



## Impact of Fetal Size on Myogenesis and Pax7+ Progenitor Cell Populations

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### Objectives

Two experiments were conducted to determine the effect of fetus size on primary and secondary muscle fiber number and size, and Pax7+ progenitor cell number.

### Materials and Methods

In experiment 1, gravid gilts ( $n = 36$ ; PIC 1050  $\times$  327) were harvested at d-60 of gestation and the smallest (SM), median (ME), and largest (LG) male fetuses were removed for muscle morphometric analysis based on crown-to-rump length. In experiment 2, gravid gilts ( $n = 24$ ) were harvested at d-90 of gestation and fetal weight was used to categorize SM, ME, and LG fetuses. Whole muscle cross-sectional area (CSA) of the *Longissimus* muscle of each fetus was measured at the fifth rib and first lumbar vertebrae. Tissue samples corresponding to each location were cryopreserved, cut into 10  $\mu\text{m}$  sections, and subjected to immunohistochemistry to identify primary and secondary muscle fibers. Muscle fiber number, CSA, and number of Pax7+ progenitor cells were determined.

### Results

On d-60 of gestation LG fetuses had greater whole muscle CSA than ME and SM fetuses ( $P < 0.05$ ), and ME fetuses had greater ( $P < 0.01$ ) whole muscle CSA than SM fetuses. Muscles of LG and ME fetuses possessed more primary muscle fibers than SM fetuses ( $P = 0.03$ ), but were similar ( $P = 0.83$ ) to each other. Primary muscle fiber CSA tended to be greater in muscles of LG and ME fetuses compared to SM ( $P < 0.09$ ), and CSA was similar ( $P = 0.73$ ) between LG and ME fetuses. Muscles of LG fetuses had

more ( $P = 0.03$ ) Pax7+ progenitor cells than SM fetuses, and muscles of ME fetuses had similar numbers of Pax7+ cells as SM and LG fetuses ( $P > 0.19$ ). On d-90 of gestation, LG fetuses had greater whole muscle CSA than ME and SM fetuses ( $P < 0.01$ ), and ME fetuses had greater ( $P < 0.01$ ) whole muscle CSA than SM fetuses. Total primary muscle fiber number and CSA were not impacted by fetal size ( $P > 0.20$ ). Muscles of LG fetuses had more ( $P = 0.03$ ) total secondary muscle fibers compared to SM fetuses, and ME fetuses had a similar number of secondary fibers as LG and SM fetuses ( $P > 0.25$ ). Secondary muscle fiber CSA was greater in muscles of LG fetuses compared to SM and ME fetuses ( $P < 0.01$ ). Additionally, muscles of ME fetuses exhibited greater ( $P < 0.01$ ) secondary muscle fiber CSA than SM fetuses. Total number of Pax7+ progenitors in muscles of LG and ME fetuses were greater than SM fetuses ( $P < 0.01$ ), and muscles of LG fetuses tended to possess more ( $P = 0.06$ ) Pax7+ progenitors than ME fetuses.

### Conclusion

Larger fetuses have greater whole muscle CSA at both d-60 and d-90 of gestation. At d-60 of gestation this is attributed to an increase in number and size of primary muscle fibers. At d-90, whole muscle CSA differences between size categories were not due to differences in primary myogenesis, but were due to increases in secondary muscle fiber number and CSA. Differences in whole muscle CSA between ME and SM fetuses and LG and ME fetuses were due solely to increases in secondary muscle fiber CSA. These data would indicate that SM and ME fetuses are on a delayed trajectory of myogenesis when compared to a LG fetus. As fetal size increases the total number of Pax7+ progenitors increases, which may be responsible for the increased efficiency of large birthweight piglets postnatally.