THE DEVELOPMENT AND GROWTH OF SKELETAL MUSCLE IN FETAL AND NEONATAL LAMBS

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GENERAL ABSTRACT

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The objective of these studies was to identify and investigate factors involved in the regulation/control of fetal growth and development in the sheep, with particular emphasis on cellular development of skeletal muscle.

Two models with the potential to impose growth-restriction on the developing fetus without invasive manipulation of the fetal environment were used in this series of studies. First, ewes mated out-of-season generally give birth to offspring with lower body weight than comparable offspring born to ewes mated in their natural breeding season. Fetal growth restriction in this situation is associated with impaired placental development in the out-of-season ewes which is evident by 84 days of gestation. Despite impaired placental growth, ewes mated out-of-season did not, in this study, consistently give birth to low-birth-weight offspring. Although differential effects on myofibre morphology were observed between fetuses from each group, the lack of differences in muscle weights and inconsistent effects on body weight indicated that this comparison proved an unreliable model with which to study fetal muscle growth and development.

The second model involved the comparison of twins versus singles. Twin lambs are consistently lighter than single lambs as a result of maternal constraint characterized by restricted placental size per fetus. Coupled with low birth weights, the growth-restricted twin lamb also had smaller hindlimb muscles compared to singles. Maternal constraint in this situation not only had a negative influence on body and muscle weight, but myofibre hypertrophy was also retarded as indicated by smaller myofibre cross-sectional area. The *adductor* muscle DNA content, and total nuclei

number in selected hindlimb muscles, were lower in twins than in singles. Myofibre number did not differ between ranks. An immunohistochemical technique involving the muscle-specific regulatory factor MyoD allowed the identification of myogenic precursor cell nuclei, which are likely to be satellite cell nuclei in muscles from fetuses in late gestation or early postnatal lambs. Bromodeoxyuridine proved to be unsatisfactory as a marker of actively dividing cells because it did not cross the feto-placental barrier in sheep. However, MyoD was a useful marker of active satellite cells. MyoD-positive nuclei were less abundant in hindlimb muscles of twins than in singles suggesting differential effects of growth restriction on cell cycle activity. The pattern of expression of this factor during development suggests that MyoD may also have an important role in late fetal and postnatal muscle growth.

These results illustrate that growth restriction during late gestation can have important consequences for birth size and skeletal muscle hypertrophy. The observation that myofibre number is not affected suggests that the full complement of fibres has been reached prior to any major nutritional impact which results in growth restriction. The delayed myofibre hypertrophy observed in twin lambs as compared to singles, coupled with lower total DNA content and fewer myogenic precursor nuclei, suggest that the late fetal developmental period is important for muscle growth and the attainment of an adequate birth weight. This result also shows that this period of development has important implications for postnatal muscle growth and may be important in determining ultimate mature muscle mass and postnatal growth potential.

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