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# Effects of Growth Pattern on Muscle Growth, Nuclei Number, Protein Accretion, and Body Composition in Heifers

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# EFFECTS OF GROWTH PATTERN ON MUSCLE GROWTH, NUCLEI NUMBER, PROTEIN ACCRETION, AND BODY COMPOSITION IN HEIFERS

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## Summary

The effects of compensatory growth on accretion of muscle mass, protein mass, and nuclei number of the supraspinatus and semitendinosus muscles were evaluated using seven serial slaughter groups of Angus x Limousin heifer calves (n = 28, BW 270 ± 9.5 kg). Fractional growth rates of carcass protein and fat were also evaluated. To achieve compensatory growth, energy intake was restricted for 88 days (Phase 1) followed by ad libitum feeding of a high energy diet (Phase 2) [LH]. Controls were allowed continuous ad libitum access to the high energy diet (HH). Muscle weights, body composition samples, and muscle biopsies were collected at various weight (465 vs 500 kg) or age (88 vs 186 days) constants. Phase 1 energy restriction limited body weight, carcass weight, carcass protein mass, and carcass fat mass ($P<.05$). This was the result of the limited tissue fractional growth rates. The fractional growth rate of protein for heifers exhibiting compensatory growth was not increased but was maintained until maximum carcass protein mass was attained. Maximum carcass protein mass was attained by a weight of 465 kg. Any further increase in carcass weight was primarily attributed to an increase of carcass fat mass regardless of previous management. Energy restriction limited muscle, protein, and nuclei accretion rates. Heifers exhibiting compensatory growth sustained a linear growth potential until maximum muscle mass occurred at an end point similar to cattle not exhibiting compensatory growth. Muscle nuclei maintained a constant relationship to muscle mass independent of nutritional treatment, muscle type (supraspinatus vs semitendinosus), or days on feed. These data indicate compensatory growth alters the growth curve without affecting the mechanisms of growth.

Key Words: Beef, Compensatory Growth, Muscle

## Introduction

Beef cattle demonstrate improvements in production and biological efficiencies during compensatory growth. The mechanisms involved in this response are not clearly understood. Muscle growth can occur through either hyperplasia (increased cell numbers) or hypertrophy (increased cell size), and differences in this growth mechanism may play a role in the efficiency of compensatory growth. Based upon the concept of the DNA unit where a given amount of DNA has physiological control over a finite amount of cell cytoplasm, hyperplasia occurs with concomitant DNA accretion. The skeletal muscle cell is multinucleated and incapable of cell division. The ultimate size of the muscle would therefore be determined by the number of nuclei. Postnatal accretion of DNA that has been observed has been attributed to the differentiation of muscle satellite cells. Thus, DNA accretion or satellite cell recruitment has been suggested as a prerequisite for muscle growth. It has been demonstrated that muscle

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growth in steers over 300 kg BW was hypertrophic in nature, whereas others have attributed a major portion of muscle growth at heavier BW to hyperplasia. The reason for these contrasting conclusions is undoubtedly important as we elucidate the mechanisms involved in regulation of skeletal muscle growth. In the present study, the effects of compensatory growth, age, and body weight (BW) on body composition, DNA, and muscle accretion were evaluated.

## Materials and Methods

Fifty-eight Limousin x Angus heifer calves were vaccinated for IBR, BVD, $PI_3$, BRSV, 7-way clostridia, and Haemophilus within 24 hours of feedlot arrival. Ivermectin[4] was used for parasite control. Anabolic implants were not utilized. Twenty-eight heifers (BW = 270 $\pm$ 9.5 kg) were selected for uniformity of BW and type from this group of 58 calves for a serial slaughter experiment. These heifers were allotted to seven slaughter groups of four for comparison of body composition at various time and BW constants (Figure 1). The remaining 30 heifers were allotted to pens of five for comparisons of feedlot performance (previously reported data[5]).

Initial BW was the average of the BW measured on each of the first 2 days of the experiment. One pen was slaughtered on day 0 for initial body composition and muscle characterization. The six remaining groups were allotted to diets (Table 1) either of low energy (LED) to impose growth restriction or high energy (HED) provided ad libitum to allow for maximal growth (Phase 1). Energy values of feedstuffs and animal requirements for gain were based on NRC (1984).

One pen from each treatment was slaughtered at the end of Phase 1 (day 88). During Phase 2, heifers fed LED were switched to HED (LH) to achieve compensatory growth, while HED heifers continued on HED (HH). The remaining pens were slaughtered when pen average BW approached 465 or 500 kg (Figure 1).

After decapitation and prior to hide removal, approximately 200-g tissue biopsies were taken from the supraspinatus and the semitendinosus muscles and immediately frozen in liquid N. Samples were pulverized in a Waring blender under liquid N and analyzed for protein and DNA concentrations. Bovine serum albumin and calf thymus DNA from Sigma[6] were used as standards.

Supraspinatus and semitendinosus muscles were dissected and weighed from the opposite side of the hot carcass from where the biopsies were obtained. Predicted muscle mass of protein (g) and nuclei number (6.2 pg DNA) were calculated using the Proc Reg procedure of SAS. Comparisons of two regression lines were performed. Data of regression equations not significantly different ($P>.10$) were pooled.

The chemical analysis of the 9-10-11th rib soft tissue was used to estimate carcass soft tissue composition. Tissue fractional growth rates were calculated using the equation $(M_1 - M_O/T) / [(M_1 + M_O)/2]$, where $M_O$ = initial tissue measure, $M_1$ = final tissue measure, and T = time, day. Carcass composition data were tested by procedures appropriate for a completely random design with carcass representing the experiment unit. Data were analyzed on a BW constant (465 vs 500 kg) or a time constant (88 vs 186 days) basis. Analysis of variance was accomplished using the GLM procedure and CONTRAST option of SAS.

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[4]MSD AGVET, Division of Merck and Co., Inc. Rahway, NJ.
[5]S.D. Agr. Exp. Sta. Beef Report CATTLE 91-12:48-51.
[6]Sigma, St. Louis, MO.

Table 1. Experimental diet compositions[a]

| Ingredient | Diet | |
|---|---|---|
| | Low energy[b] | High energy[c] |
| Hay | - | 10.00 |
| Wheat straw | 15.00 | - |
| Corn silage | 74.94 | - |
| Whole shelled corn | - | 81.61 |
| Soybean meal, 44% | 9.21 | 4.60 |
| Molasses | - | 2.25 |
| Trace mineralized salt | .30 | .30 |
| Calcium carbonate | .55 | 1.01 |
| Potassium chloride | - | .23 |
| Nutrient composition | | |
| Crude protein, % | 11.04 | 11.77 |
| Calcium, % | .439 | .505 |
| Phosphorus | .235 | .290 |
| Potassium, % | 1.146 | .803 |
| $NE_m$, Mcal/kg | 1.53 | 2.06 |
| $NE_g$, Mcal/kg | .85 | 1.36 |

[a]Percentage of dry matter unless otherwise stated.

[b]Provides 33 mg/kg lasalocid day 1 to 36, 27.6 mg/kg of monensin day 37 to 88 and 2205 IU/kg supplemental vitamin A.

[c]Provides 33 mg/kg lasalocid and 2205 IU/kg supplemental vitamin A.

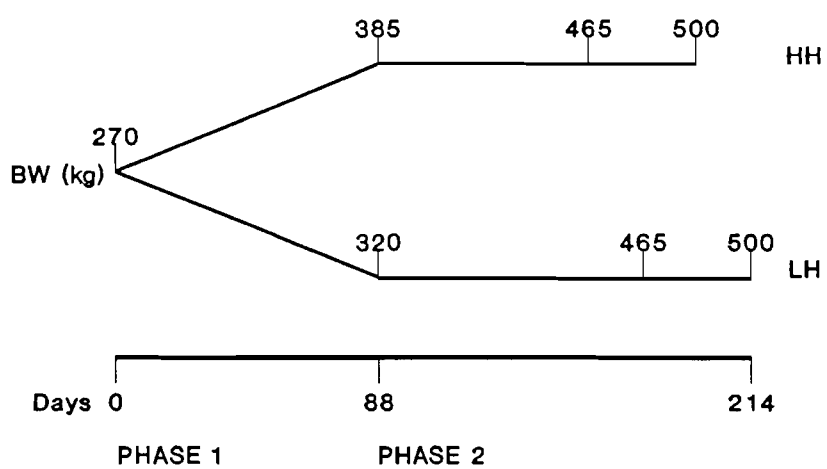


Figure 1. Serial slaughter points (n = 4).

## Results and Discussion

At the end of Phase 1 (day 88), both body weight (BW) and carcass weight (CW) were lowerfor the energy restricted heifers ($P<.05$, Table 2). This also resulted in a lower carcass protein mass (CPM) and carcass fat mass (CFM) [$P<.05$, Table 2]. During Phase 2 (day 186), CW was lower for the LH than HH heifers ($P<.05$, Table 2). After 186 days on feed, CPM was similar ($P>.05$, Table 2), whereas CFM was lower for LH than HH ($P<.05$, Table 2). The difference in CW is primarily attributable to the differences in CFM.

Contrasts performed at constant BW of 465 or 500 kg (Table 3) resulted in CW, CPM, and CFM being similar within BW group. Contrasts between BW groups (465 vs 500 kg) demonstrated that CW was greater at 500 kg BW ($P>.05$, Table 3). This increase in CW did not result from an increase in CPM, which was similar ($P>.10$, Table 3). Carcass fat mass increased from 465 to 500 kg BW ($P<.05$, Table 3). As a result, differences in CW can probably be attributed to increases in CFM.

To determine differences in the rate of tissue accretion, the fractional growth rate (FGR) of protein ($FGR_P$), and fat ($FGR_F$) were calculated. At the end of Phase 1, the $FGR_P$ and $FGR_F$ were lower for the energy restricted heifers. During realimentation, the opposite occurred. The $FGR_P$ and the $FGR_F$ were higher for LH than HH ($P<.05$, Table 4). Under a normal sigmoidal curve, the $FGR_P$ gradually decreases as the animal reaches maturity. This occurred with the HH heifers as the $FGR_P$ and $FGR_F$ decreased. During this same period, the $FGR_P$ for the LH heifers was maintained, while the $FGR_F$ increased. Compensatory growth has been characterized as having a more efficient $FGR_P$. This was not apparent in this study. Either an increase in the $FGR_P$ did not occur or had already occurred prior to the measurement made in this study. Increases in visceral mass which were not measured in this study may also account for an increase in $FGR_P$.

Table 2. Carcass tissue mass contrasts on an age constant basis

| | Phase 1 | | Phase 2 | | |
|---|---|---|---|---|---|
| Item | LH | HH | LH | HH | SEM |
| Body weight, kg[a] | 319 | 386 | 472 | 499 | |
| Carcass weight, kg[a] | 197 | 243 | 292 | 311 | |
| Protein, kg[a] | 31 | 36 | 43 | 41 | .6 |
| Fat, kg[ab] | 34 | 55 | 75 | 97 | 1.9 |

[a]Growth pattern differs within Phase 1 ($P<.05$).
[b]Growth pattern differs within Phase 2 ($P<.05$).

Table 3. Carcass tissue mass contrasts on a weight constant basis

| | 465 kg BW | | 500 kg BW | | |
|---|---|---|---|---|---|
| Item | LH | HH | LH | HH | SEM |
| Carcass weight, kg | 292 | 297 | 311 | 319 | |
| Protein, kg | 43 | 41 | 43 | 41 | .7 |
| Fat, kg[a] | 75 | 85 | 99 | 97 | 2.3 |

[a]Growth pattern differs at BW endpoint ($P<.05$).

Table 4. Fractional growth rate contrasts on an age constant basis

| | Phase 1 | | Phase 2 | | |
|---|---|---|---|---|---|
| Item | LH | HH | LH | HH | SEM |
| Protein, %/day[a] | .34 | .51 | .34 | .11 | .015 |
| Fat, %/day[a] | .43 | .92 | .81 | .52 | .023 |

[a]Growth pattern differs within phase (P<.05).

It appears that energy restriction limits protein accretion while compensatory growth maintains a linear growth potential until maximal protein mass similar to contemporaries is attained. Any further growth is comprised mainly of fat deposition (Figure 2). For these heifers, maximal protein growth had occurred by 465 kg BW.

Regression equations describing growth of tissue components over time in days for LH and HH treatments during Phase 1 are presented in Table 5. The accretion rates of the supraspinatus (SS) and semitendinosus (ST) muscle mass, nuclei number (NN), and protein mass (PROT) were greater when feed was provided ad libitum in Phase 1 (P<.10). When Phases 1 and 2 were pooled (Table 6), heifers on the LH treatment maintained linear accretion rates of muscle mass (Figure 3), PROT (Figure 4), and NN (Figure 5) for both SS and ST muscles (P<.10), while HH responses were quadratic (P<.10), reflecting a leveling off of growth in this treatment.

Protein accretion vs NN (Figure 6) was linear (P<.01) for ST but quadratic (P<.01) for SS, indicating a lag in hypertrophic PROT accretion for SS at heavier BW. This suggests that different muscles may exhibit differing rates of hyperplastic or hypertrophic growth. Increases of NN per unit of muscle mass maintained a linear relationship (P<.05) and were not affected (P>.10) by muscle (SS vs ST) or treatment (LH vs HH) [Figure 7]. This is consistent with the hypothesis that DNA accretion is a prerequisite for muscle growth and could ultimately determine muscle mass.

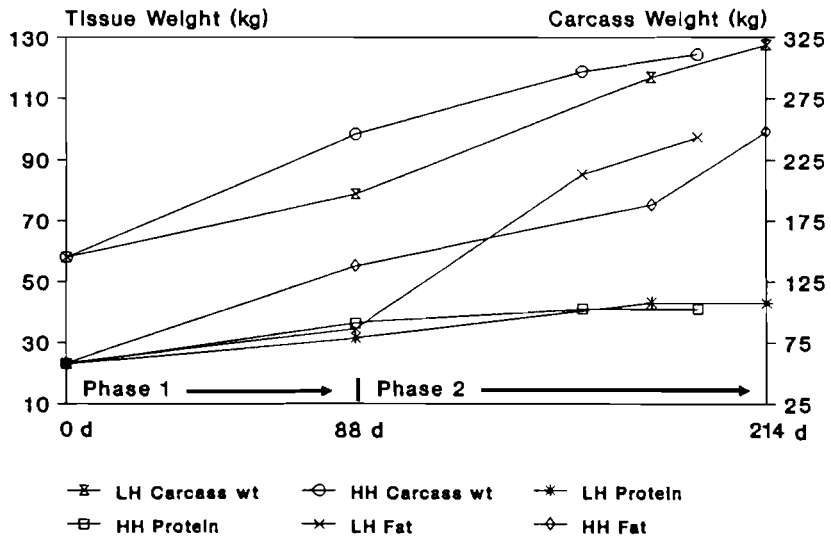


Figure 2. Carcass tissue accretion rates.

Table 5. Effect of time on tissue components (Phase 1)[a]

| Y | Treatment | Intercept | $b_1$ | $r^2$ |
|---|---|---|---|---|
| Supraspinatus (kg) | LH | .65 | .0024 | .988 |
| | HH | .65 | .0038 | .926 |
| Semitendinosus (kg) | LH | 1.22 | .0042 | .670 |
| | HH | 1.22 | .0082 | .865 |
| Supraspinatus nuclei number[b] | LH | 52.09 | .1485 | .829 |
| | HH | 52.09 | .2422 | .852 |
| Semitendinosus nuclei number[b] | LH | 86.27 | .3418 | .614 |
| | HH | 86.27 | .7142 | .913 |
| Supraspinatus protein (g) | LH | 102.14 | .3614 | .933 |
| | HH | 102.14 | .5543 | .956 |
| Semitendinosus protein (g) | LH | 204.22 | .6231 | .873 |
| | HH | 205.22 | 1.4108 | .930 |

[a]Where Y = dependent variable as kg, g, or nuclei number and X = days since inception of the experiment.
[b]Nuclei number x $10^6$.

Table 6. Effect of time on tissue components (Phases 1 and 2)[a]

| Y | Treatment | Intercept | $b_1$ | $b^2$ | $r^2$ |
|---|---|---|---|---|---|
| Supraspinatus (kg) | LH | .661 | .0021 | | .845 |
| | HH | .654 | .0042 | -.000012 | .860 |
| Semitendinosus (kg) | LH | 1.255 | .0036 | | .743 |
| | HH | 1.217 | .0129 | -.000052 | .713 |
| Supraspinatus nuclei number[b] | LH | 51.929 | .1785 | | .779 |
| | HH | 52.162 | .2920 | -.000616 | .872 |
| Semitendinosus nuclei number[b] | LH | 86.461 | .3157 | | .831 |
| | HH | 86.016 | 1.1187 | -.004430 | .896 |
| Supraspinatus protein (g) | LH | 10.684 | .3973 | | .969 |
| | HH | 102.201 | .7698 | -.002490 | .937 |
| Semitendinosus protein (g) | LH | 201.946 | .6976 | | .968 |
| | HH | 205.049 | 2.1212 | -.008626 | .902 |

[a]Where Y = dependent variable as kg, g, or nuclei number and X = days since inception of the experiment.
[b]Nuclei number x $10^6$.

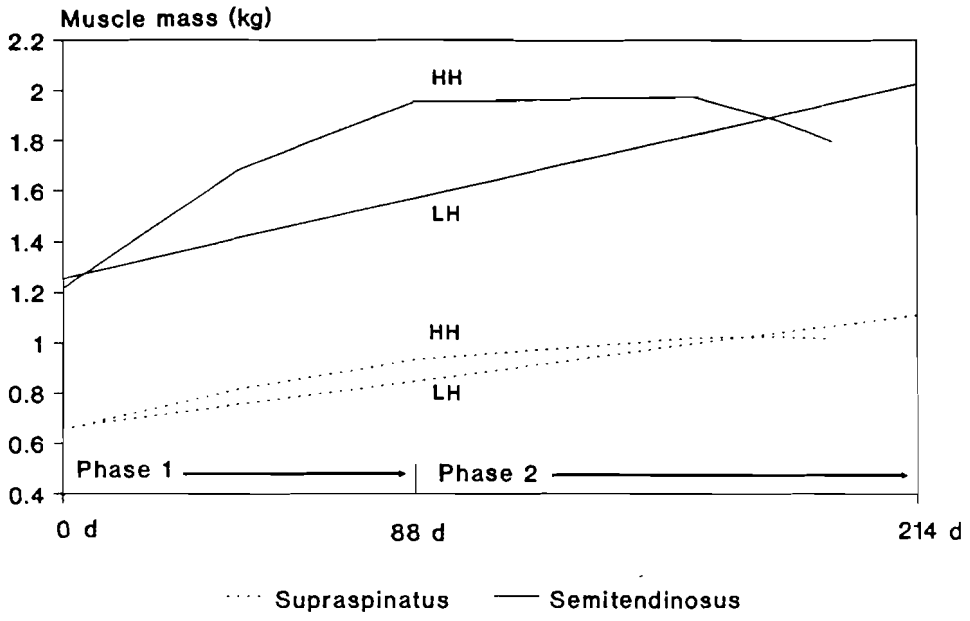


Figure 3. Muscle mass accretion rate.

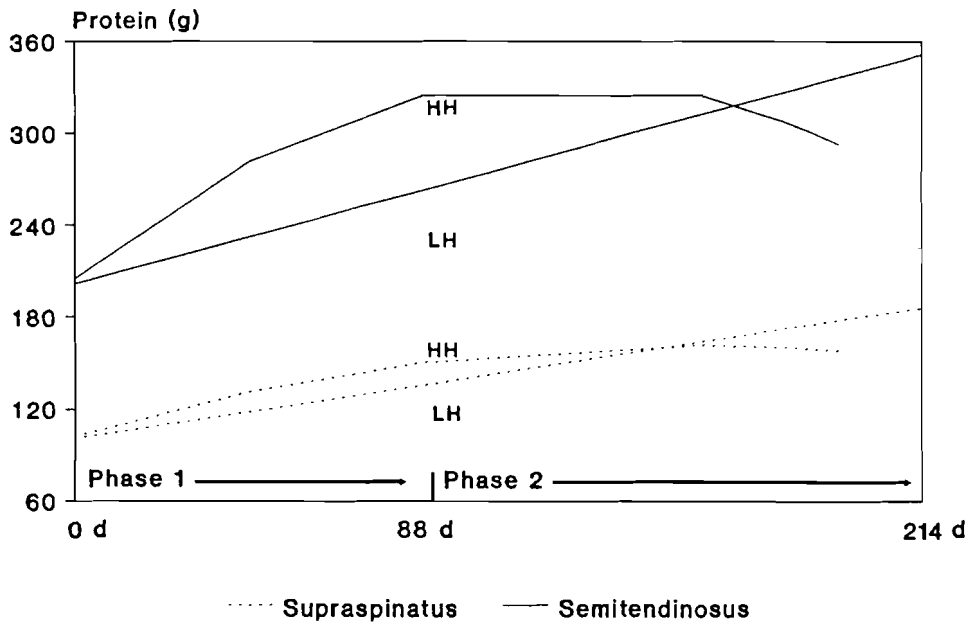


Figure 4. Muscle protein mass accretion rate.

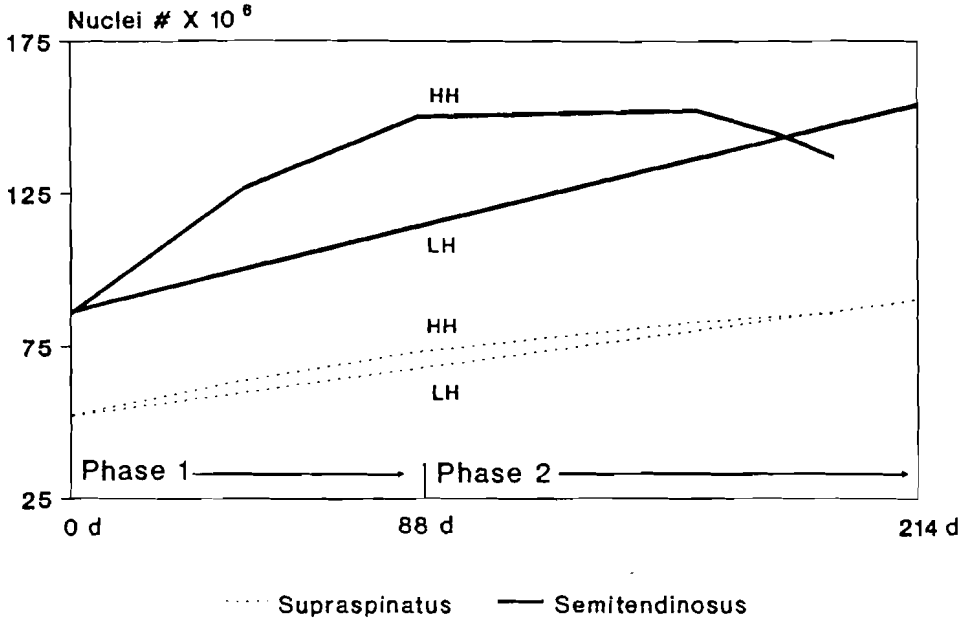


Figure 5. Muscle nuclei accretion rate.

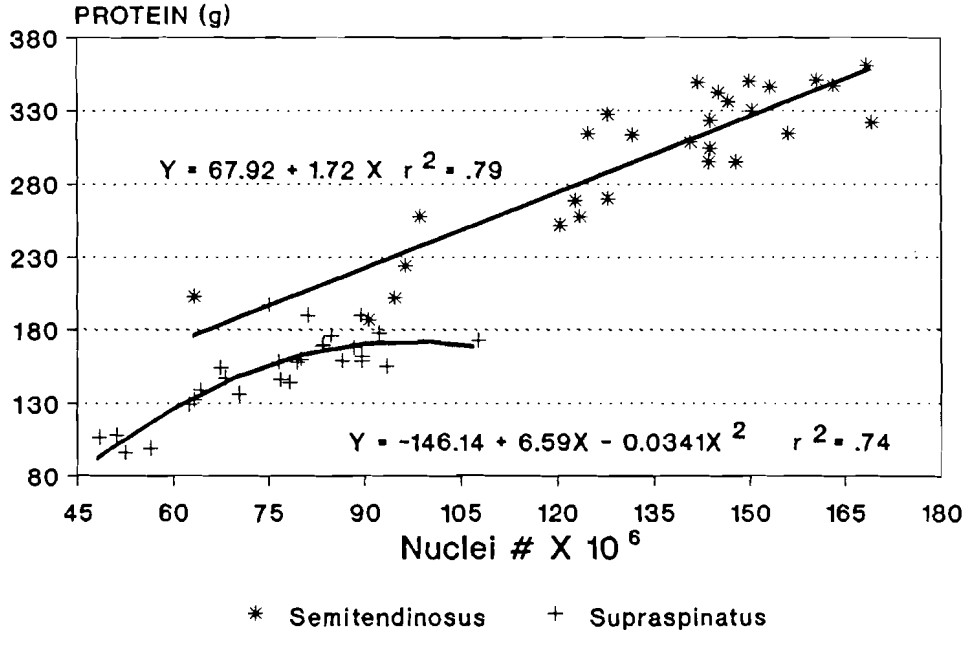


Figure 6. Relationship of nuclei number to muscle protein mass.

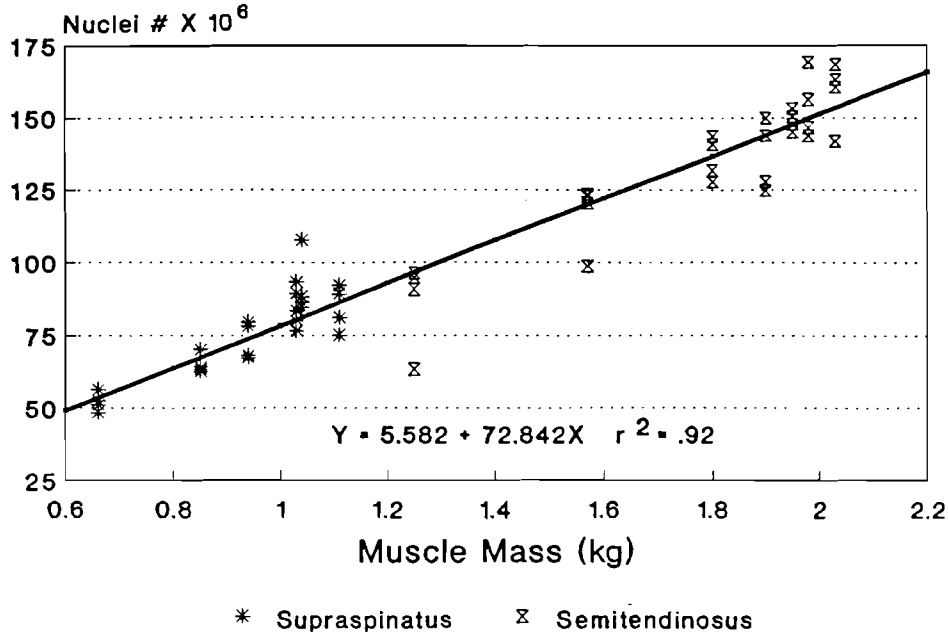


**Figure 7.** Relationship of nuclei number to muscle mass (coefficients differ from zero, $P<.01$).