Changes of the histochemical properties and meat quality traits of porcine muscles during growth. I) Effect of feed restriction in pigs slaughtered at the same age and varying body weight

Abstract
Twenty-four Swiss Large White barrows from six litters were given either ad libitum (A) or restrictive (R) access to a grower diet from 21 to 60 kg body weight (BW). At d 113 of age six pigs of the A- (BW = 62 kg) and six siblings of the R-group (BW = 51 kg) were slaughtered. The remaining 12 barrows were fed a finisher diet from 60 kg BW until slaughter. The day of slaughter barrows were 154 d old and the BW of the A- and R-group averaged 100 and 87 kg, respectively. The histochemical properties of myofibers as well as meat quality traits of the longissimus muscle (LM) and light portion of the semitendinosus (STL) were assessed. Muscle fibers were stained and classified based on the stain reaction as slow oxidative (SO), fast oxidative-glycolytic (FOG), and fast glycolytic (FG), and fiber area and distribution were determined. Regardless of the age at slaughter, pigs of the R-group had smaller ($P \leq 0.04$) SO and FG fibers in the LM and STL, and smaller ($P < 0.01$) FOG fibers in the STL than their siblings of the A-group. The STL had more ($P = 0.03$) FOG and fewer ($P = 0.04$) FG fibers in pigs of the R- than pigs of the A-group. The muscles of pigs in the R-group were less ($P \leq 0.03$) tender and percentages of cooking loss were higher ($P \leq 0.08$) than of pigs in the A-group. In conclusion, hypertrophy as well as differentiation of muscle fibers was affected by the restricted nutrient supply when pigs had the same age at slaughter. However, the extent of the dietary impact varied among muscles.

Key Words: Age at slaughter, Meat Quality, Muscle Fibers, Pig

Introduction
Quantitative and qualitative aspects of postnatal nutrition have a major effect on muscle development through their effect on growth rate and body composition. The dietary impact can vary depending on the stage of development and age of the pig. HARRISON et al. (1996) found that severe feed restriction for 4 weeks post-weaning
significantly impaired myofiber hypertrophy. Concomitantly they found higher percentages of type I fiber in the red rhomboideus but not in the white longissimus (LM) muscle. CANDEK-POTOKAR et al. (1999) suggested that the aforementioned dietary effect on myofiber size was the result of the markedly lower BW because in heavier pigs (100 and 130 kg BW) slaughtered at the same BW feed restriction neither affected the size nor the distribution of the myofibers in the LM. However, in pigs subjected to mild feed restriction and slaughtered at 55 kg BW SOLOMON et al. (1988) reported smaller slow oxidative (SO) and fast glycolytic (FG) fibers as well as fewer FG and more fast oxidative-glycolytic (FOG) fibers in the LM. Thus, it is unclear whether the effect of restricted nutrient supply differs at different developmental stages and depends on the duration of nutrient restriction. Because metabolic differentiation and myofiber hypertrophy is most intense up to four months of age (60 kg BW) (LEFAUCHEUR and VIGNERON, 1986; ONO et al., 1995) the effect of ad libitum versus restricted (80% of ad libitum) feed allowance on contractile and metabolic differentiation of myofibers after the growing and after the finishing period at a given age (different BW) were examined. In order to assess the dietary impact on myofiber development, two porcine muscles were examined, which are known to have a similar myofiber composition but to differ in their allometric growth ratios.

Material and Methods
Swiss Large White barrows (n = 24) originating from six litters were blocked by litters and assigned from within litter to the four treatment groups. The pigs had either ad libitum (A) or restricted (R) access to a standard growing-finishing diet for the total experimental period. Feed restriction was aiming to achieve an ADG of 750 g from 20 to 100 kg BW. From 20.7 ± 0.67 (start of the experiment) to 60 kg BW, barrows were offered a grower diet (crude protein: 201 g, DE: 15.5 MJ/kg dry matter). At 113 ± 2.4 d of age six pigs (one per litter) of the ad libitum (A-113) and six of their siblings of the restricted group (R-113) were slaughtered. The BW at slaughter of the A-113- and R-113-pigs averaged 62.1 ± 1.82 and 51.0 ± 1.81 kg, respectively. The remaining 12 barrows were offered a finisher diet (crude protein: 166 g, DE: 15.4 MJ/kg dry matter) from 60 kg BW until slaughter. The day of slaughter barrows were 154 ± 2.6 d old and the BW of the ad libitum (A-154) and restricted group (R-154) averaged 99.5 ± 2.45 kg and 86.6 ± 2.48 kg, respectively.

The pigs were reared in individual pens (2.6 m²/pig) on a concrete floor in environmentally controlled buildings (22 °C and 60 to 70% relative humidity). Barrows from the R-group were fed individually twice a day and had free access to water. The total daily feed allowance was adjusted weekly according to the BW (BOLTSHAUSER et al., 1993). Pigs of the A-group were weighed and feed disappearance was recorded weekly. The day prior to slaughter, feed was withheld from pigs 12 h before transportation to the research station abattoir. Slaughter and dissection were carried out according to the Swiss Pig Performance Testing Station (MLP, Sempach, Switzerland) meat cutting standards as previously described by BEE et al. (2004).

Within 40 min after exsanguination, the longissimus (LM) and semitendinosus muscle were removed from the right side of each carcass. Muscle samples for histochemical analyses were excised from the light (STL) portions of the semitendinosus and anterior
to the 10th rib location of the LM. One piece (approximately 1 × 1 × 3 cm) of each muscle was immediately fixed on a labeled flat stick, rolled in talcum powder, and immediately frozen in liquid nitrogen and stored at –80 °C. Histochemical analysis were performed as described by BEE (2004).

From the same samples used for histochemical analyses, two 1.5-cm thick LM chops were cut at the 12th rib level and two slices (approximately 70 g each) were obtained from the STL. From the muscle samples initial and ultimate pH, drip loss percentages after 72 h, thaw and cooking losses, and shear force values were determined as described by BEE et al. (2004).

Data were analyzed with the MIXED procedure of SAS (SAS Inst. Inc., Cary, NC). The model used for the analyses of muscle fiber characteristics and meat quality traits included feeding regimen and age at slaughter and the respective interactions as fixed effects and litter as the random effect. When interactions were statistically significant at \( P < 0.05 \), least squares means were separated using the PDIFF option. Pearson correlation coefficients between histological properties and meat quality traits for each muscle were calculated at 113 and 154 d of age.

Results and Discussion

Muscle Fiber Area and Distribution

In the present study, myofiber hypertrophy was delayed \((P \leq 0.05\) for each; Figure 1) in both muscles at 113 and 154 d of age in pigs of the R- compared with those of the A-group. The smaller myofiber size was paralleled by smaller LM area and smaller ST girth (data not shown). Except for the FOG fibers in the LM, these findings are in agreement with the general observations that undernutrition delay the hypertrophic growth rate of all myofiber types when comparison are made at the same age (HARRISON et al., 1996; LEFAUCHEUR et al., 2003). The size of SO in the LM and the size of FOG and FG fibers in the LM and STL were larger \((P < 0.01\) in the older and heavier pigs (A-154 and R-154) than in their younger and lighter siblings (A-113 and R-113).
and R-113). The lacking interactions between feeding regime and age at slaughter indicated that for A- and R-pigs changes in myofiber size were not ($P \leq 0.31$) different from 113 to 154 d of age. Neither the dietary nutrient supply nor the age at slaughter affected the fiber type distribution of the LM. By contrast, the STL of R-pigs had fewer ($P = 0.04$) FG (60 vs. 70%) and more ($P = 0.03$) FOG (32 vs. 26%) fibers than the STL of A-pigs. The latter findings are in agreement with results reported by HARRISON et al. (1996) who found larger amounts of oxidative fibers in the rhomboideus muscle after a period of reduced energy availability. Regardless of the dietary treatments, myofiber type distribution in both muscles did not change from 113 to 154 d of age, which confirms results reported by ONO et al. (1995) who showed that the most intense changes in myofiber type distribution occurred from 20 to 60 kg BW.

**Meat Quality Traits**

Regardless of the age at slaughter, shear force values were higher ($P < 0.05$) in the LM (4.5 vs. 3.7 kg) and STL (4.0 vs. 3.3 kg), and cooking losses of the LM were higher (18.3 vs. 13.7%; $P < 0.01$) in R- than A-pigs. In agreement with results of previous studies (HENCKEL et al., 1997; CANDEK-POTOKAR et al., 1999) the differences in the shear force values were related to neither the size nor the distribution of the myofibers. This is not surprising because pork tenderness depends primarily on the extent of postmortem proteolysis (MELODY et al., 2004). Recent results showed that restricted feeding reduced in vivo protein turn-over at the time of slaughter, which then negatively affected tenderization processes during the conversion of muscle to meat (KRISTENSEN et al., 2002). Regardless of the feeding regime, pH at 30 min (6.3 vs. 6.2) and 24 h postmortem (5.6 vs. 5.4) in the LM and at 24 h postmortem (5.7 vs. 5.6) in the STL were higher ($P = 0.01$) in younger than in older pigs. A determinant factor affecting initial and ultimate pH is the amount of stored glycogen in the muscle at slaughter (BENDALL and SWATLAND, 1988). The latter might partly depend on the size and type of myofibers (FERNANDEZ et al., 1995). In the LM the size of FOG fibers was positively correlated with initial and ultimate pH ($r = 0.63$ for each; $P < 0.03$) at 113 d of age and the size of SO ($r = 0.61; P = 0.05$) and FOG ($r = 0.58; P = 0.06$) fibers were positively correlated with pH 24 h postmortem at 154 d of age. In the STL pH values 30 min postmortem were positively correlated with higher percentages of SO ($r = 0.63; P = 0.04$) and FOG ($r = 0.61; P = 0.05$) fibers and negatively correlated with the percentages of FG ($-0.70; P = 0.02$) fibers at 113 d of age. Because within experimental treatments myofiber type distribution was unaffected by the age at slaughter the larger increase in size of the FG (+36%) compared with the SO (+27%) and FOG (+32%) fibers might partly explain the observed drop in the muscle pH from younger to older pigs. Although in both muscles the FG fibers were larger and in the STL the percentages of FG fibers were higher in pigs of the A- compared to the R-group these differences seemed not to be sufficient to affect the pH values between the treatments.

In conclusion for pigs at the same age, when slaughter weight was lower (feed restriction) the cross-sectional areas of the myofibers were smaller except for the FOG fibers in the LM. Furthermore, the STL of pigs subjected to feed restriction was more oxidative. These findings indicated that the impact of feed restriction on hypertrophic growth and differentiation vary among myofiber types and muscles.
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