1:8

Edurance exercise is associated with meat tenderness in sheep.

Aalhus, J.L. and M.A. Price
Department of Animal Science, University of Alberta, Edmonton, Alberta, Canada T6G 2P5.

Introduction

Early experiments by Mitchell and Hamilton (1933) and Bull and Rusk (1942) indicated that endurance exercise caused an increase in meat tenderness. Several experiments since then have contradicted these results (Hawrysh et al, 1974, Mandigo et al, 1971) whereas others (Spaeth et al, 1967, Crystall et al, 1982) have supported them. The following experiment was conducted to study the effect of endurance exercise on muscle tenderness and to examine some of the associated factors.

Materials and Methods

Twenty two purebred Suffolk ram lambs (27-36 kg) were paired by weight and randomly assigned to one of three slaughter groups (slaughtered after 6, 12 or 18 weeks of exercise) and one four starting dates within each slaughter group. Within each pair, animals were randomly assigned to either exercise or control treatments; they were fed a 16% protein ration ad lib. Exercised animals worked-out on a treadmill for five days per week, maintaining a workload of 75% of maximum oxygen consumption (VO_2 max) tested every three weeks. Time on the treadmill was increased continuously to a maximum of 60, 75 and 90 minutes per day for the 6, 12 and 18 week slaughter groups, respectively. Three days prior to slaughter the rams were given a final VO_2 max test and were not exercised again. Two days later their feed was removed; they were slaughtered and dressed in the normal commercial manner and the carcasses were chilled for 48 hours. After 48 hours the semimembranosus (SM) and the vastus lateralis (VL) were removed from the left side of the carcass, trimmed of fat and connective tissue and stored at -18°C for later Warner-Bratzler Shear (WBS) and Kramer Press-Ground (KP-G) analysis. The SM and VL were removed from the right side of the carcass and were subsampled for determination of dry matter, nitrogen, myofibrillar fragmentation, total collagen and soluble collagen.

1. Warner-Bratzler Shear (WBS) and Kramer Press-Group (KP-G)

After 9 to 13 weeks of storage at -18°C the left SM and VL were thawed in a 3°C cooler (40 and 24 hours, respectively). Thaw drip loss was recorded and a portion was removed from the distal end of the muscles for the determination of ultimate pH. The muscles were then roasted in a 177°C oven to an internal temperature of 70°C, and cooled to an internal temperature of 50°C before drip and volatile cooking losses were recorded. The muscles were then stored overnight in a 3°C cooler.

The following day the external surfaces of the cooked muscles were trimmed. The SM muscles were sliced into three 1.27 cm slices parallel to the grain. The slice which corresponded to the deep surface of the SM in situ was cut into core samples 2.54 cm x 1.27 cm x 1.27 cm. A similar procedure was followed for the VL muscles but all the slices were used for core samples. Core samples were sheared on an Instron 4201 equipped with a Warner-Bratzler Shear cell. Sheared core samples plus the remaining slices were ground and three 20 g subsamples were put through an OTMS equipped with a Kramer Shear cell.

2. Ultimate pH

Twenty g of muscle from the SM and 10 g of muscle from the VL were added to deionized, distilled water. Samples were homogenized in a blender and filtered. The pH was recorded on duplicate samples of the filtrate using a Fisher Accumet 610A pH meter.

3. Dry matter and nitrogen determination

Several cores (1.27 cm diameter) were removed from the frozen right side SM and VL muscles and were scissor-minced, removing any visible fat or connective tissue. Duplicate samples of approximately 2 g and duplicate samples of approximately 1 g were prepared for dry matter and kjeldahl nitrogen determinations, respectively, as outlined by the A.O.A.C. (1980).

4. Myofibrillar fragmentation index (MFI)

MFI's were determined using the Culler et al. (1978) modification of Davey and Gilbert's (1969) procedure. Samples were homogenized at 20,000 r.p.m. in a Virtis '45' tissue homogenizer. After centrifugation the sediment was passed through two non standard nylon strainers having a respective pore size of 1.69 mm^2 and .46 mm^2. Protein concentrations were determined using the method of Lowry et al. (1951).

5. Total and soluble collagen

Sample preparation combined the procedures of Culler et al. (1978) and Stegemann and Stalder (1967). Samples were autoclaved, decolorized and filtered as described by Culler et al. (1978). The HCl was evaporated rather than neutralized and the diluted samples were purified on cation exchange columns as recommended by Stegeman and Stalder (1967). Samples were analysed for hydroxyproline using the procedure of Stegeman and Stalder (1967). Absorbance at 550 nm was read on a Brinkmann PC800 Colorimeter. Hydroxyproline values were converted to total collagen using a factor of 7.25 (Goll et al., 1963) and to soluble collagen using a factor of 7.52 (Cross et al., 1973).

Results

Exercised animals had significantly lower WBS values in both the SM and VL muscles than controls (Figure 1). Exercised animals also had significantly lower KP-G values than the controls in the VL muscle (Figure 2). There was no consistent trend towards increased toughness over the 18 week time period as indicated by the WBS and KP-G values (Figures 3 and 4). KP-G values for the VL reflect an increase in toughness over the 18 week period ($P=.05$, Figure 4). In all cases the VL muscle had lower KP-G and WBS values than the SM muscle (Figures 1 to 4).

There were no significant differences in MFI between exercised and control animals in either muscle (Table 1), though the MFI's approached significance among slaughter groups in the SM.

There was significantly less total collagen per g of frozen tissue in the VL of the exercised animals (Table 2). This did not occur in the SM. Total collagen tended to increase with age, particularly in the VL ($P=.08$, Table 2). The amount of soluble collagen (expressed as a percent of total collagen) decreased significantly with age in both the SM and VL. Exercised animals also tended to have less soluble collagen than controls.

There were no significant differences among slaughter groups or between treatments in the ultimate pH. In all cases though the ultimate pH in the VL was higher than the ultimate pH in the SM (Tables 1 and 2).

Percent dry matter and percent nitrogen did not change with age or treatment. The percent dry matter and percent nitrogen in the SM were consistently higher than in the VL.
In both muscles the fresh weight increased with age from 6 to 18 weeks (Table 3). When adjusted for initial differences in fresh weight the thawed weights were significantly different (P=.04) between exercised and control animals for the SM muscle. Exercised animals also tended (P=.06) to have less thaw drip than controls in the SM. Thawed weight and percent thaw drip were not significantly different between exercised and control or among slaughter groups for the VL.
The trimmed raw weight of the SM and VL increased with age. Exercised animals tended to have heavier trimmed raw weights than the controls (Table 4). Cooked weights (adjusted to initial differences in the trimmed raw weight) were significantly different among slaughter groups (P=.01) and between treatments (P=.07) in the SM. Cooked weights did not differ among slaughter groups and or between treatments in the VL.
Cooking drip (expressed as a percent of cooked weight) was significantly affected by slaughter group and exercise in the SM. There was no difference among slaughter groups in percent cook drip in the VL. Exercised animals tended to have less percent cooking drip than controls in the VL. Percent volatile loss was only significantly different among slaughter groups for the SM muscle. Muscles from animals in the 18 week slaughter group had significantly less volatile cooking losses than those from 6 and 12 week slaughter groups.

Discussion

Several researchers have looked at the effect of exercise immediately before slaughter. Briskey et al. (1959) reported that severe exercise immediately before slaughter resulted in hogs that had muscles that were dark in colour, high in pH value and dry in appearance. Chrystall et al. (1982) reported similar effects on pH as well as an increase in tenderness of several leg muscles, including the SM in lambs subjected to exercise pre-slaughter. Information on the effect of chronic exercise on muscle tenderness has been controversial. This experiment found that the SM and VL from exercised animals were more tender than those of their control counterparts. In all but one case (WBS measurements on the SM) the muscles from exercised sheep had significantly lower WBS and KP-G values than those of the controls. Mitchell and Hamilton (1933) and Bull and Rusk (1942) working with cattle found that the tenderness of rib roasts, round steaks and ribs increased as a result of exercise. Similarly Spaeth et al. (1967) reported an increase in the tenderness of leg muscles from exercised lambs, however, these animals were also treated with diethystilbestrol, which may have affected this result. Mandigo et al. (1971) and Hawrysh et al. (1974) showed no effect of exercise on tenderness of pork. Hawrysh et al. used the longissimus muscle which is not heavily involved in treadmill walking for their determination of tenderness. Although not stated, it is assumed that Mandigo et al. also used the longissimus muscle as well. In addition, none of these researchers adjusted the intensity of the exercise to compensate for differences in the inherent athletic ability among individual animals.
Thus, exercise is associated with increased tenderness. What causes this effect? It is possible to break down tenderness into various components (Cover et al., 1962) which include a connective tissue component and a myofibrillar component. Contributions from these components can be distinguished by analysing the pattern of the WBS deformation curve. The peak force, the one reported in this paper, is due in part to both of these components (Bouton et al., 1975). Many things can influence the contribution of these components to overall mechanical tenderness including the amount and solubility of the collagen, the rate of pH and temperature fall, ultimate pH and temperature fall, ultimate pH and its effects on water holding capacity and the degree of fiber fragmentation post slaughter.
The amount of total collagen has a large effect on the tenderness of muscle as measured by mechanical methods. Mitchell and Hamilton (1933) reported a lower percent collagen nitrogen in muscles from exercised cattle which agrees with the results for the VL in this experiment. Hawrysh et al. (1974) did not find a difference in the amount of hydroxyproline between exercised and control pigs in the biceps femoris and psoas major muscles. Also, Hiner et al. (1955) reported that muscles which were used more extensively by the animal had larger amounts of collagenous fibers, whereas those with less activity had smaller amounts.
The percent soluble collagen was lower in exercised sheep than controls. This agrees with Kruggel and Field (1974) who found that muscles which are highly involved in exercise (foreshank muscles) have less collagen cross-linkages than in less active muscles (longissimus).
In animals exercise stressed immediately before slaughter the increase in tenderness is associated with a higher ultimate pH which influences the physical state of the myofibrillar protein. Chronically exercised sheep do not differ from controls in their ultimate pH. Thus differences in tenderness must be attributed to some other factors. Bouton et al. (1972) attribute variations in thoughness not directly associated with pH to differences in the "stress-state" of the animal at slaughter resulting in variations in fiber contraction. Differences in fiber toughness in the 5.4 to 6.0 pH range are decided more by fiber contraction state than by the physical state of the muscle proteins. The difference in ultimate pH between the SM and VL muscles may be responsible for the difference in WBS and KP-G values between them (Bouton et al., 1982).
Changes in ultimate pH are also associated with changes in the water holding capacity of muscle due to changes in the physical state of the muscle (McClain and Mullins, 1969; Bouton et al., 1972). Since there were no differences in ultimate pH, differences in thaw drip and cooking drip between exercised and control animals cannot be attributed to ultimate pH. Removing the effect of differences in muscle weight does not remove these differences. It seems probable that there is a 'shape' factor involved which changes the surface to volume ratio of the muscle and thus changes the rate of drip losses in thawing and cooking. Another possibility is that chronic endurance exercise induces changes in the myofibrillar proteins which changes their water holding capacity.
The MFI is used as an indication of the amount of fractionation of the myofibrillar protein at the Z-line during post mortem storage. Meat tenderness has been associated with this phenomenon (Olson et al., 1976). The MFI values obtained for the rams used in this experiment were much lower than other values reported in the literature for beef (Culler et al., 1978; Olson et al., 1976). Part of this difference may be due to the use of strainers with a smaller pore size than those reported by Culler et al., (1978). The remainder of the difference must be related to a species difference in myofibril cross sectional area or total collagen. There were no differences in the MFI between exercised and control animals which would have indicated a difference in the post mortem breakdown of the myofibrillar proteins at the Z-line.
There was no consitent trend towards tougher meat with increased age in this experiment. Jeremiah (1978) reviewed the effect of chronilogical age and physiological maturity on meat tenderness and found the literature to be inconclusive. Bouton et al. (1978) suggested that these results may be partly due to a limited age range of animals and to variations in the prerigor shortening which may occur in Achilles tendon hung animals.

Based on the results of this study long term endurance exercise can be said to increase muscle tenderness but the mechanism by which this occurs is still unclear. Collagen content and solubility seem to be influenced by exercise but not in a consistent pattern. Myofibrillar proteins may also be affected as indicated by the differences in water holding capacity in thawing and cooking. However there was no difference in the ultimate pH, nitrogen content or myofibrillar fragmentation to support this.

Acknowledgements

We would like to thank the Natural Sciences and Engineering Research Council of Canada for financial support. We would also like to thank Mr. Jim Townshend for his excellent assistance in managing the animals and maintaining the treadmill.

References

Association of Official Analytical Chemists. (A. O. A. C.). 1980. Official methods of analysis. 13th ed. Washington, D.C.

Bouton, P. E., Harris, P. V. and Shorthose, W. R. 1972. The effects of ultimate pH on ovine muscle: mechanical properties. J. Food Sci. 37:356-360.

Bouton, P. E., Harris, P. V. and Shorthose, W. R. 1975. Changes in shear parameters of meat associated with structural changes produced by aging, cooking and myofibrillar contraction. J. Food Sci. 40:1122-1126.

Bouton, P. E., Harris, P. V., Ratcliff, D. and Roberts, D. W. 1978. Shear force measurements on cooked meat from sheep of various ages. J. Food Sci. 43:1038-1039.

Bouton, P. E., Harris, P. V., MacFarlane, J. J. and Shorthose, W. R. 1982. Influence of pH on the Warner-Bratzler shear properties of mutton. Meat Science. 6:27-36.

Briskey, E. J., Bray, R. W., Hoekstra, W. G., Grummer, R. H. and Phillips, P. H. 1959. The effect of various levels of exercise on altering the chemical and physical characteristics of certain pork ham muscles. J. Anim. Sci. 18:153-157.

Bull, S. and Rusk, H. P. 1942. Effect of exercise on the quality of beef. Illinois Agric. Exp. Sta. Bull. 488.pp. 107-120.

Chrystall, B. B., Devine, C. E., Snodgrass, M. and Ellery, S. 1982. Tenderness of exercise-stressed lambs. N. Z. J. Agric. Res. 25:331-336.

Cover, S., Ritchey, S. J. and Hostetler, R. L. 1962. Tenderness of beef. 1. The connective-tissue component of tenderness. J. Food Sci. 27:469-475.

Cross, H. R., Carpenter, Z. L. and Smith, G. C. 1973. Effects of intramuscular collagen and elastin upon bovine muscle tenderness. J. Food Sci. 38:998-1003.

Culler, R. D., Parrish Jr., F. C., Smith, G. C. and Cross, H. R. 1978. Relationship of myofibril fragmentation index to certain chemical, physical and sensory characteristics of bovine longissimus muscle. J. Food Sci. 43:1177-1180.

Davey, C. L. and Gilbert, K. V. 1969. Studies in meat tenderness. 7. Changes in the fine structure of meat during aging. J. Food Sci. 34:69-74.

Goll, D. E., Bray, R. W. and Hoekstra, W. G. 1963. Age-associated changes in bovine muscle composition. The isolation and properties of a collagenous residue from bovine muscle. J. Food Sci. 28:503-509.

Hawrysh, Z. J., Murray, D. M. and Bowland, J. P. 1974. Effects of exercise on palatability and cooking characteristics of pork. Can. J. Anim. Sci. 54:191-195.

Hiner, R. L., Anderson, E. E. and Fellers, C. R. 1955. Amount and character of connective tissue as it relates to tenderness in beef muscle. Food Tech. 9:80-86.

Jeremiah, L. E. 1978. A review of factors affecting meat quality. Agriculture Canada Technical Bulletin 1. pp. 8-9.

Kruggel, W. G. and Field, R. A. 1974. Cross linking of collagen in active and quiescent muscle. J. Anim. Sci. 38:1326. (Abstr.).

Lowry, O. H., Rosebrough, N. J., Farr, A. L. and Randall, R. J. 1951. Protein measurement with the folin phenol reagent. J. Biol. Chem. 193:265-275.

Mandigo, R. W., Peo Jr., E. R.,Kumm, R. C. and Moser, B. D. 1971. Influence of exercise on pork carcass composition. J. Anim. Sci. 33:220. (Abstr.).

McClain, P. E. and Mullins, A. M. 1969. Relationship of water binding and pH to tenderness of bovine muscles. J. Anim. Sci. 29:268-271.

Mitchell, H. H., and Hamilton, J. S. 1933. The effect of long-continued muscular exercise upon the chemical composition of the muscles and other tissues of beef cattle. J. Agric. Res. 46:917-941.

Olson, D. G., Parrish Jr., F. C. and Stromer, M. H. 1976. Myofibril fragmentation and shear resistance of three bovine muscles during post mortem storage. J. Food Sci. 41:1036-1041.

Spaeth, C. W., Carpenter, Z. L., King, G. T. and Shelton, M. 1967. Effects of forced exercise on lamb carcass merit. J. Anim. Sci. 26:901-902. (Abstr.).

Stegemann, H. and Stalder, K. 1967. Determination of hydroxyproline. Clinica Chimica Acta. 18:267-273.

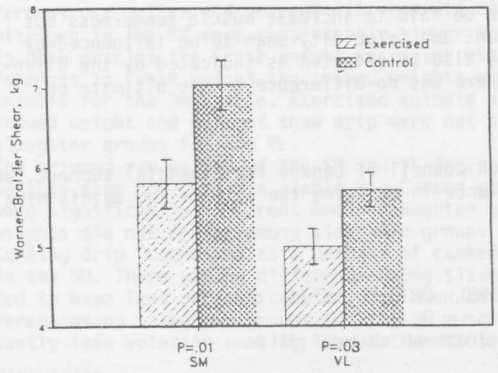

Figure 1. Exercised and control Warner-Bratzler Shear measurements for the SM and VL muscles.

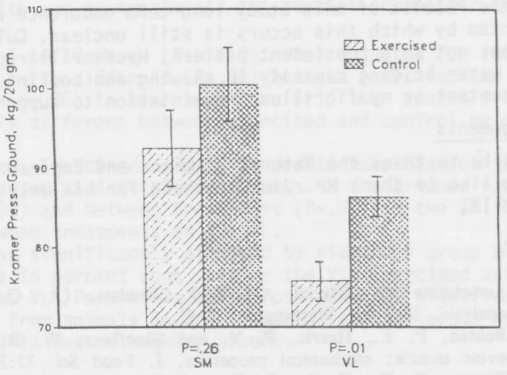

Figure 2. Exercised and control Kramer Press-Ground measurements for the SM and VL muscles.

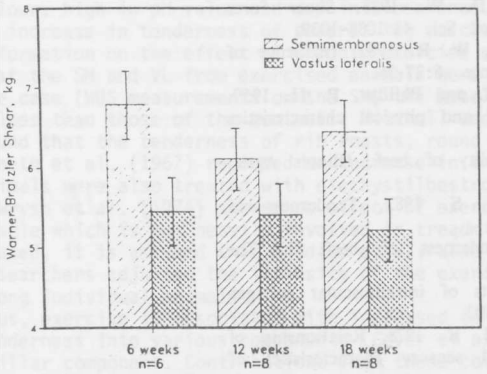

Figure 3. Change in Warner-Bratzler Shear measurements with time (SM P=0.51; VL P=0.78).

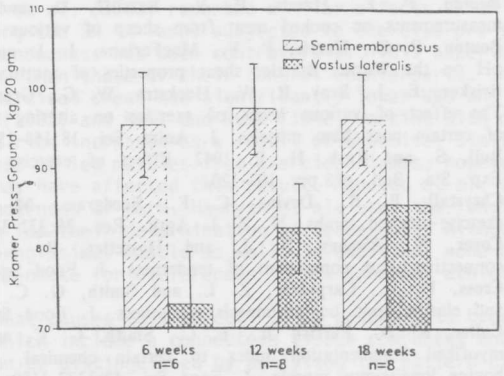

Figure 4. Change in Kramer Press-Ground measurements with time (SM P=0.97; VL P=0.05).

Table 1. Certain chemical properties of the semimembranosus muscle (frozen weight basis).

	Number of animals	MFI[c]	Total Collagen mg/g	Soluble Collagen %	Ultimate pH	Dry Matter %	Nitrogen %
Overall Mean	22	14.96±0.94	6.18±0.25	3.96±0.14	5.55±0.02	26.17±0.18	3.22±0.03
Saughter Group							
6 weeks	6	11.15±1.80	6.07±0.47	4.73±0.26[a]	5.62±0.05	26.43±0.34	3.22±0.05
12 weeks	8	16.89±1.56	5.95±0.41	3.78±0.22[b]	5.49±0.04	25.70±0.29	3.19±0.04
18 weeks	8	15.90±1.56	6.49±0.41	3.56±0.22[b]	5.55±0.04	26.44±0.29	3.24±0.04
		P=.07	P=.63	P=.01	P=.14	P=.17	P=.63
Treatment							
Exercised	11	14.71±1.33	6.20±0.35	3.70±0.19	5.53±0.03	26.05±0.25	3.18±0.04
Control	11	15.22±1.33	6.15±0.35	4.22±0.19	5.56±0.03	26.29±0.25	3.25±0.04
		P=.79	P=.92	P=.07	P=.50	P=.50	P=.15

Table 2. Certain chemical properties of the vastus lateralis muscle (frozen weight basis).

	Number of Animals	MFI[c]	Total Collagen mg/g	Soluble Collagen %	Ultimate pH	Dry Matter %	Nitrogen %
Overall Mean	22	14.16±0.91	6.33±0.19	4.33±0.13	5.78±0.02	24.81±0.25	3.08±0.03
Slaughter Group							
6 weeks	6	13.03±1.74	6.14±0.36	5.05±0.26[a]	5.76±0.03	24.78±0.48	3.11±0.06
12 weeks	8	13.71±1.51	5.89±0.31	4.16±0.22[b]	5.80±0.03	24.56±0.42	3.09±0.05
18 weeks	8	15.45±1.51	6.93±0.31	3.95±0.22[b]	5.77±0.03	25.09±0.42	3.05±0.05
		P=.55	P=.08	P=.01	P=.52	P=.68	P=.67
Treatment							
Exercised	11	14.09±1.29	5.93±0.26	4.28±0.19	5.76±0.02	24.87±0.36	3.04±0.04
Control	11	14.23±1.29	6.74±0.26	4.37±0.19	5.80±0.02	24.75±0.36	3.12±0.04
		P=.94	P=.05	P=.75	P=.32	P=.82	P=.20

[a,b] Means and standard errors not followed by the same letter are significantly different at the 5% level of probability as determined by the Student-Neuman-Kuels multiple range test.
[c] Absorbance per 0.5mg myofibril protein x200.

Table 3. Thawing characteristics of the semimembranosus and vastus lateralis muscles from 22 ram-lambs.

	SEMIMEMBRANOSUS			VASTUS LATERALIS		
	Fresh weight (g)	Thawed weight (adjusted to mean of fresh wt.)	% Thaw drip	Fresh weight (g)	Thawed weight (adjusted to mean of fresh wt.)	% Thaw drip
Overall Mean (n=22)	613.27±11.96	583.82±2.09	5.30±0.36	299.95±7.69	295.74±0.46	1.60±0.17
Slaughter Group						
6 weeks (n=6)	556.50±22.91[a]	576.65±4.00	6.53±0.70	265.33±14.73[a]	295.48±0.87	1.79±0.33
12 weeks (n=8)	618.00±19.84[a,b]	584.51±3.47	5.26±0.60	309.50±12.76[b]	296.29±0.76	1.36±0.30
18 weeks (n=8)	651.13±19.84[b]	588.51±3.47	4.42±0.60	316.38±12.76[b]	295.39±0.76	1.70±0.30
	P=.02	P=.22	P=.21	P=.04	P=.67	P=.60
Treatment						
Exercised (n=11)	630.91±16.92	588.66±2.96	4.51±0.51	302.64±10.88	296.01±0.64	1.49±0.25
Control (n=11)	595.64±16.92	578.98±2.96	6.09±0.51	297.27±10.88	295.47±0.64	1.71±0.25
	P=.16	P=.04	P=.06	P=.73	P=.57	P=.55

Table 4. Cooking characteristics of the semimembranosus and vastus lateralis muscles from 22 ram-lambs.

	SEMIMEMBRANOSUS				VASTUS LATERALIS			
	Trimmed raw wt. (g)	Cooked wt. (g) (Adjusted to mean of trimmed raw wt.)	Cooking drip (%)	Volatile loss (%)	Trimmed raw wt. (g)	Cooked wt. (g) (Adjusted to mean of trimmed raw wt.)	Cooking drip (%)	Volatile loss (%)
Overall Mean (n=22)	551.24±11.34	416.88±3.06	3.45±0.15	20.99±0.49	273.34±7.65	201.11±1.25	5.94±0.18	20.26±0.42
Slaughter Group								
6 weeks (n=6)	494.30±21.71[a]	408.57±5.86[a]	3.58±0.29[a,b]	22.40±0.93[a]	242.12±14.65	200.53±2.40	6.37±0.35	20.01±0.81
12 weeks (n=8)	554.70±18.80[a,b]	406.59±5.08[a]	3.95±0.25[a]	22.24±0.81[a]	281.12±12.69	198.63±2.08	5.90±0.30	21.19±0.70
18 weeks (n=8)	590.49±18.80[b]	433.40±5.08[b]	2.86±0.25[b]	18.68±0.81[b]	288.96±12.69	204.03±2.08	5.65±0.30	19.52±0.70
	P=.01	P=.01	P=.03	P=.02	P=.07	P=.22	P=.43	P=.26
Treatment								
Exercised (n=11)	573.00±16.03	423.57±4.33	3.04±0.21	20.23±0.69	276.85±10.82	202.17±1.77	5.60±0.26	20.16±0.60
Control (n=11)	529.48±16.03	410.19±4.33	3.86±0.21	21.75±0.69	269.83±10.82	200.05±1.77	6.28±0.26	20.36±0.60
	P=.07	P=.07	P=.02	P=.17	P=.65	P=.41	P=.09	P=.82

[a,b] Means and standard errors not followed by the same letter are significantly different at the 5% level of probability as determined by the Student-Neuman-Kuels multiple range test.