The excretion of trigonelline by normal individuals on a coffee-free diet ranged from 10 to 16 mgm. per day. After dosing with nicotinic acid the increased output of trigonelline varied from 10 to 28 per cent of the ingested dose, depending upon the size of the dose and the body weights of the subjects.

The metabolic connexion between trigonelline and nicotinic acid is of interest in relation to the test proposed for assessing the nutritional status of human subjects in the anti-pellagra vitamin (Harris and Raymond, 1939[7]). This depends on the measurement under controlled conditions of the urinary excretion of nicotinic acid or related substances. We have confirmed the conclusion of Harris and Raymond[7] that their method does measure essentially the nicotinic acid (which is biologically active), and that trigonelline (which is inactive), does not interfere. With the low concentration of alkali and the short period of hydrolysis used, no detectable amount of trigonelline is converted to nicotinic acid. The disadvantage of a prolonged digestion with stronger alkali, as used by Swaminathan[10], is that a considerable conversion of trigonelline into nicotinic acid occurs. The method of alkaline hydrolysis used by Harris and Raymond seems preferable to that of acid hydrolysis used by Melnick and Field[8], since the latter may involve an incomplete conversion of nicotinuric acid, and a darker digestion mixture. Also Melnick and Field[9], in criticizing the blank correction, seem to have overlooked the fact that the Cambridge workers employed an acid reaction medium, in which the urine reacts less with interfering substances than in the neutral medium used by Melnick and Field, and in which the bleaching effect of CNBr is entirely suppressed.

We have introduced certain modifications in the method of analysis which increase its sensitiveness and accuracy; these include the removal of interfering substances by preliminary adsorption on charcoal, control of period of hydrolysis and adjustments in the blank correction.

In conjunction with Dr. Harris we have confirmed again that the estimation of nicotinic acid, as such, in urine, in the absence of test-dosing (that is, measurement of 'resting level' of excretion) does give an indication of the past intake of nicotinic acid and hence of the anti-pellagra status of the subject: thus in deficiency in humans, in dogs and in guinea pigs the excretion of nicotinic acid falls either to zero or to a very low level[6]. Indeed, in ordinary circumstances it seems preferable to estimate the nicotinic acid rather than the trigonelline in a specimen of urine, because, unless special precautions have been taken to make the diet free of trigonelline, it may be excreted in large amounts as such in the urine, even when there has been no nicotinic acid in the diet and the subject is actually deficient. For test-dosing on the other hand ('saturation tests') the most satisfactory procedure would appear to be to administer nicotinamide and estimate both the trigonelline and the nicotinic acid excreted while the subject is kept on a controlled diet low in trigonelline. Our reason for this recommendation is the finding that when doses of nicotinic amide are given the product excreted in the urine is almost entirely trigonelline, while with a nicotinic acid dose both trigonelline and nicotinic acid (or nicotinic acid-like substances) are found. The possible explanation may be that nicotinic acid given by mouth has first to be converted by the organism into nicotinamide before being excreted as trigonelline, and any excess which escapes this conversion will be excreted as nicotinic acid-like substances. On the other hand, nicotinamide is utilized as such and there is no overflow of nicotinic acid-like substances, but only an increased excretion of trigonelline.

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[1] Ackermann, *Z. Biol.*, **59**, 17 (1912).
[2] Linneweh and Reinwein, *Z. physiol. Chem.*, **207**, 48 (1932).
[3] Linneweh and Reinwein, *ibid.*, **209**, 110 (1938).
[4] Sarett, Perlzweig and Levy, *J. Biol. Chem.*, **135**, 483 (1940).
[5] Melnick, Robinson and Field, *ibid.*, **136**, 131, 145 (1940).
[6] Harris, Kodicek and Wang, in the Press.
[7] Harris and Raymond, *Biochem. J.*, **33**, 2037 (1939).
[8] Melnick and Field, *J. Biol. Chem.*, **134**, 1 (1940).
[9] Melnick and Field, *ibid.*, **135**, 53 (1940).
[10] Swaminathan, *Ind. J. Med. Res.*, **27**, 417 (1939).

Sulphanilylguanidine

IN view of the present interest in the trial of sulphanilylguanidine for the treatment of bacillary dysenteries[1] it may be useful to direct attention to a convenient method for the preparation of this substance by the fusion of sulphanilamide with dicyandiamide which we described some years ago. In our original description of this reaction[2] it was assumed that the substance isolated was formed by the addition of the cyanamide group at the N4 position to give 4-sulphonamidophenylguanidine; $NH:(NH_2)CNHC_6H_4.SO_2NH_2$; actually addition takes place at the N1 position to give sulphanilylguanidine, $NH_2C_6H_4.SO_2NH.C(NH_2):NH$ completely identical with the product recently obtained by Marshall *et al.*[3] by the interaction of *p*-acetylaminobenzesulphonyl chloride and guanidine. Marshall's synthesis leaves no doubt about the correct constitution of the substance, which is confirmed also by the insolubility of the substance in alkali and the development of colour on diazotizing and coupling.

We are much indebted to Prof. E. K. Marshall, jun., who has kindly directed our attention to the identity of the substance produced by these two reactions.

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[1] Marshall, E. K., Bratton, A. C., Edwards, L. B., and Walker, E., *Bull. Johns Hopkins Hosp.*, **94** (January, 1941).
[2] Buttle, Dewing, Foster, Gray, Smith and Stephenson, *Biochem. J.*, **32**, 1101 (1938).
[3] Marshall, Bratton, White and Litchfield, *Bull. Johns Hopkins Hosp.*, **67**, 163 (1940).

Vitamin A in Canned Salmon

IN a communication in NATURE[1], Pyke and Wright have commented on the high values for the vitamin A contents of salmon-body and salmon-flesh oils given in the tables of Fixsen and Roscoe[2]. In their own experiments they have failed to detect any vitamin A by the antimony trichloride method