Developmental Changes in the Plasma Protein Pattern of the Mouse

KNOWLEDGE of the changes in the pattern of plasma proteins during the development of the mouse is desirable not only from the embryological but also from the genetical point of view, because of the existence of many inbred lines of this versatile laboratory animal.

Smithies's technique of starch-gel electrophoresis[1] with Poulik's discontinuous buffer system[2] was applied to blood plasma from strain *A* mice. Each sample was carried by a small strip of filter paper inserted in a slit in the starch-gel. This procedure apparently immobilizes the γ-globulins[1], so that the observations that follow do not concern this fraction.

Plasma from adult females at various stages of pregnancy gave twelve fractions in the regions under consideration (Fig. 1, 1).

The earliest embryos from which blood could be reliably collected were aged 12 days from conception. Many of the fractions of the adult are already present (Fig. 1, 2) but certain interesting differences can be noted. First, fraction 1 (albumin) is present in the early embryos at a very low concentration.

Secondly, fractions 2 and 3 of the adult are absent; but there is a second fraction slower than both the above, and by far stronger than albumin.

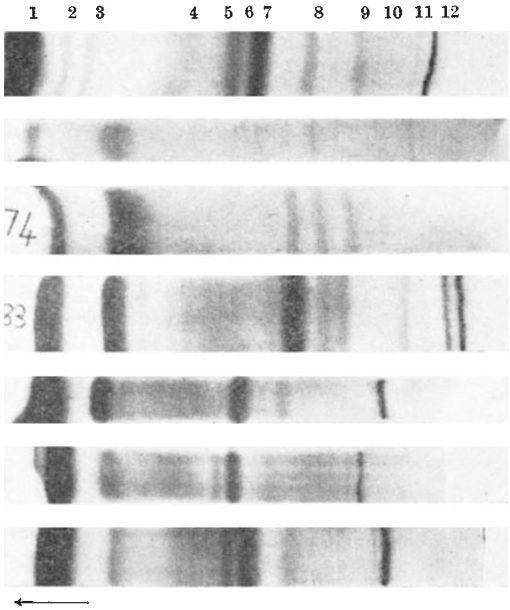

Fig. 1. Starch-gel electrophoresis pattern of plasma proteins of mouse. 1, 15-day pregnant female; 2, 12-day fœtus; 3, 15-day fœtus; 4, 19-day fœtus; 5, new-born; 6, 12-day-old young; 7, 18-day-old young

Thirdly, all other fractions are found in a concentration that is both low and about equal for all, that is, the bands in the transferrin region and the α_2-globulin are not stronger than the others, as in the adult.

The second fraction is still the strongest in the 15-day fœtus (Fig. 1, 3), but by the latter days of intrauterine life (Fig. 1, 4) and in the new born (Fig. 1, 5) albumin equals it in concentration, to exceed it by the twelfth day after birth (Fig. 1, 6).

The single second fraction persists after birth in the young, providing a contrast with the two fractions (2 and 3) of their mothers as tested during pregnancy.

Finally, the transferrin and slow α_2-globulin bands (6, 7, 12) do not reach their typical prominence over other fractions until a few days before birth (Fig. 1, 4).

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α-Galactosidase Activity of Rumen Bacteria

IN a recent search for bacterial α-galactosidases, Carrere, Lambin and Courtois[1] tested many organisms isolated from the intestinal tracts of a wide range of animals including several ruminants. Only one bacterial species, from the guinea pig, containing α-galactosidase was obtained. The authors commented on the apparent rarity of this enzyme in bacteria, although it is fairly widespread in yeasts and plant tissues[2]. Bacteria producing α-galactosidase might be expected in rumen micro-flora, particularly in animals grazing pasture, as small amounts of oligo- and poly-saccharides containing α-linked galactose are present in many pasture plants. In addition the leaves of plants such as red clover (*Trifolium pratense*) contain up to 1 per cent (dry wt. basis) of lipid bound galactose[3], present either as monosaccharide or 6-*O*-α-D-galactopyranosyl-D-galactose glycosidically joined through a β-link to the lipid glycerol[4]. The ability of rumen bacteria to release this α- and β-linked galactose has not been demonstrated. One rumen organism, *Streptococcus bovis*, appears to be a potential producer of α-galactosidase, as an extract from cells of one strain of this species has been shown able to hydrolyse melibiose[5]. The action of cell extracts from mixed rumen bacteria and *S. bovis* on various compounds containing α-linked galactose has, therefore, been investigated.

Mixed rumen bacteria, freed from protozoa and plant debris by repeated centrifuging, were prepared from the rumen contents of a cow feeding on fresh red clover. *S. bovis* cells (strain I)[6] were collected from cultures grown in a medium[6] containing glucose (2 per cent w/v) as sole carbohydrate. Bacterial cell extracts (25 ml. from 1–2 gm. wet wt. of cells) were prepared in acetate buffer (0·05 *M*, *p*H 6·0) and tested for carbohydrase activity by the procedures described previously[5], all incubations being done at 39° C. Total plant lipids were extracted from fresh cut red clover leaves with boiling 80 per cent (v/v) aqueous ethanol and, after removal of the ethanol, freed from sugars by suspending in water and centrifuging at high speed. Galactose was the only sugar detected after hydrolysis of the lipids in *N*-sulphuric acid for 1 hr. at 100° C. Approximately one-third of the plant lipid galactose in clover lipids is present as the disaccharide[3]. Mono- and di-galactosyl glycerol, prepared from the lipids by alkaline hydrolysis and column chromatography, were supplied by Mr. R. O. Weenink, Fats Research Laboratory, Department of Scientific and Industrial Research, Wellington.

Extracts from the mixed rumen bacteria readily hydrolysed sucrose, maltose, cellobiose, lactose, salicin and mono-galactosyl glycerol. Melibiose and methyl α-D-galactoside were also hydrolysed, but at a much slower rate. With raffinose and stachyose the rapid release of fructose, producing melibiose and