CHAPTER 13: INFLUENCES OF TESTOSTERONE ON CONTRACTILE PROTEINS OF THE GUINEA PIG TEMPORALIS MUSCLE

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We have recently shown in the rat that the switch from neonatal to adult fast myosin is orchestrated by thyroid hormone (391); it is delayed in hypothyroid animals and switches precociously in hyperthyroid animals. In view of this hormonal effect on the development of fast muscle, we have been interested in other hormonal effects on myosin gene expression.

Testosterone is known to stimulate protein synthesis and influences muscle growth. However, comparatively little is known about androgenic influences on muscle structure or the contractile proteins. In the guinea pig, striking effects of castration and of testosterone administration have been described (394) on the size and metabolic enzymes of the fast twitch temporalis muscle. Gutmann et al. have reported that this muscle can be classified as "fast white" (low oxidative enzyme concentration) in the mature male and "fast red" (high oxidative enzyme concentration) in the mature female (395). This suggested to us that testosterone may alter the expression of myosin isoforms in the male temporalis since it is widely believed that the subdivision of fast fibers into fast red and fast white phenotypes is correlated with differences.

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in forms of myosin. For these reasons, and because the transition from IIa to IIb fibers has not previously been studied biochemically, we investigated the myosin isozymes in the male and female guinea pig temporalis, and have followed the development of this sexual dimorphism from late fetal life to maturity. The myosins in these muscles are compared with the myosin isozymes in two other testosterone sensitive muscles, the psoas and the masseter of the guinea pig.

At birth, a fundamental pattern of specialization emerges in both the male and female temporalis. With the myosin ATPase reaction virtually all fibers stain well after alkali preincubation (pH 10.4) (Figure 35a). After acid preincubation (pH 4.5) there is a continuous range of staining intensities between IIa, IIb and darkly stained IIc fibers (Figure 35b). As a consequence, discrimination between fiber types is difficult. Approximately 40% are IIa in type, 40% IIb and 20% are intermediate and IIc fibers. Small numbers of type I cells are also present. They are also found in the masseter at this stage and are presumably transformed into type II fibers soon after birth for they are rare in both muscles at 50 days and in the adult. There are no IIm fibers (396).

We have examined the early stages of fiber differentiation using a monoclonal antibody, 2B6, to rat embryonic myosin. This antibody cross reacts with an embryonic myosin in all mammals we have studied (390). In the neonatal temporalis there is a spectrum of fiber staining intensities with this antibody (Figure 35d), which is consistent with the myosin ATPase results, and suggests that the heterogeneity of ATPase fiber staining at birth is due to the coexistence of varying proportions of embryonic and adult fast myosins within each of the developing fiber types. Antibody staining is most intense in small IIb and IIc fibers suggesting that IIa fibers are the first to differentiate and that hypertrophy is correlated with elimination of embryonic myosin. By 5 days of age, only occasional fibers react with the monoclonal antibody.

Sexual dimorphism first becomes evident by 30 days, and by 50 days the male and female temporalis are clearly distinct in terms of fiber size (Figure 36) and histochemical differentiation. In the female, IIa and IIb fibers are approximately the same size in cross sectional area. The relative proportions of these two fiber types are comparable to the ratio at birth. Type IIc fibers are infrequent. The guinea pig grows throughout life with the result that the temporalis slowly hypertrophies (Figure 36) in both sexes. However at 18 months of age the pattern of fiber differentiation is approximately the same as at birth and 50 days in the female (Figure 37). This stability of specialization