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EFFECT OF STORAGE CONDITION ON SARCOMERE LENGTH MEASUREMENT IN DIFFERENT BOVINE MUSCLES

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I. INTRODUCTION

Sarcomere length is a critical factor influencing meat tenderness and is commonly measured using laser diffraction [1]. However, this analysis is commonly used in fresh samples, since the effect of freezing and subsequent thawing on sarcomere length remains unclear. This study evaluates whether freezing affects sarcomere length in three bovine muscles with diverse fibre composition. Understanding these effects is essential for ensuring accurate muscle structure assessment in meat science research, particularly when using frozen samples for tenderness evaluation.

II. MATERIALS AND METHODS

Three bovine muscles, *Longissimus thoracis et. Lumborum* (LTL), *Semimembranosus* (SM), *Psoas major* (PM), were sampled from 6 beef carcasses (N = 8 LTL, 7 SM, and 8 PM), subjected to different processing conditions. The muscle samples were sampled at 3 days post-mortem and either preserved immediately or frozen at the same timepoint (stored at -20 °C for 3 weeks and thawed at 4 °C). Muscle cores (2 x 1 x 1 cm) with fibres running longitudinally were excised and stored in 5% glutaraldehyde solution for 4h at 4 °C, followed by storage in 0.2 M sucrose solution. Cores in sucrose solution were stored at 4 °C and used for analysis within 7 days. Sarcomere length was measured in µm using the laser diffraction unit [1] to assess if there was an impact on sarcomere length due to freeze/thaw treatment. Statistical analysis was performed using paired t-test and REML ANOVA in Minitab to assess differences between treatment groups. Main effects and interactions were visualized using Bland and Altman plot [2] and interaction plots.

III. RESULTS AND DISCUSSION

The impact of storage condition on the sarcomere length of three different muscles (LT, PM, and SM) was assessed, with results presented using Bland and Altman plot (Figure 1) and interaction plot (Figure 2). The paired t-test ($p = 0.14$) and ANOVA ($p = 0.19$) revealed no significant difference between fresh and frozen sarcomere length measurements, indicating that the freezing process did not significantly alter sarcomere length in these muscles; although a trend for shorter sarcomeres after freezing was observed in the PM muscle. This suggests that sarcomere length measurements from frozen samples can reliably be used to determine the corresponding values from fresh samples, regardless the muscle type. Additionally, the Bland and Altman plot suggests that, on average, there is little systematic bias between fresh and frozen measurements, as indicated by the green line that lies near to zero.

Most points fall within the upper and lower limits of agreement, which suggests that fresh and frozen measurements are in agreement. Regarding the sarcomere length variation between muscles, PM samples exhibited the longest sarcomeres, ranging from approximately 2.5 to 3.5 µm. In contrast, LT and SM muscles showed shorter sarcomere lengths, predominantly clustering around 1.8 to 2.2 µm. This difference in sarcomere length between muscles is consistent with known variations in muscle fiber composition and function, with PM being a predominantly slow-twitch muscle, which typically has longer sarcomeres compared to the fast-twitch fibers found in LT and SM.

The lack of significant differences between fresh and frozen samples suggests that freezing may be a viable method for preserving sarcomere length in these muscles, allowing for more flexible sampling processing options without compromising measurement accuracy. This finding is particularly useful in meat research and quality control, where timely analysis of fresh samples may not always be feasible due to logistical constraints. Future studies should focus on assessing the impact of freezing process on muscles in a broader range of sarcomere lengths; including those suffering of cold shortening, this will be essential to further validate the reliability of freezing

for sarcomere length analysis. If any interaction is observed a predictive model could be developed to determine the actual correlation between sarcomeres measured fresh or frozen in different muscle types and considering actual sarcomere length.

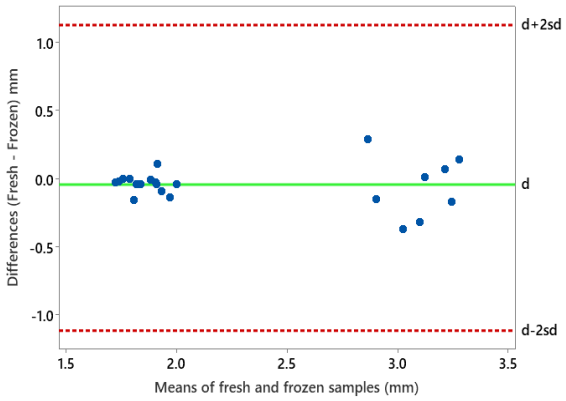


Figure 1 Bland and Altman plot showing the difference between sarcomere lengths measured using fresh and frozen muscle samples. Red lines represent the limits of agreement, while the green line indicates the mean value of the differences.

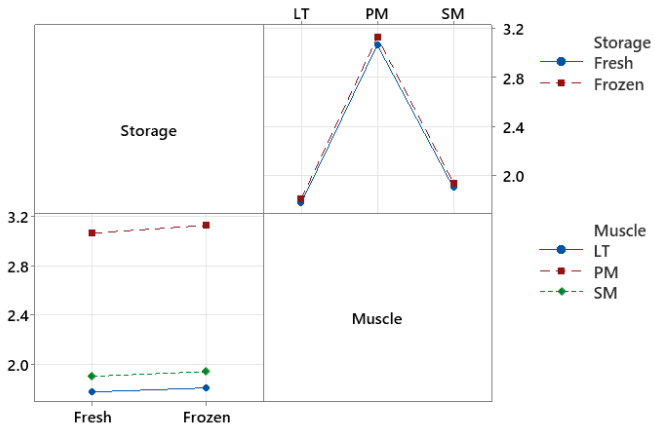


Figure 2. Effect of storage in three muscles and the interaction of storage and muscle; LT *Longissimus thoracis et.lumborum,* SM *Semimembranosus,* PM *Psoas major.*

IV. CONCLUSION

Based on the findings, freezing can be considered a suitable alternative for fresh sarcomere length analysis across all tested muscles, including LT, PM, and SM, especially when time is a constraint. This has practical implications for meat research and quality control, where fresh sample processing may not always be feasible. Further validation under more diverse processing conditions, including cold shortening, could refine this approach for broader applications; enabling the generation of a predictive model correlating frozen and fresh samples under a variety of processing conditions.

ACKNOWLEDGEMENTS

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