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COMPARATIVE ANALYSIS OF MUSCLE FIBRE CHARACTERISTICS IN MAJOR CAPRINE MUSCLES

Jaehoon Baek[1*], Junyoung Park[1], Sumin Song[1], Huilin Cheng[1], Hyun-Jun Kim[2], Gap-Don Kim[1, 2, 3]

[1]Graduate School of International Agricultural Technology, Seoul National University, Pyeongchang 25354, Republic of Korea

[2]Institutes of Green Bio Science & Technology, Seoul National University, Pyeongchang 25354, Republic of Korea

[3]Research Institute of Agriculture & Life Science, Seoul National University, Seoul 08826, Republic of Korea

*Corresponding author email: gapdonkim@snu.ac.kr

I. INTRODUCTION

Muscle fibre characteristics play a central role in understanding both the transformation of skeletal muscle into meat and the physicochemical properties of meat. Specifically, muscle fibre type influences postmortem pH decline: glycolytic fibres are linked to a faster rate of glycolysis, and consequently lowering the ultimate pH compared to oxidative fibres [1]. Furthermore, identifying the similarities and differences in muscle fibre characteristics among skeletal muscles provides valuable insights for interpreting meat quality among different types of muscles (cut). Unlike cattle or pigs, studies on goat muscle fibre characteristics and their relationship with meat quality have not been sufficiently conducted. Therefore, this study was conducted to elucidate the similarities of muscle fibre characteristics across each type of caprine skeletal muscle with different architectures.

II. MATERIALS AND METHODS

Longissimus thoracis et lumborum (LTL), *semimembranosus* (SM), *semitendinosus* (ST), *psoas major* (PM), *diaphragm* (DP), *gracillis* (GA), *rectus abdominis* (RA), and *superficial digital flexor* (SDF) muscles were obtained from both sides of two goat carcasses (12 kg, male, 4 months old) after 24 h postmortem chilling at 4°C. Three different samples from each muscle were collected for histological analysis following Song et al. (2020) with some modification. For histological analysis, 1.0 × 1.0 × 1.0 cm muscle samples were trimmed and frozen in 2-methylbutane pre-cooled in liquid nitrogen. Transverse and longitudinal sections were cut using a cryostat microtome (CM1520; Leica Biosystems, Wetzlar, Germany). Each sections were stained with primary antibodies specific to myosin heavy chain (MHC) isoforms: BA-F8 (anti-MHC slow/I), SC-71 (anti-MHC IIA, and IIX), 6H1 (anti-MHC, slow/I, and IIX) (all from DSHB, Iowa City, IA, USA). For each primary antibody, IgG and IgM fluorescent dye conjugate secondary antibody (AlexaFluor 405, 488, and 594; Thermo Fisher Scientific, Waltham, MA, USA) was used. Images were captured using a fluorescence microscope (EVOS M5000; Thermo Fisher Scientific). Cross-sectional area (CSA, μm²), relative fibre area (RFA, %), fibre density (fibres/mm²), fibre length (μm), and muscle bundle size (μm²) were measured using Image-Pro Plus software (Media Cybernetics, Rockville, MD, USA). Muscle fibre characteristics among muscle types were analysed using one-way analysis of variance (ANOVA), followed by Duncan's multiple range test in SAS (version 9.4; SAS Institute, Cary, NC, USA), with significance set at $p < 0.05$. Hierarchical cluster analysis (HCA) was also performed to assess the similarity in muscle fibre profiles across muscles.

III. RESULTS AND DISCUSSION

LTL exhibited the largest bundle size among all muscles, whereas ST, GA, RA, and SDF showed significantly smaller values ($p<0.05$). Type IIX fibres were not observed in DP muscle. Only type IIA fibre length showed significant differences, with the greatest LTL length ($p < 0.05$). For CSA, RA exhibited the largest area among all fibre types ($p<0.05$), whereas GA had the lowest values ($p<0.05$). The CSA of type IIA fibres in the DP was significantly larger than that in all muscles except the RA ($p<0.05$). Regarding RFA, the highest values for type I and IIA fibres were found in the DP group ($p<0.05$). The highest number of type IIX fibres was found in the LTL muscle, followed by the SM, ST, and SDF ($p<0.05$). Type I fibre density was significantly higher in the PM than in the SM, RA, ST, and LTL, whereas the GA showed a significantly higher type IIA fibre density than the other muscles ($p<0.05$). The RA group showed the lowest type IIX fibre density ($p<0.05$). In terms of total fibre density, GA showed the highest value, whereas RA showed the lowest ($p<0.05$). HCA revealed the greatest similarity between SM and PM, which shared similar trends in bundle size, fibre length, CSA, and total fibre density.

These two muscles were clustered with the ST muscle (0.43). In contrast, the LTL and SDF showed distance of 0.32. However, the RA and DP muscles showed lower similarities showing higher than 1.0 distance.

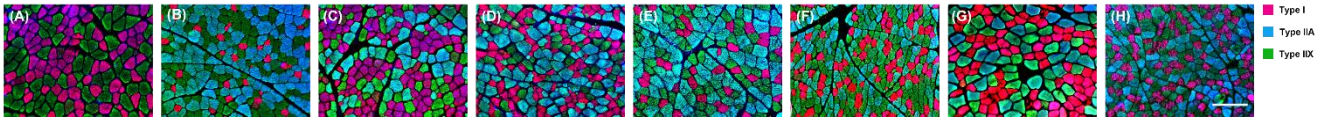


Figure 1. Representative stained images of caprine skeletal muscles. Each section of caprine muscle was stained using myosin heavy chain isoform-specific primary antibodies (BA-F8, SC-71, BF-35, and 6H1) with secondary anti-IgG and anti-IgM antibodies conjugated to Alexa Fluor dyes 405, 488, and 594. (A) DP, *diaphragm*; (B) LTL, *longissimus thoracis et lumborum*; (C) PM, *psoas major*; (D) SM, *semimembranosus*; (E) ST, *semitendinosus*; (F) GA, *gracilis*; (G) RA, *rectus abdominis*; (H) SDF, *superficialis digital flexor*. Bar = 150um

Table 1. Comparison of muscle fiber characteristics among the major caprine skeletal muscles.

Muscle	Bundle size (mm^2)	Length (um)			Cross-sectional area (um^2)				Relative area (%)			Fiber density ($number/mm^2$)			
		I	IIA	IIX	I	IIA	IIX	Mean	I	IIA	IIX	I	IIA	IIX	Total
DP	1.83^{ab}	920.62	1324.82^{ab}	*n.d*	898.36^{b}	1132.77^{b}	*n.d*	1000.97^{b}	48.31^{a}	51.69^{a}	*n.d*	548.83^{ab}	493.30^{b}	*n.d*	1042.13^{d}
(se)	±0.02	±139.46	±154.83	*n.d*	±70.44	±127.90	*n.d*	±84.73	±1.96	±1.96	*n.d*	±30.88	±63.86	*n.d*	±93.05
LTL	1.95^{a}	1567.61	1400.79^{a}	1187.59	478.73^{d}	571.10^{de}	999.90^{bc}	762.64^{c}	6.82^{e}	31.10^{bc}	62.08^{a}	146.10^{d}	562.78^{b}	654.30^{a}	1363.18^{bc}
(se)	±0.02	±389.64	±120.56	±119.44	±39.59	±43.08	±110.19	±67.35	±0.26	±1.04	±0.97	±10.31	±53.26	±61.91	±120.43
PM	1.90^{ab}	856.51	854.89^{c}	1003.18	734.44^{c}	786.64^{cd}	1204.62^{b}	876.40^{bc}	44.15^{ab}	15.81^{f}	40.03^{c}	615.98^{a}	210.47^{de}	348.15^{c}	1174.60^{cd}
(se)	±0.01	±52.85	±85.20	±136.42	±53.98	±70.81	±98.71	±63.52	±1.30	±0.60	±1.19	±38.57	±19.83	±36.88	±85.84
SM	1.50^{bc}	874.10	802.05^{c}	1041.38	583.00^{d}	776.71^{cd}	1251.81^{b}	876.38^{bc}	26.59^{c}	20.24^{ef}	53.17^{b}	463.89^{bc}	269.36^{de}	433.16^{bc}	1166.42^{cd}
(se)	±0.01	±133.10	±47.69	±104.76	±43.13	±45.97	±87.39	±59.26	±1.98	±1.90	±0.63	±37.95	±33.10	±22.20	±68.99
ST	1.27^{c}	684.57	1013.86^{abc}	1356.62	843.70^{bc}	882.73^{c}	1303.32^{b}	1048.71^{b}	17.61^{d}	29.14^{bc}	53.25^{b}	211.56^{d}	337.89^{cd}	438.38^{bc}	987.83^{de}
(se)	±0.01	±99.56	±102.87	±218.12	±34.79	±58.43	±168.98	±88.02	±0.82	±1.27	±1.63	±15.44	±25.71	±43.42	±74.82
GA	0.33^{d}	933.07	884.72^{bc}	1104.08	437.92^{d}	457.79^{e}	633.03^{d}	513.14^{d}	24.05^{c}	32.74^{b}	43.21^{c}	572.21^{ab}	721.98^{a}	699.84^{a}	1994.04^{a}
(se)	±0.00	±295.01	±132.93	±148.79	±43.95	±47.63	±76.17	±53.38	±3.16	±3.18	±1.83	±405.32	±50.29	±62.56	±177.45
RA	0.44^{d}	1275.24	939.28^{abc}	1489.68	1133.38^{a}	1430.89^{a}	2005.60^{a}	1402.74^{a}	40.78^{b}	27.36^{cd}	31.87^{d}	361.84^{c}	193.43^{e}	161.57^{d}	716.85^{e}
(se)	±0.00	±371.23	±217.30	±454.22	±49.53	±73.35	±94.79	±46.19	±1.34	±1.16	±1.81	±12.83	±11.08	±13.15	±22.01
SDF	0.31^{d}	1275.54	1184.80^{abc}	1042.72	509.00^{d}	564.18^{de}	889.33^{cd}	670.03^{cd}	26.17^{c}	23.65^{de}	50.18^{b}	508.60^{ab}	440.26^{bc}	578.35^{ab}	1527.21^{b}
(se)	±0.00	±49.78	±115.27	±63.72	±46.80	±62.63	±61.94	±48.81	±3.70	±1.59	±4.05	±44.29	±45.86	±52.24	±87.91

Data are means±SD. Different superscripts ($^{a-f}$) indicate significant differences between the muscles ($P < 0.05$). DP, *diaphragm*; LTL, *longissimus thoracis et lumborum*; PM, *psoas major*; SM. *semimembranosus*; ST, *semitendinosus*; GA, *gracilis*; RA, *rectus abdominis*; SDF, *superficialis digital flexor*; *n.d*, not detected.

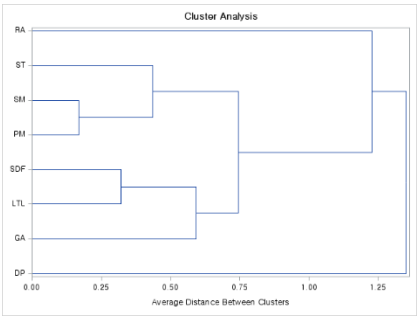


Figure 2. The hierarchical cluster analysis among the caprine muscles. DP, *diaphragm*; LTL, *longissimus thoracis et lumborum*; PM, *psoas major*; SM. *semimembranosus*; ST, *semitendinosus*; GA, *gracilis*; RA, *rectus abdominis*; SDF, *superficialis digital flexor*

IV. CONCLUSION

Among the major skeletal muscles examined in goats, the RA and DP muscles exhibited distinct characteristics, primarily due to the large CSA and absence of type IIX fibres, respectively, leading to the lowest similarity in fibre traits compared to the others. The PM and SM showed the greatest similarity, sharing similar muscle fibre sizes and densities. The LTL muscle is characterised by a relatively low proportion of type I fibres. These preliminary results, due to the limited sample, suggest that the fusiform muscles were more similar, whereas other architecture types did not show a consistent trend. Further studies are needed to validate these findings and advance our understanding of caprine meat quality and its utilization.

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