HISTOCHEMISTRY

Effect of Denervation on the Lactate Dehydrogenase Isozymes of Skeletal Muscle

The lactate dehydrogenase (LDH) isozyme pattern of many mammalian tissues has been shown to change with development[1,2]. Persistence of the fœtal LDH pattern in skeletal muscle has been noted in chickens with hereditary muscular dystrophy[3] and in children with myopathy[4]. In the work recorded here an adult skeletal muscle was denervated in order to ascertain the effect of an acquired lesion on the LDH isozyme pattern.

The soleus muscle of an adult guinea-pig was unilaterally denervated by severing the sciatic nerve at the trochanteric level under general anæsthesia. A portion of nerve 1·5 cm long was removed. The animal was then permitted free movement in its cage and killed after 11 weeks. The soleus muscles from both hind-legs were excised, homogenized, submitted to starch-gel electrophoresis, and incubated to demonstrate LDH activity, according to the methods described previously[5]. LDH isozyme patterns of soleus muscles from new-born and normal adult guinea-pigs were prepared in the same manner. The LDH activities of all extracts were measured in a spectrophotometer and equalized at 30,000 units/ml. by dilution before application to the gel.

Predominance of the faster-moving LDH isozymes (LDH 1 and LDH 2) was found to characterize normal adult guinea-pig soleus[6] (Fig. 1*C*). The pattern of new-born soleus differed from that of the normal adult (Fig. 1*B*). It showed less activity of the fast-moving isozymes and greater activity of the slow-moving isozymes. The pattern of the denervated soleus also differed from that of the normal adult and resembled the pattern of the new-born (Fig. 1*A*). The soleus from the unoperated side of the denervated animal had an isozyme pattern identical to that of normal adult soleus.

Microscopic examination of the denervated soleus revealed only slight infiltration of connective tissue in addition to neurogenic atrophy of muscle fibres.

These results were confirmed with three adult guinea-pigs and with new-born guinea-pigs from three different litters. The observations are in accord with a recent chemical analysis of the forms of LDH in rabbit soleus by Dawson *et al.*[7], who found that '*H*–LDH' (which predominates in the fast-moving isozymes) increases more rapidly in the first several weeks of life than '*M*–LDH' (which predominates in the slow-moving isozymes). Denervation was found by these workers to produce greater loss of the *H* type than of the *M*.

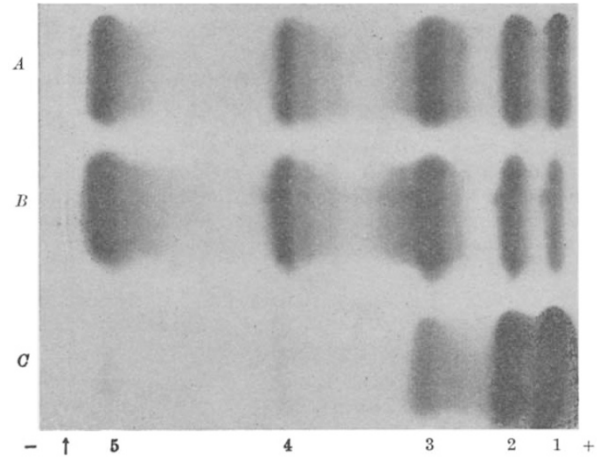

Fig. 1. Photograph of a starch gel showing electrophoretic patterns of guinea-pig muscles. The LDH activity of each extract was adjusted to 30,000 units/ml. before application to gel. Arrow indicates origin; LDH isozymes are numbered from anode. *A*, adult soleus denervated 11 weeks; *B*, new-born soleus; *C*, normal adult soleus. Note predominance of fast-moving isozymes in normal adult soleus. In denervated soleus, the fast-moving isozymes are decreased in intensity, the slow-moving isozymes are increased, and the pattern resembles that of the new-born

The work recorded here indicates that experimental denervation of muscle may cause reversion to the immature LDH isozyme pattern. Not only does the intensity of the fast-moving isozymes decrease in the reversion of the guinea-pig soleus pattern but also the intensity of the slow-moving isozymes increases.

Resemblance to the immature LDH isozyme pattern is, therefore, a non-specific effect of either myopathic or neurogenic change in skeletal muscle and occurs in hereditary and acquired conditions.

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RADIOBIOLOGY

Retention of ^{90}Sr in Lactating Rats

In previous investigations we were able to demonstrate that elimination of ^{90}Sr is enhanced in lactating rats[1–4]. Similar findings were reported by other investigators under various experimental arrangements and with various experimental animals[5–10]. To gain more quantitative information on Sr-metabolism in the course of lactation the following experiment has been performed.

Experimental animals were 32 albino rats of the 'Heiligenberg' strain, 15 weeks of age. 29 were dams carrying litters, the mean number of young being 9–10. Three virgin rats served as controls. Each rat received an intravenous injection of 2 μc. of ^{90}Sr–^{90}Y chloride in physiological saline and was killed 48 h thereafter. Time of injection varied with experimental groups so that 48-h retention could be evaluated for the end of gestation, the beginning of lactation and the end of lactation. Femora of dams and total litters were ashed dry. Radioactivities of samples of the ash from each femur were measured, and means were calculated for the litter. The experimental arrangement and results are recorded in Table 1.

As can be seen, ash-weights were elevated with respect to controls at the end of gestation and at the beginning of lactation. At the end of lactation they were lowered. Radioactivities of the femora were lower with all dams, the difference between groups 2 and 3 not being significant ($P > 0.05$). In an earlier experiment[2], different retentions of injected *Sr, compared with virgin controls, could not be established. With the present arrangement retention at the end of gestation was significantly lower. Diminution of radioactivity in the femora at days 15–17 post partum was highly significant. Radioactivities of the juveniles were roughly doubled for days 3–5 post partum compared to days 17–19 of gestation, and then once more for days 15–17 post partum. Therefore, participation of the litter on a dose of ^{90}Sr injected during lactation was growing continuously.

An approximation to the amount retained by the whole animal can be calculated from the figure derived from the femora by multiplication by a factor of 20. Retentions calculated for groups 1–4 are 51, 36, 42 and 17 per cent of the injected dose, respectively. Radioactivities retained in the biological system comprising mother and litter 48 h