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THE URINARY EXCRETION OF PENICILLIN AFTER INGESTION WITH AND WITHOUT ADJUVANTS AND FOLLOWING INTRAMUSCULAR INJECTION¹

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In their classic paper Florey and co-workers (1) found that a large part of the antibacterial effectiveness of penicillin was lost when the drug was taken orally as compared with parenteral administration. This has since been confirmed by numerous investigators (2, 3, 4, 5, 6, 9, 11, 12, 13). It has generally been thought that successful administration of penicillin by mouth could be accomplished by protecting it from destruction by gastric acidity. Several studies have been reported in which attempts were made to circumvent this assumed mechanism of destruction either by neutralizing the gastric acidity by various antacid adjuvants taken with the penicillin or by encapsulating it in such a manner that it would not be released until it had reached the less acid or even slightly alkaline medium of the intestine.

Sodium bicarbonate ingested with or prior to penicillin (1, 5, 15) failed to protect most of the drug from destruction. Little and Lumb (8) reported that when penicillin mixed with raw egg white was ingested 10 minutes after taking sodium bicarbonate with milk the urinary excretion of the drug was comparable to that after a like dose injected intramuscularly; but Heatley (9) was unable to confirm their results, although he did find that the urinary excretion after taking the dose in egg white was more than double that when taken in water alone. Charney et al (5) gave penicillin orally with trisodium citrate, disodium phosphate, Amphojel (hydrrous alumina), and milk with calcium carbonate, but only the trisodium citrate and the disodium phosphate in the fasting state increased the amount recoverable in the urine to only about one-third that which would have been excreted had the same dose been given intramuscularly (2, 22). The hydrrous alumina gave an intermediate value when compared with the controls. In the absence of trisodium citrate only five per cent of the dose of penicillin taken after breakfast appeared in the urine whereas eleven per cent was excreted when the dose was taken with suitable amounts of trisodium citrate after breakfast. Even so this was only about one-fifth the amount which would have appeared in the urine had the dose been injected intramuscularly. Welch and co-workers (23) reported therapeutic blood levels of penicillin which were maintained for prolonged intervals of time after the ingestion of penicillin solutions to which suspensions of aluminum hydroxide or magnesium hydroxide were added dropwise. When 100,000 units thus treated with aluminum hydroxide were taken by eleven subjects the average recoverable in the urine during the next twenty-four hours was only

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13.6 per cent of the dose. These findings are supported by the findings of Charney et al (5) as well as by the present report. McDermott and co-workers (6) gave sufficiently large amounts of magnesium trisilicate over a period of three hours to change the pH of the stomach contents to about 8.0 before giving penicillin in water, in peanut oil, and in corn oil plus beeswax. The average urinary excretion during the next two hours was about twelve per cent in each case although there were indications that the oils delayed the excretion somewhat. György and his associates (7) reported mixed results in the clinical use of oral penicillin with trisodium citrate. With doses of penicillin comparable to those customarily given parenterally the combination was effective in the treatment of gonococic urethritis in males as well as in a few other clinical conditions, but doses as high as 300,000 units (0.18 gm.³) given orally in combination with trisodium citrate brought about only temporary clinical improvement of gonococic vaginitis in young girls even after three such courses of treatment whereas permanent cures were obtained when the same dose was administered intramuscularly. Krantz and co-workers (14) reported effective serum levels of penicillin after oral administration with basic aluminum aminoacetate. However, the effectiveness of this antacid is impossible to evaluate on the basis of their paper since the results of the control experiments where penicillin was taken by the same subjects without the antacid were not disclosed.

Florey et al (1) first suggested that the destruction of penicillin by gastric acidity might be prevented by using enteric-coated capsules but their attempts to use phenyl salicylate coated vehicles were unsuccessful. Burke (12) and his associates sealed penicillin in a capsule which was then surrounded by a second capsule that was treated by immersion in diluted formaldehyde for five seconds followed by 95 per cent alcohol for five minutes. The ingestion of 100,000 units (60 mg.³) in these capsules gave serum levels of penicillin comparable to those reported by others after the parenteral administration of 40,000 units. Libby (19) suggested the administration of penicillin by mouth suspended in digestible oils contained in enteric-coated capsules. In experiments in which the results of suitable controls were not reported, he found that therapeutic blood levels of penicillin were attainable by giving gelatin capsules containing 90,000 units of the calcium or sodium salt of the drug suspended in cottonseed oil. In view of the results reported by McDermott et al (6) it is probable that Libby would have found no therapeutic advantage to the use of the suspensions in oil had he tried either penicillin in capsules without the oil or simple aqueous solutions. (See under discussion of Table I). Recently Perlstein et al (21) gave penicillin calcium suspended in equal parts of lanolin and corn oil in gelatin capsules by mouth and found that the urinary excretion was thereby prolonged. However, the average total recovery was only about fifteen per cent or about one-fourth of what it would have been had the same amount been injected intramuscularly (2, 22).

In the following studies penicillin was taken by mouth in conjunction with various antacids and other adjuvants in attempts to decrease the destruction within the stomach and intestine so that the amounts recoverable in the urine might be made to approach or equal those recoverable after intramuscular administration. The results with a single subject are reported to avoid the confusion the data would present had the findings with several subjects been considered. It will be shown below that a remarkable constancy in the rate of excretion as well as the total amount of penicillin excreted occurred at different times in the same subject

³Since the international unit of penicillin has been defined as 0.6 microgram of the International Standard consisting of the pure crystalline sodium salt of penicillin G or II (17, 18), it has been proposed (16) that penicillin dosages might be expressed advantageously on the basis of the weight of the pure principle present in preparations now on the market rather than by the cumbersome unitage method. The situation is analogous to that proposed by Waksman (10) for expressing quantities of streptomycin used clinically.

under comparable experimental conditions. To avoid frequent, repeated venipunctures as well as the assay difficulties encountered in determining concentrations of penicillin in the blood, assays were carried out in triplicate by the Oxford Cup Method⁴ on voided urines collected at standardized intervals throughout all of the experiments. This procedure permitted screening tests to be carried out very readily. The assumption on which it is based is supported by the work of Rantz and Kirby (15) who found that several-fold differences in the rate of urinary flow in certain subjects were not associated with variations in the plasma clearance of penicillin injected intravenously and that the rate of excretion was proportional to the plasma concentration.

URINARY EXCRETION AFTER INGESTION OF 100,000 UNITS WITH AND WITHOUT ADJUVANTS COMPARED WITH INTRAMUSCULAR INJECTION

In EXPT. 1, the contents of one vial of the sodium salt of penicillin (100,000 units) was dissolved in 20 ml. of sterile normal saline for injection deep intramuscularly at two sites on the thigh. The urine was collected one, two, three, four, six and eight hours later (Table I) and immediately diluted with cold one per cent phosphate buffer, pH 6.0, for assay. The total percentage of the dose recovered, 65.1, as well as the rate of excretion was comparable to the data in the literature (2, 22) after intramuscular administration in other subjects.

EXPT. 2 was similarly carried out except that the penicillin was dissolved in only 10 ml. of sterile normal saline and the dose was injected into a single site. The very marked catharsis due to the simultaneous ingestion of five grams of U.S.P. magnesium oxide did not change the total amount excreted in the urine, 59.2 per cent, nor did it alter the rate curve significantly. This may be taken as evidence that the difference between an intramuscular dose and the amount recoverable in the urine is not due to elimination via the feces.

The average of the total amount excreted in the urine in the first two experiments is 62.1 per cent of the intramuscular dose. This value will serve through much of the discussion as a reference with which values obtained after oral administration may be compared.

EXPT. 3. When 100,000 units (60 mg.³) were taken on an overnight fasting stomach, 11.4 per cent of the dose was excreted in the urine in the first six hours. The results of this experiment wherein a very crude calcium salt of penicillin extracted from a surface culture of a strain of *Penicillium notatum*⁵ was ingested should be compared with the 11.2 per cent recovered in Expt. 6 where the same unitage as a highly refined sodium salt of penicillin extracted from a submerged culture of the mold was taken orally. Obviously there was no difference.

The average of the total amount recovered in the urine in Expts. 3 and 6 was 11.3 per cent. Comparison of this value with the average of 62.1 per cent after intramuscular injection reveals that only 18.2 per cent as much appears in the urine after ingestion on a *fasting stomach* as after intramuscular injection in the subject used. In other words five times as much would be required for oral administration *in the fasting state*.

The situation when penicillin was administered orally on a non-fasting stomach in the same subject is illustrated by the data for Expt. 12 where the total excretion was 4.7 per cent of the dose. This means that, based on these data, the subject used would have to ingest thirteen times the intramuscular dose on a *non-fasting stomach* (compare with Expt. 108).

⁴I am indebted to Marguerite M. Sullivan for these assays.

⁵Obtained from the Northern Regional Research Laboratory.

TABLE I
URINARY EXCRETION OF PENICILLIN AFTER ADMINISTRATION OF 100,000 UNITS

EXPT. No.	ROUTE	DATE	CONDITIONS UNDER WHICH PENICILLIN WAS ADMINISTERED	PER CENT OF DOSE EXCRETED IN THE INTERVALS (Hrs.)						TOTAL % OF DOSE RECOVERED
				0-1	1-2	2-3	3-4	4-6 ¹	6-8	
1	I-M ¹	3/22/45	In normal saline, non-fasting.....	39.7	15.6	5.1	2.8	1.6	0.3	65.1
2	I-M ¹	3/23/45	In normal saline, non-fasting; 5.0 gram MgO taken at same time in 200 ml. tap water. Marked catharsis when each specimen was collected.....	28.1	17.8	8.0	2.3	2.4	0.6	59.2
3	Oral ²	12/6/44	In 200 ml. water on an overnight fasting stomach.....	4.7	3.5	1.6	1.0	0.6	11.4
4	Oral ²	12/8/44	Suspended in 75 ml. U.S.P. Cottonseed oil followed by 200 ml. tap water on an overnight fasting stomach.....	4.5	4.2	1.7	0.7	0.5	11.6
5	Oral ²	12/12/44	1 mg. Atropine followed 10 minutes later by the penicillin dissolved in 200 ml. cold tap water on an overnight fasting stomach.....	3.5	3.1	2.5	2.1	1.7 ³	0.4	13.3
6	Oral ¹	3/27/45	In 350 ml. tap water on an overnight fasting stomach.....	4.7	3.2	1.3	1.1	0.9	11.2
7	Oral ¹	4/4/45	In water on an overnight fasting stomach, 10 minutes after taking 5 gms. Al(OH) ₃ suspended in water.....	5.3	4.0	1.5	0.8	0.6	12.2
8	Oral ¹	4/6/45	In water on an overnight fasting stomach followed immediately by 500 ml. tap water at 4° C. Another 200 ml. at 4° C. was drunk when first urine specimen was collected.....	3.9	3.8	1.2	1.3	1.1	0.2	11.5
9	Oral ¹	4/17/45	Dissolved in whites of 2 eggs 10 minutes following 10 gms. of NaHCO ₃ dissolved in ½ pint of homogenized milk on an overnight fasting stomach.....	1.0	5.2	2.3	1.0	2.1	11.6
10	Oral ¹	5/14/45	In water on an overnight fasting stomach 10 minutes after taking 20 gms. of powdered egg white dissolved and suspended in about 400 ml. of tap water.....	3.5	2.6	2.1	1.5	1.1	0.1	10.9
11	Oral ¹	5/9/45	Dissolved in egg-nog containing 3 egg whites, milk, sugar and vanilla immediately after a breakfast consisting of bacon, egg, toast with butter and hot chocolate made with milk.....	1.5	3.1	1.3	0.9	1.2	0.2	8.2
12	Oral ¹	5/7/45	In water 40 minutes after breakfast consisting of orange juice, toast and butter, poached egg, and dry cereal with milk and sugar.....	1.2	1.6	0.9	0.6	0.4	4.7
13	Oral ¹	4/10/45	In water on an overnight fasting stomach 10 minutes following 25 gms. of Wilson Granular Concentrated Mucin washed down with 250 ml. of tap water.....	2.4	1.7	0.7	0.3	0.2	5.3
14	Oral ¹	5/11/45	Dissolved in 400 ml. water containing 20 gms. glycine on an overnight fasting stomach.....	2.4	3.7	2.0	0.9	0.6	0.1	9.7
15	Oral ¹	5/16/45	In water immediately following 5 gms. activated charcoal, Merck, suspended in 300 ml. water on an overnight fasting stomach.....	0.2	0.2	0.2	0.1	0.1	0.8
16	Oral ¹	3/28/45	In 400 ml. water containing 3 gms. trisodium citrate on an overnight fasting stomach.....	7.0	7.2	2.4	1.4	0.8	18.8
17	Oral ¹	6/13/45	In water on overnight fasting stomach followed immediately by a solution of 5.0 gms. ferric ammonium citrate in about 300 ml. tap water.....	0.7	3.0	2.4	1.1	0.8	0.2	8.2

TABLE I—(Continued)

EXPT. No.	ROUTE	DATE	CONDITIONS UNDER WHICH PENICILLIN WAS ADMINISTERED	PER CENT OF DOSE EXCRETED IN THE INTERVALS (Hrs.)						TOTAL % OF DOSE RECOVERED
				0-1	1-2	2-3	4-5	4-6 ^a	6-8	
18	Oral ¹	6/14/45	In water on overnight fasting stomach followed immediately by a solution of 5.0 gms. d-glutamic acid adjusted to pH=8.0 with NH ₄ OH in about 300 ml. tap water.....	2.6	5.2	2.1	0.9	1.0	0.1	11.9
19	Oral ¹	6/15/45	In water on overnight fasting stomach together with an elixir of ammonium citrate ⁶ calculated to contain 5.0 gms. of the salt in about 250 ml. tap water.....	3.4	3.1	2.1	1.2	1.0	0.1	10.9
20	Oral ¹	6/19/45	In water on overnight fasting stomach followed immediately by 5.0 gms. of diammonium acid citrate in solution in about 300 ml. tap water.....	2.0	2.3	0.8	0.4	0.5	0.6	6.6
21	Oral ¹	6/21/45	In water on overnight fasting stomach followed immediately by a suspension of 20.0 gms. of casein in about 500 ml. of tap water.....	3.6	1.5	1.4	0.8	0.8	0.2	8.3
22	Oral ¹	6/23/45	In water on overnight fasting stomach followed immediately by a suspension of 20.0 gms. of zein in about 400 ml. tap water.....	2.9	3.3	1.7	1.0	0.9	0.2	10.2
23	Oral ¹	6/27/45	One vial dissolved in 20 ml. water, 30 ml. hydrous alumina (Creamalin, Liquid) slowly stirred in. Immediately drunk on overnight fasting stomach.....	5.3	5.4	2.3	0.9	0.6	14.5

¹Penicillin Sodium, deep culture.²Calcium salt containing 54.5 units/mg. surface culture.³Estimated value.⁴Regular lunch eaten in this interval.⁵Kindly supplied by the Wm. S. Merrell Co.⁶13.3 ml. of a solution of 12 gm./fl. oz. of ammonium citrate kindly supplied by the Wm. S. Merrell Co.

EXPT. 4. It was felt that suspending the penicillin in a digestible oil might protect it from the destruction by the gastric juice while permitting its release for absorption in the duodenum and jejunum. The results of this experiment, 11.6 per cent, indicate that there was no therapeutic advantage to be gained from suspending penicillin in cottonseed oil for ingestion over giving it in simple aqueous solution. This is in contrast to the inferences of Libby's paper (19) in which the results of suitable control experiments were not reported.

EXPT. 5. These data reveal only a slight advantage to the use of atropine with a view to decreasing the destruction by gastric juice of penicillin taken orally.

EXPT. 6. Discussed under Expt. 3 above.

EXPT. 7. U. S. P. aluminum hydroxide, dried, ingested with the penicillin increased the amount recoverable in the urine slightly to 12.2 per cent. In Expt. 23, 30 ml. of a suspension of approximately 5.5 per cent aluminum hydroxide (Creamalin, Liquid) was slowly stirred dropwise into 100,000 units of penicillin dissolved in 20 ml. of distilled water and immediately drunk on an overnight fasting stomach. This method of mixing increased the amount recoverable in the urine during the first six hours to 14.5 per cent. According to these data four times as much of the penicillin treated in this way would have to be ingested on a fasting stomach instead of five times the dose in simple aqueous solution required over the intramuscular route. The method of preparation of the "modified" penicillin was essentially that of Welch *et al* (23) and the amount of the ingested dose recoverable in the urine was the same as they reported.

EXPT. 8. Chilling the stomach with large volumes of cold water simultaneously with the ingestion of the penicillin did not change the amount excreted in the urine.

EXPT. 9. This work was done to try to duplicate the findings of Little and Lumb (8) who reported that sodium bicarbonate in milk followed by penicillin dissolved in raw egg white resulted in urine levels of penicillin comparable to those following similar doses administered intramuscularly. Heatley (9) was unable to confirm this and obviously the data in Table I do not support it in the case of the subject used but show instead that the excretion was the same as would have occurred had the same dose been taken on a fasting stomach without the alkali and egg white.

EXPT. 10. This work was designed to see whether egg white alone would be as effective as with sodium bicarbonate and the data showed that this was the case. This suggests then that by giving penicillin with egg white it is unnecessary to keep the individual in the fasting state if five times as much penicillin is given as would be required by intramuscular injection instead of the use of thirteen times as much in the non-fasting state.

EXPT. 11. In order to test the protective action of egg white further, penicillin was given in an egg-nog containing the whites of three eggs on a non-fasting stomach. The total recovered in the urine was 8.2 per cent compared with 4.7 per cent in Expt. 12 where the same dose of penicillin was taken without egg white on a non-fasting stomach. This means that eight instead of thirteen times as much would be required to bring the urinary excretion levels up to the values attained by intramuscular injection.

Little and Lumb (8) attribute the protective action of egg white to its sulfur content. The acid buffering capacity of the egg white protein itself as well as its digestion products as they are gradually and continuously split off by the action of acid-pepsin might be important factors in the protective action.

EXPT. 12. Discussed under Expts. 3 and 11.

EXPT. 13. It is generally thought that gastric mucin elaborated by cells lining the stomach protect the mucosa from the action of the acid-pepsin. However, 25 grams of a concentrated mucin preparation swallowed and washed into the stomach with tap water followed ten minutes later by 100,000 units of penicillin in water was ineffective.

EXPT. 14. The buffering capacity of various substances with respect to 0.100 N HCl was determined as shown in Table II. It will be noted that only five grams of glycine prevented the pH from dropping below 3.1 when 150 ml. of the acid was added. Since previous experiments⁶ had shown that penicillin was much more stable at pH 3.1 than would have been the case at the usual pH of gastric juice (about 1.8 or less), 20 grams of glycine dissolved in water was drunk on an overnight fasting stomach followed immediately by 100,000 units of penicillin in water. The resulting total urinary excretion was only 9.7 per cent.

EXPT. 15. Data in Table II show that acid is taken up by an activated carbon to a considerable degree. Since penicillin is readily absorbed by activated carbon it was decided to take penicillin orally with it to see whether the penicillin so absorbed would be protected from the action of the gastric juice with the thought that it might be eluted from the carbon for absorption in the duodenum and jejunum. Less than one per cent of the dose was recovered in the urine when five grams of Merck's activated carbon was used.

EXPT. 16. This experiment was undertaken to determine whether similar results could be obtained with the subject under study as those reported by Charney, *et al* (5). The data confirm their findings. When five grams of trisodium

⁶Unpublished data.

citrate were ingested on an overnight fasting stomach with 100,000 units of penicillin, 18.8 per cent of the dose was recovered in the urine. This is 30 per cent of the recovery after intramuscular administration and 166 per cent of the amount recoverable in the urine after ingestion under similar conditions except that no sodium citrate was taken simultaneously.

EXPT. 17. A heavy dose of ferric ammonium citrate proved to be of no advantage. It was taken with the thought of combining the oxidizing properties of the ferric iron with the acid buffering capacity of ammonium citrate. An oxidizing agent was used in an attempt to prevent the inactivation of penicillin in the strongly reducing medium of the upper intestine since Abraham and Chain (24) stated that penicillin was inactivated by such reducing agents as NaHSO_3 and $\text{Na}_2\text{S}_2\text{O}_4$ among others as well as by the primary alcohols and Cavillito and Bailey (25) and others (26, 27) reported that cysteine readily inactivated penicillin.

TABLE II

IN THE FOLLOWING EXPERIMENTS 5.0 GMS. OF EACH REAGENT WERE DISSOLVED OR SUSPENDED IN ABOUT 75 ML. OF DISTILLED WATER AND STIRRED VIGOROUSLY DURING THE ADDITION OF 0.100 N HCl WHILE NOTING CHANGES IN pH WITH A GLASS ELECTRODE

ml. 0.100 N HCl	Trisodium Citrate	Glycerine	Egg Albumin Powder	Merck Activated Charcoal	Ammonium ¹ Glutamate Solution	Ferric Ammonium Citrate	Di-am- monium Acid Citrate	Ammonium ² Citrate Solution
0	pH 8.6	pH 5.8	pH 7.1	pH 6.4	pH 7.8	pH 6.4	pH 5.6	pH 5.0
1	7.7	5.1	6.8	5.9	7.7	6.3	5.6	5.0
2	7.3	4.8	6.5	5.6	7.6	6.2	5.6	5.0
3	7.2	4.7	6.3	5.3	7.5	6.1	5.6	5.0
4	7.1	4.5	6.1	5.1	7.3	6.0	5.6	5.0
5	7.0	4.4	6.0	4.8	7.1	5.9	5.6	5.0
10	6.7	4.2	5.3	3.9	6.4	5.3	5.6	5.0
15	6.5	4.2	4.8	3.4	6.0	4.8	5.6	5.0
20	6.4	4.0	4.3	3.0	5.7	4.5	5.6	4.9
25	6.3	4.0	3.9	2.7	5.6	4.2	5.6	4.8
30	6.2	3.9	3.6	2.5	3.8	5.6
40	6.1	3.7	3.1	2.3	3.2	5.5
50	6.0	3.6	2.8	2.2	5.1	2.8	5.4	4.7
100	5.5	3.3	2.0	2.2	4.6	2.2	5.0	4.4
150	5.0	3.1	1.8	1.7	4.4	4.7	4.1
200	4.3	4.1	4.4	3.9
250	3.5	4.1	3.6
300	3.8	3.3

¹5.0 gms. d-glutamic acid suspended in water, pH=3.3. Solution affected by addition of 5.5 ml. of concentrated NH_4OH . Final pH=7.8.

²13.3 ml. of a solution of 12 gm./fl. oz. of ammonium citrate kindly supplied by the Wm. S. Merrell Co.

EXPT. 18. Only an insignificant increase to 11.9 per cent excreted in the urine resulted from the simultaneous ingestion of ammonium glutamate over the 11.3 per cent recovery when the simple aqueous solution was drunk.

EXPT. 19. An elixir of ammonium citrate proved to be of no particular advantage in spite of its great *in vitro* buffering capacity as shown in Table II.

EXPT. 20. When five grams of diammonium acid citrate dissolved in water were drunk immediately following an aqueous solution of 100,000 units of penicillin on an overnight fasting stomach, only 6.6 per cent of the dose was excreted in the urine. This was only about one-third of the recovery resulting when the same amount of trisodium citrate was simultaneously ingested (compare with Expt. 16).

EXPT. 21. A heavy suspension of casein was found to have a protective capacity mid-way between that of powdered egg white and the non-fasting state (compare Expts. 10 and 12).

EXPT. 22. A heavy suspension of zein was tried because this protein is quite insoluble in water and it was felt that it might resist digestion in the stomach for some time and thus exert a protein acid-buffering protective capacity longer than in the case of soluble proteins. Compared with Expt. 10 it was found to be almost as efficacious as powered egg white.

**URINARY EXCRETION AFTER INGESTION OF 43,250 UNITS
WITH MAGNESIUM TRISILICATE AND OTHER ADJUVANTS**

The penicillin used in compiling the data in Table III was in the form of tablets made up with magnesium trisilicate⁷ to contain 4,325 units each. Ten of the tablets were taken by mouth for each of the experiments under the conditions described in the table.

TABLE III
**URINARY EXCRETION OF PENICILLIN AFTER ORAL ADMINISTRATION OF 43,250 UNITS OF
TABLETS MADE UP WITH MAGNESIUM TRISILICATE¹**

EXPT. No.	DATE	CONDITIONS UNDER WHICH PENICILLIN WAS ADMINISTERED	PER CENT OF DOSE EXCRETED IN THE INTERVALS (Hrs.)					TOTAL % OF DOSE RECOVERED
			0-1	1-2	2-3	3-4	4-6 ²	
101	1/31/45	With 300 ml. tap water on an overnight fasting stomach....	3.7	3.3	1.8	1.0	1.0	10.8
102	2/2/45	In tap water on an overnight fasting stomach followed immediately by 3 gms. U.S.P. sodium benzoate dissolved in tap water.....	1.1	2.3	1.1	0.6	0.4	.5
103	2/7/45	In tap water on an overnight fasting stomach followed immediately by 0.7 mg. atropine and 75 mg. phenobarbital.....	2.5	2.6	1.9	1.5	0.9	9.4
104	2/9/45	Same as No. 103 except the penicillin was taken 45 minutes after the atropine and phenobarbital were ingested.....	1.9	3.2	.8	1.2	0.9	9.0
105	2/12/45	In tap water on an overnight fasting stomach followed by a suspension of 5 gms. of magnesium trisilicate in 350 ml. of tap water.....	1.2	2.5	0.9	0.3	0.2	5.1
106	2/14/45	In tap water on an overnight fasting stomach followed immediately by a suspension of 5 gms. of magnesium oxide in 400 ml. of tap water.....	0.9	0.6	0.1	0	0	1.6
107	2/16/45	In tap water on an overnight fasting stomach followed immediately by 5 Creamalin tablets (hydrous alumina) which had soaked overnight in water.....	3.6	2.9	1.3	0.6	0.4	8.8
108	2/24/45	With 1 pint of homogenized milk 1 hour after breakfast consisting of bacon, 1 egg, toast and butter, hot chocolate made with milk, and half grapefruit.....	0.2	1.0	1.0	0.4	0.5	3.1

¹Kindly supplied by the Wm. S. Merrell Co.

²Regular lunch eaten in this interval.

EXPT. 101. 43,250 units were taken on an overnight fasting stomach. The 10.8 per cent total of the dose excreted in the urine compared closely with the amounts recoverable when 100,000 units were ingested (see discussion of Expts. 3 and 6). Obviously the incorporation of magnesium trisilicate with the penicillin in the tablet was not advantageous.

EXPT. 102. It is well known that the ingestion of sodium benzoate results in conjugation with glycine in the liver to form hippuric acid which is then excreted in the urine. Beyer et al (28) showed that the simultaneous intravenous administration of p-amino-hippuric acid with penicillin resulted in greatly prolonging the urinary excretion of the drug with attendant increased blood levels for long

⁷Obtained from the Wm. S. Merrell Co.

intervals of time. It was thought that the ingestion of sodium benzoate with penicillin might result in the delayed excretion of the latter because of preferential excretion by the kidneys of the hippuric acid formed in the liver from the sodium benzoate. Neither the total amount recoverable nor the rate of excretion data support the hypothesis.

EXPTS. 103 and 104. These data demonstrate that destruction by the gastric juice of penicillin taken orally was not significantly altered either by taking the drug simultaneously with atropine or by taking it after atropine at a sufficient interval for any maximal effect of the atropine on the secretion of gastric juice to have occurred.

EXPT. 105. These data show that the simultaneous ingestion of a large dose of magnesium trisilicate with penicillin had a deleterious effect on the amount of antibiotic activity recoverable in the urine. Possibly the magnesium trisilicate absorbed the penicillin and did not release it, or it may greatly prolong the emptying-time of the stomach similar to the view held by some concerning the action of sodium bicarbonate (13).

EXPT. 106. When five grams of magnesium oxide were taken as an antacid simultaneously with the tablets containing the penicillin, marked catharsis resulted. The data show that the penicillin may have been washed out of the stomach and intestine in the voluminous, watery stools. This was true both with respect to the total activity recoverable as well as the rate at which penicillin appeared in the urine. If the interpretation that the penicillin was washed out in the feces be valid, then the question arises as to whether the absorption from the gut into the blood stream normally may be only a slow process.

EXPT. 107. The results of this experiment are at variance with those of Expts. 7 and 23 in that hydrated alumina (Creamalin, Tablets) were not effective in preventing the destruction of penicillin taken orally under the conditions of the experiment.

EXPT. 108. Here the penicillin was taken about an hour after a meal and the data confirm the results of Expt. 12 (Table I). The recovery was only 3.1 per cent. When this is compared with the results of Expt. 101 it is seen that less than thirty per cent of the recovery when the drug was taken on a fasting stomach was obtained. These data mean that about twenty times the intramuscular dose would be required on a non-fasting stomach to bring the urinary excretion to the same level (compare with Expts. 3 and 11).

DISCUSSION

The data in these studies show that the maximal amount of oral penicillin recoverable in the urine was less than one-third of the antibiotic activity excreted in the urine after the same dose was injected intramuscularly. That this was true in spite of the simultaneous ingestion of large amounts of various antacids leads to the thought that gastric acidity is not solely responsible for the destruction of ingested penicillin. Rammelkamp and Helm (3) found that only 22.7 and 39.6 per cent of the ingested dose appeared in the urine of two achlorhydric pernicious anemia patients. Previous experiments⁸ have shown that sixty-six per cent of the original activity was retained for twenty-four hours at 37° C. at pH 5 *in vitro*. In Table II it is seen that sufficient amounts of trisodium citrate and diammonium acid citrate were ingested with penicillin to have maintained this pH in the presence of 150 ml. and 100 ml. of one-tenth normal hydrochloric acid respectively. In spite of this similar *in vitro* buffering capacity, the data in Table I (Expts. 16 and 20) demonstrate that there was a three-fold difference in

⁸Unpublished data.

antibiotic activity in the urine. Since penicillin was taken with these buffers on a fasting stomach and was not followed by food, it is probable that little if any free hydrochloric acid was present or appeared soon after ingestion took place. Further, the bitter and nauseating concoctions probably would have inhibited gastric secretion for some time. Moreover, the data show that more than one-third of the total amount excreted appeared in the urine during the first hour when, according to the data of Bergeim (30), an average of only 0.07 per cent of free hydrochloric acid appears in the gastric contents during the first forty-five minutes after a meal.

It is thought that the decreased efficacy of oral penicillin, aside from that which does occur due to destruction previously by gastric acidity, may be due in large part to inactivation in the liver when the penicillin is absorbed into and carried by the portal system directly to that organ from the gastro-intestinal tract. The critical experiments of Florey et al (1) on this point merit repetition since their results were not conclusive in view of the small amounts of the drug which they had available for use in such experiments at that time. Recently Perlstein et al (20) presented evidence that the hypothesis could be correct and that the penicillin may be inactivated in the liver by coupling with glucuronic acid similar to the inactivation of aromatic acids, phenols and alcohols by conjugation with this acid (29). No suitable adjuvant which might prevent such conjugation is foreseen.

CONCLUSIONS AND SUMMARY

Bearing in mind all of the advantages and limitations imposed by the use of a single subject, the data presented in this report seem to justify the following conclusions with respect to that subject.

1. Five times as much penicillin is required to be ingested *on a fasting stomach* to give the same excretion of penicillin via the kidneys as by intramuscular injection in the subject used.
2. Thirteen to twenty times as much is required orally *in the non-fasting state*.
3. There is no difference between the penicillin extracted from surface cultures of the mold compared with submerged cultures with respect to destruction when taken orally as judged by recovery of antibiotic activity in the urine.
4. The same proportion of the antibiotic activity is recoverable in the urine when the same dose (based on antibiotic activity) of a very crude calcium salt of penicillin is ingested compared with a highly refined sodium salt of the drug.
5. There is no therapeutic advantage to the oral administration of penicillin suspended in cotton-seed oil over a simple aqueous solution of the drug.
6. Atropine taken simultaneously with oral penicillin or forty-five minutes before the antibiotic was ingested did not increase the proportion excreted in the urine over that recoverable when atropine was not taken.
7. Aluminum hydroxide taken with penicillin either did not change or increased only slightly (depending on the method of mixing) the amount of antibiotic activity recoverable in the urine. At best four times as much penicillin would have had to have been taken with the aluminum hydroxide on a fasting stomach to have given the same amount of excretion via the kidneys as by the intramuscular route of administration of penicillin.
8. Ferric ammonium citrate, ammonium glutamate, elixir of ammonium citrate, diammonium acid citrate, magnesium trisilicate, magnesium oxide, glycine, activated carbon, sodium benzoate, concentrated mucin, casein, and zein when ingested simultaneously with penicillin on a fasting stomach either decreased or did not alter the proportion of the antibiotic activity recoverable in the urine compared with oral administration in tap water.

9. The ingestion of five grams of trisodium citrate with penicillin increased the amount of the dose recoverable in the urine by sixty-six per cent over that recoverable without the salt but the amount in the urine was still only thirty per cent of that recoverable had the same dose been given intramuscularly.

10. Ingestion of penicillin with raw egg-white with or without sodium bicarbonate resulted in the same proportion of the penicillin being excreted in the urine as after taking the same dose with tap water on a fasting stomach.

11. Chilling the stomach with large volumes of cold water did not increase the amount of orally administered penicillin which appeared in the urine.

12. The capacity for the increased destruction of penicillin taken with food compared with oral administration in the fasting state persisted for several hours after the food was eaten.

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