# Fiber-type proportions and diameters in the longissimus muscle of beef heifers undergoing catch-up (compensatory) growth

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Yambayamba, E. and Price, M. A. 1991. **Fiber-type porportions and diameters in the longissimus muscle of beef heifers undergoing catch-up (compensatory) growth.** Can. J. Anim. Sci. **71**: 1031–1035. Fifty-three Hereford crossbred heifers ($211 \pm 28$(mean$\pm$SD) kg, $197 \pm 13$ d of age at day 1) were used to study the effects of mild, chronic feed restriction followed by refeeding on some longissimus muscle fiber characteristics. Five animals were slaughtered on day 1 for analysis of initial muscle fiber characteristics. The remaining 48 animals were randomly penned in groups of 6 and assigned to treatments as follows: three pens to ad libitum feeding; three pens to 2 mo of feed restriction followed by refeeding, and two pens to 4 mo of feed restriction followed by refeeding. Animals in one ad libitum and one restricted pen were slaughtered after 2 mo and those in one pen from each treatment after 4 mo, and the remainder at the final slaughter weight of about 410 kg. Two months of feed restriction had no effect on the proportions of longissimus muscle fiber-types, "red" ($\beta$R), "white" ($\alpha$W), and "intermediate" ($\alpha$R), but fiber diameters were smaller ($P < 0.05$) in the restricted than in the ad-libitum-fed animals. Four months of feed restriction was associated with a relatively higher ($P < 0.05$) proportion of $\beta$R fibers and lower ($P < 0.05$) proportion of $\alpha$W fibers than ad libitum feeding. Muscle fiber diameters were larger ($P < 0.05$) in the ad-libitum-fed than in the restricted heifers. No significant feeding treatment differences were found in fiber-type proportions or fiber diameters at the final slaughter weight.

Key words: Heifers, feed restriction, realimentation, muscle fibers, fiber-type, compensatory growth

Yambayamba, E. et Price, M. A. 1991. **Proportion et diamètre des différentes fibres du muscle longissimus chez des génisses à viande en croissance de rattrapage.** Can. J. Anim. Sci. **71**: 1031–1035. Cinquante-trois génisses croisées pesant en moyenne $211 \pm 28$ kg et âgées de $197 \pm 13$ jours au début de l'expérience ont été utilisées dans une étude sur les effets d'un rationnement léger prolongé suivi d'un retour à l'alimentation à volonté sur certains caractères des fibres du muscle longissimus. Cinq bêtes étaient abattues au premier jour de l'expérience pour établir les caractères initiaux des fibres musculaires. Les 48 autres étaient groupées par lots de 6 selon les traitements suivants: alimentation à volonté (trois groupes), deux mois de rationnement suivis de retour à l'alimentation à volonté (trois groupes), quatre mois de rationnement puis retour à la normale (deux groupes). Un lot de chaque traitement était abattu au bout de deux et de 4 mois, ainsi qu'au terme de l'expérience (poids marchand de 410 kg). Deux mois de rationnement n'ont pas eu d'effet sur la proportion des diverses fibres du longissimus ($\beta$R, $\alpha$W et $\alpha$R) mais leur diamètre était plus petit ($P < 0{,}05$) en régime rationné qu'en alimentation libre. Un rationnement de 4 mois s'est traduit par une proportion relativement plus forte ($P < 0{,}05$) de fibres $\beta$R et plus faible ($P < 0{,}05$) de fibres $\alpha$W que dans le régime d'alimentation à volonté. Le diamètre des fibres musculaires était plus gros ($P < 0{,}05$) dans le régime d'alimentation à volonté. Au poids final d'abattage, il n'y avait plus de différence significative dans les proportions des divers fibres du muscle.

Mots clés: Génisses, rationnement, retour à l'alimentation à volonté, fibres musculaires, type de fibres, croissance de compensation

The ways of classifying muscle fibers include one based on the ATPase activity of their myosin (Arnold and Meyer 1988). On this basis, three fiber-types can be identified: "red" (βR) fibers, "white" (αW) fibers, and "intermediate" (αR) fibers. Red fibers are physiologically slow, with high oxidative and low myosin ATPase activities, and white fibers are fast, with high glycolytic and myosin ATPase activities (Moody and Cassens 1968). The intermediate fibers have a mixed oxidative-glycolytic activity.

Transformations from one fiber-type to another can occur (Joubert 1956), the transformation of αR to either αW or βR fibers being well established (Ashmore et al. 1972; Pette 1984). Factors such as the breed of the animal and its live weight, gender, and age are known to influence fiber-type transformations (Joubert 1956; Johnston et al. 1981); environmental factors such as nutrition may also influence fiber-type transformations (Moody et al. 1980). The objective of this study was to investigate the effects of feed restriction followed by refeeding on muscle fiber-type proportions and diameters in young heifers.

## MATERIALS AND METHODS

The animals used in the study were those described previously (Yambayamba and Price 1991). Briefly, following slaughter of a randomly selected initial group (INIT) of five heifers at 211±28 (mean± SD) kg and 197±13 d of age, the remaining 48 heifers were randomly allocated to treatment groups: ad libitum access to feed to a market weight of about 410 kg (ADLIB); 2 mo of feed restriction followed by realimentation ad libitum to market weight (2REST); and 4 mo of feed restriction followed by realimentation ad libitum to market weight (4REST). After the first 2-mo period, six ADLIB and six 2REST heifers were slaughtered; after the second 2-mo period six ADLIB, six 2REST and six 4REST heifers were slaughtered, and at market weight (the end of the third period) the remaining six ADLIB, six 2REST, and six 4REST heifers were slaughtered.

On the day following slaughter, two muscle samples (each 1 $cm^3$) were removed from the center of the 11th rib portion of the longissimus muscle of each carcass for fiber typing. A third sample (about 0.2 g) was removed from the same site, fixed in 10% formalin, and stored at 4°C for later fiber diameter determination.

The two 1-$cm^3$ samples were mounted on cryostat chucks within an hour of removal, with the muscle fibers oriented perpendicularly to the path of movement of the cutting blade. They were mounted with embedding medium and quick frozen in isopentane, cooled to −160°C in liquid nitrogen and stored overnight at −20°C. The following morning, 8-μm cross sections were cut, mounted on microscope slides (two slides per sample), and air-dried at room temperature for 30 min before staining for alkali-stable ATPase (Guth and Samaha 1970).

The following day, a single randomly selected field from each slide was photographed at 10× magnification using a Nikon Leitz System camera (Wetzlar, Germany) mounted on a Leitz–Dialux 20 EB microscope (Wetzlar, Germany). Using the micrographs, all the fibers in the field were classified on the basis of color into one of the three fiber types, βR, αW, and αR.

Fiber diameters were measured following homogenization of the 0.2-g sample in 5 mL physiological saline (Hegarty and Naude 1970) at 900 rpm for 5–10 s, using a Polytron homogenizer. A drop of the homogenate was transferred to a hanging-drop slide with a 15-mm-diameter well. The homogenate was covered with a cover slip and while still fresh, 50 randomly selected fibers were measured at their greatest width with an ocular micrometer.

### Statistical Analysis

The muscle fibers for each animal were counted and the number of each type expressed as a percentage of the total in the field. Muscle fiber diameters for individual animals were similarly recorded. Analysis of variance (Harvey 1960) for treatment effects on muscle fiber-types and diameters used the following model:

$$Y_{ijk} = \mu + F_i + T_j + FT_{ij} + E_{k(ij)}$$

where:

$Y_{ijk}$ = muscle fiber proportions or diameters for each individual,
$F_i$ = feeding treatment,
$T_j$ = time period,
$FT_{ij}$ = feeding treatment × time period interaction,
$E_{k(ij)}$ = random error term.

The Student- Newman–Keuls procedure was used to separate the means when significant ($P<0.05$) main effects were found.

## RESULTS AND DISCUSSION

Despite a significant gain in live and muscle weight by both the ADLIB and 2REST heifers during the first 2-mo period, there were no changes in muscle fiber type proportions (Table 1). Reid et al. (1980) and Moody et al. (1980) have shown that normal growth is associated with an increased proportion of αW fibers, while feed restriction that inhibits growth is associated with an increase in the proportion of βR fibers. Such a trend was not evident in the first 2 mo of feed restriction, though there was an effect ($P<0.001$) on fiber diameter (Table 1). The 2REST heifers had significantly larger muscle fibers than the INIT group, and the ADLIB heifers had greater ($P<0.05$) muscle fiber diameters than the 2REST group, reflecting their live weights.

Four months of feed restriction was associated with significant changes in muscle fiber-type proportions and fiber diameters (Table 2). The 4REST heifers had relatively more ($P<0.05$) βR fibers in the longissimus muscle than the INIT animals, which in turn had relatively more ($P<0.05$) than the ADLIB and 2REST animals. Conversely, the proportion of αW fibers was lower ($P<0.05$) in the 4REST and INIT animals than in the ADLIB and 2REST animals. The proportion of αR fibers was higher ($P<0.05$) in the INIT animals than in the other groups.

Under normal feeding conditions (ADLIB) and positive growth, the proportion of αW fibers would be expected to increase as the animals gained weight (Holmes and Ashmore 1972). The results also indicate that though feed restriction (4REST) inhibited this change, 2 mo of refeeding (2REST) was enough to allow catch-up. It can be seen from Tables 1 and 2 that there was a decrease in the proportion of

Table 1. Least-squares means (±SE) of longissimus muscle fiber-type proportions and diameters of heifers before (INIT) and after being fed an ad libitum (ADLIB) or restricted (2REST) ration for 2 mo

| | INIT | ADLIB | 2REST | P |
|---|---|---|---|---|
| No. of animals | 5 | 6 | 6 | |
| Slaughter wt (kg) | 211±17*a* | 296±23*b* | 246±12*b* | 0.02 |
| Slaughter age (d) | 199±4*a* | 255±5*b* | 254±5*b* | 0.001 |
| **Fiber proportion (%)** | | | | |
| Red (βR) | 27.6±1.1 | 28.2±1.6 | 31.8±2.3 | 0.25 |
| White (αW) | 44.4±3.0 | 49.4±2.3 | 46.4±0.9 | 0.30 |
| Intermediate (αR) | 28.0±3.2 | 22.4±1.5 | 21.8±1.7 | 0.14 |
| Fiber diameter (μm) | 38.2±1.5*a* | 68.5±1.9*c* | 46.0±1.4*b* | 0.001 |

*a,b*Means within a row followed by a different letter differ significantly ($P<0.05$).

Table 2. Least-squares means (±SE) of longissimus muscle fiber-type proportions and fiber diameters of heifers before (INIT) and after being fed an ad libitum for 4 mo (ADLIB), restricted for 2 mo and realimented for 2 mo (2REST), or restricted for 4 mo (4REST)

| | INIT | ADLIB | 2REST | 4REST | P |
|---|---|---|---|---|---|
| No. of animals | 5 | 6 | 6 | 6 | |
| Slaughter wt (kg) | 211±17*a* | 343±4*b* | 314±7*b* | 244±12*a* | 0.001 |
| Slaughter age (d) | 199±4*a* | 315±4*b* | 317±5*b* | 311±5*b* | 0.001 |
| **Fiber proportion (%)** | | | | | |
| Red (βR) | 27.6±1.1*b* | 21.0±0.6*a* | 23.9±1.1*a* | 34.9±1.1*c* | 0.001 |
| White (αW) | 44.4±3.0*a* | 57.0±2.3*b* | 55.4±2.0*b* | 44.0±0.8*a* | 0.001 |
| Intermediate (αR) | 28.0±3.2*b* | 22.0±0.8*a* | 20.7±1.4*a* | 21.1±0.6*a* | 0.03 |
| Fiber diameter (μm) | 38.2±1.5*a* | 86.8±0.9*d* | 77.0±0.1*c* | 52.3±0.2*b* | 0.001 |

*a–d*Means within a row followed by a different letter differ significantly ($P<0.05$).

βR fibers from 28.2 to 21.0% and from 31.8 to 23.9%, and an increase in the proportiion of αW fibers from 49.4 to 57.0% and from 46.4 to 55.4% in the ADLIB and 2REST animals, respectively. No significant changes were found in the proportions of αR fibers in these two treatments. On the contrary, there was a significant increase in the proportion of βR fibers, whereas the proportion of αW fibers remained relatively constant in the 4REST animals.

As expected, the mean muscle fiber diameter in the longissimus muscle of the ADLIB was greater ($P<0.05$) than in the 2REST animals, which was greater ($P<0.05$) than in the 4REST animals, which in turn was greater ($P<0.05$) than in the INIT animals. When the animals were slaughtered at about 410 kg, no significant differences were found among treatments in any of the muscle fiber-type proportions or muscle fiber diameters (Table 3). Thus it can be concluded that the realimentation schedule in this study allowed complete recovery to "normal" proportions of muscle fiber-types.

There was no difference ($P>0.05$) in the mean muscle fiber diameters among treatments at the final slaughter weight, indicating that during catch-up growth, the muscle fibers in the longissimus muscle also recovered sufficiently in size to catch up with the "normal" muscle fiber.

Considering the whole experimental period, the rate of increase in mean muscle fiber diameter in the ADLIB heifers was high during the first 2 mo, and slowed down as time progressed. As for the 2REST animals, the increase was slow during the first 2 mo, very rapid during realimentation, and slow again in the last phase of the experiment. In the case of the 4-mo restricted animals, the increase was slow in the first 4 mo and then more rapid during realimentation. Clearly, muscle fiber diameter is a function of total muscle weight, which is in turn a function of live weight.

## Significant Interaction Effects

The proportions of βR and αW fibers and muscle fiber diameter were all affected by significant treatment × period interactions. At the end of the second period (4 mo), the proportion of βR fibers was higher whereas that of αW fibers was lower in the 4REST animals than in the ADLIB and 2REST animals. During the third period (4 mo to slaughter weight), the proportions of βR and αW fibers significantly decreased and increased, respectively, in the 4REST animals, but remained relatively constant in the ADLIB and 2REST animals. The result was a nonsignificant treatment effect in the proportions of these fiber-types at the final slaughter weight.

The mean muscle fiber diameters were all different among treatments at the end of the second period. During the third period the increase in the muscle fiber diameter was greater in the 4REST than in the 2REST animals, and was in turn greater in the 2REST

Table 3. Least-squares means (±SE) of longissimus muscle fiber-type proportions and fiber diameters of heifers after being fed ad libitum (ADLIB), or following 2 mo (2REST) or 4 mo (4REST) of feed restriction to a market weight of about 410 kg

| | ADLIB | 2REST | 4REST | *P* |
|---|---|---|---|---|
| No. of animals | 6 | 6 | 6 | |
| Slaughter wt (kg) | 414±19 | 419±13 | 408±13 | 0.88 |
| Slaughter age (d) | 373±8*a* | 401±5*b* | 422±7*b* | 0.001 |
| **Fiber proportion (%)** | | | | |
| Red (βR) | 22.4±0.9 | 24.0±1.4 | 25.3±0.7 | 0.18 |
| White (αW) | 55.5±1.0 | 55.2±1.6 | 53.7±1.2 | 0.60 |
| Intermediate (αR) | 22.1±1.0 | 20.7±1.4 | 21.0±1.4 | 0.74 |
| Fiber diameter (μm) | 91.7±3.3 | 84.0±3.6 | 81.2±3.0 | 0.10 |

*a,b*Means within a row followed by a different letter differ significantly ($P<0.05$).

than in the ADLIB animals. The result was that at the final slaughter weight there were no significant differences.

## CONCLUSIONS

Overall, it is concluded from the present study that feed restriction of growing beef heifers is associated with a relatively higher proportion of red fibers and a lower proportion of white fibers than ad libitum feeding. Feed restriction in the present study appeared to have less effect on the proportion of intermediate fibers. On the other hand, muscle fiber diameter was affected in a shorter period, indicating a direct relationship between fiber size and live weight. On refeeding, relatively more red fibers were transformed into white fibers, and at a market weight of about 410 kg, the proportions of the three fiber-types were similar in all treatments. Muscle fiber diameters were also similar at constant weight. Thus, catch-up growth was associated with the reversal of feed restriction effects, in terms of both muscle fiber-type proportions and fiber diameter.

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