Selection of Iodinated Protein in the Young Mouse

THE suckling mouse absorbs antibody readily when whole immune serum is administered by mouth⁴ and transmission to the circulation is selective in the sense that antibodies prepared in a variety of species are characterized by different concentration quotients in the recipient plasma². During the course of the work with serum protein preparations labelled with iodine-131 in other species a criterion of the physiological integrity of the labelled material was sought which would be sensitive and readily determined. Recourse was therefore made to gut transmission in the young mouse.

The mice employed throughout the work were L.A.B. Greys, obtained by courtesy of Dr. Lane-Petter in 1955 and bred randomly since then. Preliminary experiments suggested the standard conditions of testing. Protein, labelled by the jetburette method³, was made up finally in 1 per cent solution. This was fed to 8-day old mice which were killed 3 hr. later, the blood collected, and the activity of the serum protein determined after tungstic acid precipitation.

Values are expressed as the ratio :

concentration quotient = $\frac{c.min.^{-1} ml.^{-1} serum protein}{c.min.^{-1} ml.^{-1} preparation fed}$

Fig. 1A presents the regressions of the concentration-quotient with dosage for two levels of iodination of aliquots of a frozen-dried stock preparation of rabbit globulin which was prepared by final precipitation from 12 per cent sodium sulphate⁴. It is apparent that a very sharp selection occurs between these two preparations, although they differ only in the level of iodine-127 per molecule of globulin.

Fig. 1B illustrates the relation of concentration quotient to iodination-level at a chosen dose of 0.03ml. fed, and is thus in effect a calibration curve for the testing of subsequent preparations of this protein



Fig. 1. Absorption of 1 per cent solutions of rabbit globulin from the gut of the 8-day old mouse. The means ± 2 standard error of groups of six mice are plotted; within each relationship each group was composed of one sibling of each of six litters. (A) Dosage relation: ---, regression where the mean iodination-level is 0.5 atom/mol. b = 4.4; --, regression where the level is 0.5 atom/mol. b = 4.9.4; -- mean concentration quotient of rabbit anti-sheep red cell serum from ref. 2. (B) The test calibration curve : dose fed 0.03 ml.

at any future date. It is clear that there is no significant difference between the concentration quotient at the mean iodination-level of 0.5 atom per molecule and that for antibody from whole rabbit serum at the same dosage (Fig. 1*A*), so that it is possible to take the concentration quotient of isotope at this level as being maximal for the stock preparation of globulin in these experimental conditions. Since the reduction of concentration quotient at the mean level of 3 atoms per molecule is already highly significant, the proposed test is clearly a sensitive one. It is thus possible, by feeding small groups of mice, to check the integrity of labile preparations in the course of a few hours.

Selection between molecules differing only in the number, and presumably the distribution, of labelled tyrosine residues, since it is so sharp, may suggest that some at least of the tyrosine residues of the molecular surface normally play a part in selection of the protein by the entoderm cell.

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¹ Morris, I. G., Proc. Roy. Soc., B, 148, 84 (1957).

² Hemmings, W. A., and Morris, I. G., *Proc. Roy. Soc.*, B, **150**, 403 (1959).

³ McFarlane, A. S., *Biochem. J.*, **62**, 135 (1956). ⁴ Kekwick, R. A., *Biochem. J.*, **34**, 1248 (1940).

Cæsium-137 in Toronto Milk during 1959

MEASUREMENTS of the changing cæsium-137 content of milk have been carried out in many parts of the world, particularly in the United States¹ and the United Kingdom². The Los Alamos group measured the activity of Ontario milk during 1958³. Here we present results of measurements on milk taken from the Toronto milk-shed during the period April 1959– February 1960.

The measurements were carried out using as gamma-ray detector a 5-in. sodium iodide crystal placed in the University of Toronto 'steel room'⁴. 670 gm. of dried skimmed milk, received within a few days of production, were placed in a thin copper vessel immediately below the crystal. Counting was normally for 75 min.

The only photo-peaks observed were at energies of 0.66 MeV. and 1.46 MeV., corresponding to cæsium-137 and potassium-40 activities. By comparison of the areas of these photo-peaks, and making suitable corrections⁴, the ratio casium-137/potassium of a sample is obtained. Calibration of the apparatus was carried out regularly using a weighed amount of potassium chloride in a standard copper vessel, and thus the potassium content of the milk could be determined, corrections once again being necessary for the differential absorption of the potassium gamma-rays in the potassium chloride and in the dried milk. It was in fact found that the potassium content of each sample of dried milk was approximately the same, the average value being 3.7 mgm. of potassium per gm. of dried milk.

The statistical error associated with the measurement of the potassium content of any one particular milk sample did not exceed 2 per cent.

The results for cæsium content during the period of observation, expressed in cæsium units (C.U.), that is, $\mu\mu c.$ cæsium-137 per gm. of natural potassium, are given in the lower part of Fig. 1. For comparison,