POLYVINYLPYRROLIDONE AS A DRUG RETARDANT

PART III. EFFECT ON SODIUM *p*-AMINOSALICYLATE

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CLINICAL evidence has been accumulated by Weiss, Seven and Eisenberg[1], demonstrating the improved blood levels of *p*-aminosalicylate when the drug is administered intravenously with polyvinylpyrrolidone. While *p*-aminosalicylate is usually given orally, gastrointestinal intolerances sometimes indicate its intravenous use. In such instances the reported prolonged retention of effective blood concentrations and the increase in maximum blood levels of the drug when given with polyvinylpyrrolidone suggest that this is a distinct advance in therapeutics.

As part of a broader study of the drug retardent action of polyvinylpyrrolidone[2,3], the combination of *p*-aminosalicylate with polyvinylpyrrolidone was investigated in rabbits and rats. This investigation was particularly desirable in view of the earlier report of Bertolani and Farinetti[4] which indicated that the intravenous injection of polyvinylpyrrolidone had little or no effect on the blood and urine concentrations of *p*-aminosalicylate given orally or intravenously.

EXPERIMENTAL METHODS AND RESULTS

Male and female rabbits used in these experiments were of mixed breeding and weighed 1·6 to 2·8 kg. Samples of free-flowing blood were taken from the ear vein just before and at intervals after the injection of *p*-aminosalicylate solutions. Measured aliquots of the blood, usually 0·2 ml., were added immediately to sufficient distilled water to give a total volume of 7·0 ml. Protein was removed by adding 3·0 ml. of 20 per cent. trichloracetic acid solution and filtering, after at least 5 minutes, through Whatman No. 42 filter paper. *p*-Aminosalicylate determinations on 5·0 ml. or smaller aliquots of the filtrate were carried out by the method of Klyne and Newhouse[5] using *p*-dimethylaminobenzaldehyde reagent (2 per cent. solution in 95 per cent. ethanol). If aliquots smaller than 5·0 ml. were used they were diluted to 5·0 ml. by the addition of 6 per cent. trichloracetic acid solution. After the addition of 1·0 ml. of citrate buffer (39·4 g. of citric acid dissolved in 100 ml. of 2 N sodium hydroxide and diluted to 250 ml. with distilled water) and 2·0 ml. of *p*-dimethylaminobenzaldehyde reagent, the percentage transmission of the coloured solutions was read at 420 mμ in the Evelyn colorimeter. The blank consisted of 5·0 ml. of 6 per cent. trichloracetic acid solution, 1·0 ml. of citrate buffer, and 2·0 ml. of *p*-dimethylaminobenzaldehyde reagent.

In the experiments in which rats were used, the analytical procedure was the same. Blood samples were obtained from the cut neck veins

of the rat when killed. Aliquots (0·3 ml.) were added at once to 6·7 ml. of distilled water without the use of an anticoagulant. The rats used were all male albinos and all within the weight range 130 to 180 g.

In some of the earlier experiments the *p*-aminosalicylate solutions for injection were prepared by suspending *p*-aminosalycylic acid in distilled water and adjusting the *p*H to approximately 6 with 2 N sodium hydroxide solution before diluting to the final volume. In the later experiments sodium *p*-aminosalicylate was used. Usually the salicylate derivative was made up at 5 times the required concentration and diluted with 4 volumes of polyvinylpyrrolidone solution to the required strength.

Rabbits were dosed in groups of 5, one rabbit receiving each of the 10 per cent. sodium *p*-aminosalicylate solutions containing 0, 5, 10, 20, or 40 per cent. polyvinylpyrrolidone. These solutions were injected at the rate of 2·0 ml./kg. of body weight. Blood samples were taken for analysis at 0 minutes and 20, 40, 60, 90, 120 and 180 minutes after injection. The analytical values on blood samples obtained after injection of the drugs were corrected by subtraction of the appropriate 0 time level. The results of 5 such experiments involving 25 rabbits are shown in Table I.

TABLE I

EFFECT OF POLYVINYLPYRROLIDONE ON THE BLOOD LEVELS AND RETENTION OF *p*-AMINOSALICYLATE IN RABBITS

Polyvinyl-pyrrolidone per cent.	Blood level of *p*-aminosalicylic acid μg./ml. of blood ± standard error					
	Minutes after injection (intravenous)					
	20	40	60	90	120	180
0	384 ± 9	236 ± 31	112 ± 14	49 ± 12	14 ± 5	trace*
5	378 ± 11	237 ± 33	87 ± 9	36 ± 9	10 ± 2	"
10	403 ± 21	225 ± 39	95 ± 24	43 ± 15	14 ± 3	"
20	402 ± 32	190 ± 27	62 ± 16	27 ± 7	7 ± 2	"
40	217 ± 39	119 ± 13	40 ± 14	24 ± 6	13 ± 4	"

* The corrected values at 180 minutes usually (22 cases) were less than 50 per cent. of the control level and are considered to be of very doubtful validity.

These data were subjected to appropriate application of the "t" test and it was found that the blood level of *p*-aminosalicylate at 20, 40, or 60 minutes after injection of the drug in 40 per cent. polyvinylpyrrolidone was significantly lower than the control. By 90 minutes after injection this difference was lost. After 120 minutes the blood level of *p*-aminosalicylate was very low and in 16 of 25 rabbits was zero at 180 minutes.

Most of the clinical investigations employed 12·5 to 25 per cent. polyvinylpyrrolidone. Since the clinical data of Weiss *et al.*[1] were obtained using 3·5 per cent. of polyvinylpyrrolidone, additional tests on rabbits were made to determine whether or not concentrations of polyvinylpyrrolidone less than 5 per cent. might be effective. The results with 3·5 per cent. of polyvinylpyrrolidone were essentially the same as those obtained with 0 or 5 per cent. of the retardant.

On the chance that the plasdone* brand of polyvinylpyrrolidone used in most of the experiments reported here might fail as a retardant because

* Plasdone was supplied by General Aniline and Film Corporation. K (Fikentscher) (1 per cent. solution) 30 ± 2.

of its chemical or physical properties, experiments using subtosan† brand of polyvinylpyrrolidone were conducted. The results, regardless of concentration over the range 2·6 to 40 per cent., were indistinguishable from those obtained with plasdone.

In the experiments with rats the *p*-aminosalicylate (10 per cent.) in aqueous polyvinylpyrrolidone solutions (0 to 40 per cent.) were injected intraperitoneally at the rate of 5·0 ml./kg. The injections were made at carefully timed intervals and the rats were killed 25, 50, 75, 125, or 200 minutes later. In each of two identical experiments 5 rats on each dose were killed at each of the 5 intervals. The results of the chemical determinations were subjected to an analysis of variance which indicated that there was no significant difference between the results of the two experiments.

The analytical data for the pooled experiments are presented in Table II.

TABLE II

EFFECT OF POLYVINYLPYRROLIDONE ON THE BLOOD LEVELS AND RETENTION OF *p*-AMINOSALICYLATE IN RATS

Polyvinyl-pyrrolidone per cent.	Blood level of *p*-aminosalicylic acid μg./ml. of blood ± standard error				
	Minutes after injection (intraperitoneal)				
	25	50	75	125	200
0	857 ± 29	693 ± 32	437 ± 42	196 ± 20	33 ± 6
5	726 ± 72	722 ± 53	426 ± 31	168 ± 25	45 ± 17
10	753 ± 35	672 ± 15	444 ± 23	149 ± 17	36 ± 4
20	678 ± 18	643 ± 18	439 ± 18	217 ± 26	57 ± 7
40	498 ± 26	518 ± 12	396 ± 42	250 ± 22	104 ± 8

Appropriate application of the "t" test to the means of the combined experiments indicated that the presence of 10, 20, or 40 per cent. of polyvinylpyrrolidone in the injected material significantly decreased the blood level of *p*-aminosalicylate measured 25 minutes after injection. At 50 minutes after injection this effect was significant only in the 40 per cent. polyvinylpyrrolidone group and at 75 minutes there were no significant differences among the mean blood levels for all 5 groups. In the presence of 40 per cent. polyvinylpyrrolidone 125 minutes after injection the blood *p*-aminosalicylate concentration was significantly greater than that in the presence of 5 or 10 per cent., but not in the presence of 0 or 20 per cent. of retardant. At the end of 200 minutes the blood level of *p*-aminosalicylate was significantly elevated over the control in the presence of 20 or 40 per cent. polyvinylpyrrolidone.

Since the depressed early blood levels might have been caused by excessive amounts of polyvinylpyrrolidone interfering with the analytical method for *p*-aminosalicylic acid, this point was investigated. Amounts of polyvinylpyrrolidone corresponding to as much as 12·5 mg./ml. of blood were added to known amounts of *p*-aminosalicylic acid and the analyses performed. There was no detectable influence of polyvinylpyrrolidone on the results obtained.

It seemed possible that the more highly concentrated solutions of

† Subtosan was supplied by Poulenc, Ltd. K (Fikentscher) value (1 per cent. solution) 33.

polyvinylpyrrolidone injected intravenously might cause a temporary hæmodilution thereby leading to low blood levels of the salicylate. In an attempt to test this possibility, serial hæmoglobin determinations were run on 6 rabbits given 2·0 ml./kg. of 10 per cent. *p*-aminosalicylate in water or in 40 per cent. polyvinylpyrrolidone solutions. The hæmoglobin determinations were made on 0·05 ml. aliquots of blood by a slight modification of Rimington's pyridine hæmochromogen method.[6] The results indicated that the hæmoglobin levels increased slightly during the first 20 to 30 minutes after injection of the drug and then returned to the original level whether the carrier solution was water or an aqueous solution of polyvinylpyrrolidone. This finding is not compatible with a hæmodilution which would have to be rather considerable to explain the 40 per cent. initial depression of the blood *p*-aminosalicylate level.

With intraperitoneal injections in rats it is likely that the high viscosity of the more concentrated polyvinylpyrrolidone solutions mechanically hindered absorption of *p*-aminosalicylate into the blood stream. This could account for the initial low blood levels and more prolonged retention of *p*-aminosalicylate in the blood stream.

On the basis of these experiments in rabbits and rats it is not possible to account for the findings of Weiss *et al.*[1] that 3·5 per cent. polyvinylpyrrolidone in the *p*-aminosalicylate solutions for intravenous injection in man doubled both the maximum blood level and the duration of measurable blood levels of the antituberculosis drug. The results reported here would suggest that polyvinylpyrrolidone was without effect except at relatively high concentration and in such instances lowered the maximum blood concentration of *p*-aminosalicylate.

Summary

1. Polyvinylpyrrolidone at 40 per cent. concentration in the injected material effectively lowered the maximum blood level of *p*-aminosalicylate when the drugs were given together intravenously to rabbits or intraperitoneally to rats.

2. The duration of retention in the blood stream of significant amounts of the intravenously injected *p*-aminosalicylate was not influenced by polyvinylpyrrolidone.

3. When the combined drugs were given intraperitoneally appreciable blood levels persisted longer in the presence of 20 or 40 per cent. of polyvinylpyrrolidone.

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References

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