XIII. *On the Conversion of Benzoic Acid into Hippuric Acid in the Animal Economy. By* Mr. Alfred Baring Garrod, *of University College.*

Read January 18, 1842.

A PAPER has appeared in the Medico-Chirurgical Transactions for last year, and also in the first Number of the Pharmaceutical Transactions, by Dr. Alexander Ure, in which it is stated, that by the internal administration of benzoic acid, or any of its salts, hippuric acid is formed in the system, and is eliminated from the kidneys in the form of a soluble hippurate, and that this hippurate is formed by the benzoic acid uniting with uric acid. It is also stated, that no trace of uric acid, or any of its salts, could be found in the urine after the administration of the benzoic acid.

I have repeatedly performed Dr. Alex. Ure's experiment, swallowing from a scruple to half a drachm of benzoic acid at a time, and have always obtained a copious crop of crystals of hippuric acid, amounting to from fifteen to twenty-nine grains, by the addition of hydrochloric acid to the urine passed about three or four hours afterwards (evaporated or not, according to its state of dilution). These crystals possessed all the characters of hippuric acid, with the crystalline form, the small solubility in cold water and æther, the ready solubility in alcohol, the evolution of nitrogen, and also the odour of the tonquin bean when heated to destruction; and my experiments therefore so far confirm Dr. A. Ure's fundamental ob-

servation. He also mentions another test of hippuric acid, viz. that when evaporated to dryness with dilute nitric acid, and ammonia added, a beautiful purple colour is produced. This is certainly true of the crystals obtained from the urine, but it is not a character of pure hippuric acid. The cause of this colour will be shown presently.

Dr. A. Ure states that no trace of uric acid could be found in the urine; but on examination I have always been able to obtain a distinct trace of uric acid from a drop or two of the urine, by adding a little nitric acid, carefully evaporating, and holding the capsule containing it over ammonia, when a distinct trace of murexide was formed; also, when the dish containing the crystals of hippuric acid is carefully examined, minute grains are found at the bottom, which are uric acid crystals; and on examining the crystals of hippuric acid with the microscope, uric acid crystals are found adhering to them in immense numbers, and this is the cause of the production of the purple colour spoken of, and which has been given as a test of hippuric acid. When the crystals are dissolved in alcohol the uric acid is precipitated, and the hippuric acid crystallized from the alcoholic solution no longer gives the purple colour. On collecting the uric acid from the same quantity of urine, formed on successive days, the same food being taken, one containing about twenty-seven grains of hippuric acid, and the other none, the following results were obtained:—

From 4½ oz. of urine, when no benzoic acid had been taken, uric acid 1·07 grain.

From 4½ oz. of urine, after taking 30 grains of benzoic acid, uric acid 0·96 grain.

Difference in favour of first, 0·11 grain.

In the second experiment also, a small loss might have occurred from the greater washing of the crystals necessary in that experiment. Now if we suppose that uric acid is decomposed to afford the elements necessary to be added to benzoic acid to form hippuric acid, we find that each equivalent of benzoic acid requires the addition of $C_4 H_6 O_4 N$. To obtain the nitrogen, four atoms of benzoic acid would require one atom of uric acid, or half a drachm of benzoic acid would require rather more than ten grains of uric acid. Now the quantity of urine, in the experiment without the benzoic acid, only contained 1·07 grain of uric acid, and yet that quantity was not materially diminished when twenty-eight grains of hippuric acid were found in the urine. It cannot therefore be from the uric acid that the hippuric acid is formed.

If we examine the subject theoretically, it does not seem

probable that such a body as benzoic acid, possessing such feeble affinities, and producing no sensible action on the body when taken, should be able to break up such a stable compound as uric acid; to abstract from the latter the requisite elements for its conversion into hippuric acid. But as hippuric acid is really formed in the urine, from whence does it obtain the necessary addition? The quantity of urea was noticed in several experiments to be deficient; could this be the source? We can find no rational formula for the explanation of the conversion if we suppose it to be from urea alone. We can, it is true, select the elements required; but, as in the last case, we should leave some compound in the system, which cannot be resolved into any known compounds, as ammonia, water, carbonic acid, &c., while from the ready conversion of the benzoic acid into hippuric acid we should expect that the change was one which could easily take place, without the action of any unusual affinities being brought into play. It occurred to me that it might be the lactate of urea, instead of pure urea, which is taken up; and upon comparing the formulæ for hippuric acid, benzoic acid, and the lactate of urea, it appeared that one equivalent of lactate of urea minus three eqs. of water, gave exactly the requisite elements for the conversion of 2 eqs. of benzoic acid into 2 eqs. of hippuric acid. 2 eqs. of benzoic acid + 1 eq. of lactate of urea = 2 eqs. of hippuric acid + 3 eqs. of water.

Hippuric acid (anhydrous)	$C_{18}\ H_8\ O_5\ N_1$
Benzoic acid (Do.)	$C_{14}\ H_5\ O_3$
Difference	$C_4\ H_3\ O_2\ N_1$
Twice the difference . .	$C_8\ H_6\ O_4\ N_2$
Lactic acid	$C_6\ H_5\ O_5$
Urea	$C_2\ H_4\ O_2\ N_2$
Lactate of urea	$C_8\ H_9\ O_7\ N_2$

Lactate of urea $- 3\ H\ O = C_8\ H_6\ O_4\ N_2$.

Now the urea has by MM. Cap and Henry been found to exist in human urine as lactate, and the separation of the elements of water is a change which might be expected to take place in the system under such circumstances. The benzoic acid merely taking up the lactate of urea, and throwing off water, is certainly a more probable occurrence than the destruction of such a stable compound as uric acid.

In analyses for the quantity of lactate of urea, according to the method of Cap and Henry, I found that although I could not obtain it in crystals, yet the quantity in a syrupy state was much reduced after taking the benzoic acid, and the same appeared on forming nitrate of urea from it. I obtained 14 grs.

less of urea in 4½ ounces of urine when the benzoic acid had been taken. In another experiment I obtained 17 grs. less of urea when 30 grs. of benzoic acid had been taken; this is a greater loss than can be accounted for by the formation of the hippuric acid; but this can be referred to the urine, from some accidental circumstance, being of nearly as high specific gravity in this case as when the benzoic acid had been taken. 30 grs. of benzoic acid, swallowed, usually increased the specific gravity of the urine from four to six-thousandths.

From these results two inquiries suggest themselves:—1st, May not hippuric acid be formed artificially out of the body? 2nd, If sufficient benzoic acid were swallowed at such a time when least urea was contained in the urine, would the benzoic acid not cease to be all converted into hippuric acid, part of it then appearing in the urine unchanged?