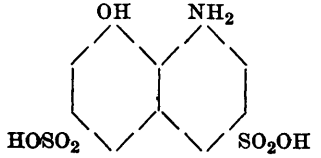
more sensitive to the basic dyes than to the acid dyes, but the Gram negative bacili are more sensitive to the acid dyes.

In order to study this questions further we have tested the antiseptic action of H acid (aminonaphthol disulphonic acid),



an acid which is present in both trypan blue and the new trypanosome remedy (Baeyer 205[5])* The results are shown in the table above.

It is evident that the Gram positive cocci tested here are more sensitive to H acid than are the Gram negative bacilli, and therefore that Churchman's findings cannot be regarded as evidence of an entirely general rule.

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The synthesis and excretion of hippuric acid: The glycine factor.

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The rate of synthesis and excretion of hippuric acid in normal individuals and those with various renal disorder has already been studied by Kingsbury and Swanson[1] and this study made the basis of renal function test. In this work 2.4 grams of sodium benzoate was the dosage regardless of the size or weight of the individual. In the present paper the original work has

[4] Churchman, J., *Jour. Exper. Med.*, 1913, xvii, 373; PROC. SOC. EXPER. BIOL. AND MED., 1922, xix, 288-31.

* A sample of pure H acid was kindly prepared for us by Drs. Derick and Strachan, of the laboratories of the National Aniline Company, and the bacterial cultures were furnished by Dr. J. F. Noble, to whom we extend our sincere thanks.

[5] Bayer 205, *Science*, 1922, lvi, 514.

[1] Kingsbury, F. B., and Swanson, W. W., *J. Biol. Chem.*, 1921, xlv, 4; *Arch. Int. Med.*, 1921, xxviii, 220.

been extended using dosages of sodium benzoate based on body weight in one series of 21 (50 milligrams per kilo) and on body surface in another series of 66 (1.8 grams per square meter of body surface). Body surfaces were obtained from the height-weight formula and chart of Dubois and Dubois.[2] The dosage based on surface was obtained by calculation after determining the body surface of one individual of average size and weight who had had 50 milligrams of benzoate per kilo of body weight. All individuals of both series were medical students who made the tests on themselves as part of their work in Physiolical Chemistry and whose urine specimens I made check analyses on by the method of Kingsbury and Swanson.[3] The benzoate was ingested quantitatively the first thing in the morning, having voided the night urine immediately before. Breakfast was omitted. The total water intake was 500 c.c. in every case. The urine was collected for the next three hours and immediately analyzed. The small and variable 3 hour normal output of hippuric acid independent of the ingested benzoate is included in these figures. Experience has shown that it is unnecessary to determine this and correct for it in the practical application of the test. In the first series of 21 it was noted that the groups of heavier individuals generally put out a smaller amount of hippuric acid per kilo of body weight than did the groups of lighter individuals, although all received the same dosage of sodium benzoate per kilo of body weight. This is shown in Table 1 in the form of percentages of excretion for the different weight groups.

This indicates that the actual amount of functioning tissue in the kidneys is less in its relation to body weight in the heavy person than in the light. This was also shown by a fairly high, but negative correlation coefficient· 0.50, between the per cent. excretion and the body weight with a probably error of ± 22 per cent.

In a second group of 66 normal individuals given benzoate, 1.8 grams per square meter of body surface, it was noted that the average percentage of excretion for groups of different body weights was nearly constant, indicating that the actual amount of functioning kidney substance is more nearly in proportion to the body surface than to the body weight. Calculations have shown that by using the square root of the body weight as a

2 Dubois and Dubois, *Arch. Int. Med.*, 1917, xvii.

3 Kingsbury, F. B., and Swanson, W. W., *J. Biol. Chem.*, 1921, xlviii, 13.

basis for the benzoate dosage that nearly the same amount is obtained for any one individual as by the body surface plan. This agrees with the results obtained by Austin, Stillman and Van Slyke,[4] who found that the square root of the body weight was more adequate than the body weight itself in their constant for urea excretion. The correlation coefficient between the milligrams of hippuric acid excreted per square meter of body surface and the volume of urine excreted on the same basis was small, 0.13, negative and with a very high probable error, ±64 per cent., indicating that in general there it little relationship between the excretion of hippuric acid and of water and that there is a slight tendency for the kidney that shows the best output of hippuric acid also to show the best concentrating ability. In the first group of 21 quite the reverse of this was true· namely that the greater volume of water that was excreted in a test was accompanied by a greater relative amount of hippuric acid. This was well shown by correlating the milligrams of hippuric acid excreted per kilo of body weight with the volume of urine excreted on the same basis. The coefficient obtained was 0.42, positive, and with a probable error of ±28 per cent. A comparison of some of the findings with the two groups are shown in Table 1.

It will be noted from Table 1 that 91 per cent. of the individuals of the group of 66 were able to excrete more than 85 percent. of hippuric acid theoretically obtainable from the ingested benzoate. Values lower than 80 per cent. in this test for renal function should be regarded as indicating probable renal insufficiency.

Two normal individuals in a total of 89, given benzoate according to the body surface plan gave evidence that glycine was not furnished with sufficient rapidity for the synthesis and excretion of hippuric acid at the normal rate. The first, a healthy medical student, gave on two occasions tests of 53 and 67 per cent. He was thoroughly examined by Dr. George E. Fahr of the Department of Medicine, who also determined his Van Slyke[4] urea excretion constant. This was within the normal limits. The second student showed benzoate test figures of 69 on two occasions and while not examined physically was apparently in perfect health. Glycine in amount equivalent to the benzoate was

[4] Austin, J. H., Stillman, E., and Van Slyke, D. D., *J. Biol. Chem.*, 1921, xlvii, 91.

given with the benzoate to each of these students in subsequent tests. The first showed 90 per cent. and the second 84 per cent. It should be noted that there was an interval of at least a week between any two of these tests on the same individual. The benzoate dosage in each case was approximately 50 milligrams per kilo of body. Lewis and Griffith[5] gave rabbits the enormous dosage of 1 gram of sodium benzoate per kilo of body weight and found that the simultaneous administration of glycine markedly increased the rate of synthesis and elimination of hippuric acid. This was to have been expected since the dosage which they used was approaching the lethal dose of 1.7 grams per kilo. In previous tests using 2.4 grams of benzoate I had tried the effect of the simultaneous administration of glycine without result in the case of normal human beings, and I was therefore surprised that with the somewhat larger dose of 50 milligrams per kilo, but still a relative small dose, to find two persons out of a series of 89 who showed this peculiar condition. This apparent difficulty in certain persons of readily supplying glycine must be taken into account in making benzoate tests: Experiment with normal given benzoate per body surface basis shows administration of glycine increases rate of hippuric acid output in two hours' interval markedly but slightly in three hours interval. It will be necessary to supply sufficient glycine in all tests to rule out the possibility of an occasionally low benzoate test being due to this glycine factor. Gelatine can probably be adequately substituted for glycine for this purpose if given in proper amount and a short time before the administration of the benzoate to allow time for digestion.

TABLE I.

	Group 1, 21 cases		Group 2, 266 cases		Group 2		
Body wt. kilos.	No. of cases	Aver. H. A. % excreted	No. of cases	Aver. H. A. % excreted	% excretion	No. of cases	% of total
41- 50	1	100	1	96	80- 85	6	9
51- 60	5	91	16	91	86- 90	20	30
61- 70	11	90	26	96	91- 95	22	33
71- 80	3	83	21	91	96-100	18	28
81- 90	1	85	1	92			
91-100	0		1	100			

[5] Lewis, H. B., and Griffith, W. H., *J. Biol. Chem.*, 1923, lv, 22.

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