommended its intravenous injection in

1 Antopol, W., and Robinson, H., PROC. SOC. EXP. BIOL. AND MED., 1939, **40**, 428.

2 Molitor, H., and Robinson, H., *Arch. Internat. de Pharm. et Ther.*, in press.

3 Gross, P., Cooper, F., and Lewis, M., PROC. SOC. EXP. BIOL. AND MED., 1939, **40**, 448.

4 Marshall, E. K., Bratton, A., and Litchfield, J., *Science*, 1938, 597.

patients in whom administration of sulfapyridine by mouth is impossible, or intestinal absorption is very poor, or prompt action of the drug is imperative.[5] Since sodium sulfapyridine is excreted both as free and acetylated sulfapyridine it might be expected that its administration would result in a greater incidence of urolithiasis. The strong alkalinity of the compound also suggests the possibility of undesirable local effects depending upon the method of administration.

In the present paper we wish briefly to report the results of a series of experiments in which 10% aqueous solutions of sodium sulfapyridine (pH 11.4) were administered by stomach tube to 160 mice, 150 rats, 24 rabbits and 21 monkeys. Single doses of 3 and 4.5 g per kg produced gastric congestion and irritation in mice, rats and rabbits, as well as frequent urolithiasis in rats and rabbits. These phenomena became more pronounced after repeated administration (daily feeding for 10 days). In monkeys 0.5 g per kg or more produced gastric congestion with occasional erosions and marked urolithiasis in 8 out of 10 animals; 0.25 g per kg resulted in urolithiasis in 3 out of 5 animals, whereas no concretions were found with 0.1 g per kg. Rats fed doses of 2 and 3 g per kg lost weight and showed a general debility. Hematuria was frequent after the third day and all animals dying during the feeding period showed renal calculi and a severe hydronephrosis. Urolithiasis was also found in 13 out of 14 rats which survived the 10-day feeding period and were sacrificed on the 11th day. The degree of changes in the urinary tract of rats, rabbits and monkeys varied from mild dilatation of the ureters with little or no change in the kidney to an enormous dilatation of the ureter and kidney with partial disappearance of the kidney substance.

As is the case with sulfapyridine, no evidence of urolith formation was found in mice.*

Summary. Oral administration of sodium sulfapyridine produces marked gastric irritation and results in rats, rabbits and monkeys in the formation of urinary concretions. The minimal doses necessary to produce these phenomena vary from 0.25 g per kg in monkeys to 2 g per kg in rats, administered daily for 10 days. Larger doses (3 to 4.5 g per kg) may cause urolithiasis even after a single administration.

[5] Marshall, E. K., and Long, P. H., *J. Am. Med. Assn.*, 1939, **112**, 1671.

* However, since submission of this paper we have been able to produce regularly urolithiasis also in mice by oral or intravenous administration of acetylated sulfapyridine in the form of its sodium salt.