

In vitro analysis of mushroom proteases that may tenderize beef

Jing-Wei Lee, Michelle N. LeMaster, Eric M. England

INTRODUCTION

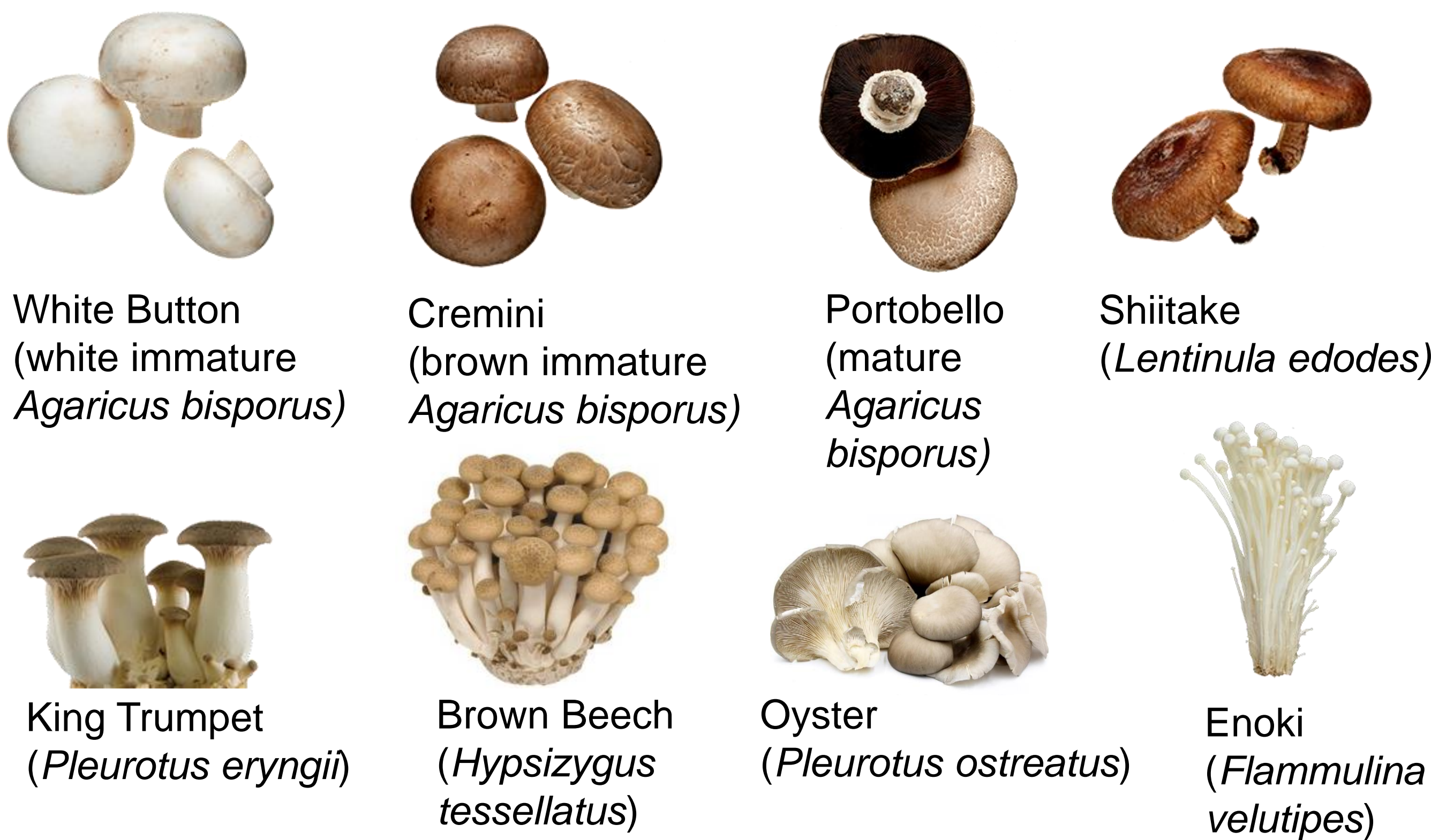
Meat tenderness is an important characteristic that influences consumer purchasing decisions (1). Protease extracts from pineapple, ginger, papaya and kiwi have exhibited broad proteolytic activity that can over-tenderize meat and negatively affect texture and quality (2). Therefore, identification and evaluation of other proteases capable of tenderizing beef is necessary. Previously, mushrooms have been shown to enhance flavor and nutritional composition of meat dishes, as well as having beneficial antioxidant and health effects (3). Mushrooms also contain a variety of proteases that were analyzed in this study for their ability to proteolyze bovine myofibrillar proteins using an *in vitro* model system.

AIM

Determine mushroom species containing proteases capable of proteolyzing bovine myofibrillar proteins.

MATERIALS

The following eight mushroom varieties were obtained from local grocery stores for analysis:



<http://www.blendandextend.ca/fresh-mushrooms-and-your-health/mushroom-varieties/>
http://www.specialtyproduce.com/produce/Oyster_Mushrooms_703.php
<https://matome.naver.jp/odai/2139452359361475301/2139459103899322103>

METHODS

- Fresh mushrooms were homogenized in 20 mM Tris buffer (pH 8.0). The homogenate was combined with purified bovine myofibrils and incubated at 25°C. Samples were collected at 0, 30, 60, 240, and 1440 min for analysis.
- Myofibrillar proteins from each time-point were solubilized and separated using sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE). Gels were then stained overnight, destained for 2 hours, and imaged using an Azure c600 with NIR capabilities.
- A caseinolytic assay quantified protease activity of mushroom homogenates at 280nm using a NanoDrop spectrophotometer.

RESULTS

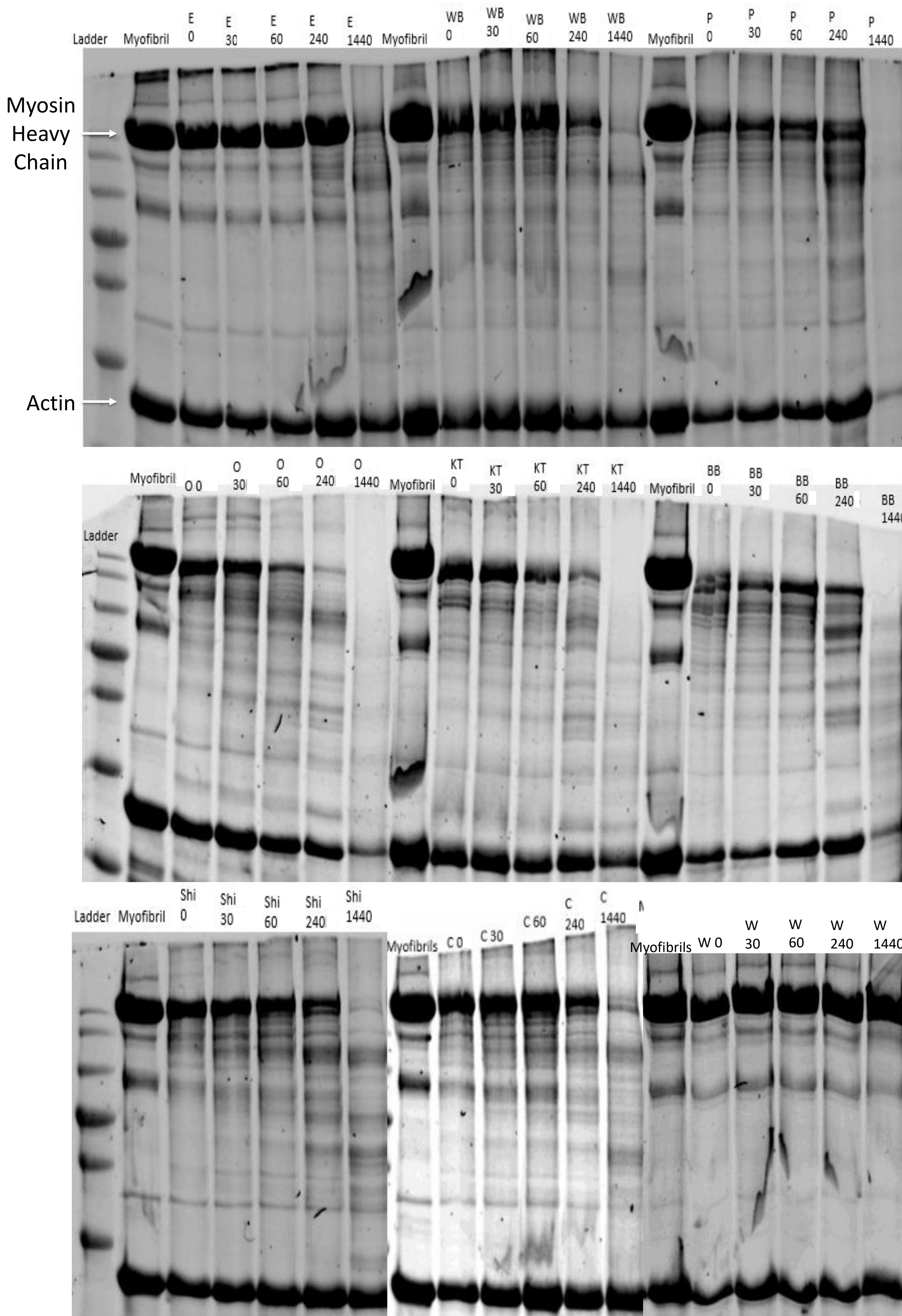


Figure 1. SDS-PAGE gels of water/control (W) enoki (E), white button (WB), portobello (P), oyster (O), king trumpet (KT), brown beech (BB), shiitake (Shi) and cremini (C) mushroom homogenates on bovine myofibrillar proteins after 0, 30, 60, 240 and 1440 min of incubation.

RESULTS

Table 1. Caseinolytic activity assay of the proteases from eight mushroom varieties

Mushroom Variety	Protease Activity (U/mL)
Oyster	1.06
Portobello	0.70
King Trumpet	0.68
Shiitake	0.45
Brown Beech	0.34
White Button	0.31
Cremini	0.19
Enoki	0.03

DISCUSSION

The SDS-PAGE gel images align with the caseinolytic activity assay. Oyster, portobello, and king trumpet varieties proteolyzed bovine myofibrillar proteins and exhibited numerically increased caseinolytic activity. Conversely, enoki exhibited numerically lower caseinolytic activity and relatively limited proteolysis of bovine myofibrils.

CONCLUSIONS

The data indicated that all eight mushroom varieties proteolyzed myofibrillar proteins. Therefore, these results support the possibility that mushroom proteases may be able to tenderize beef. Future research trials will focus on utilizing the mushroom homogenates *in vivo* on beef muscles to determine if proteolysis and/or over-tenderization occurs.

BIBLIOGRAPHY

- Voges, K., Mason, C., Brooks, J., Delmore, R., Griffin, D., Hale, D., . . . Savell, J. (2007). National beef tenderness survey – 2006: Assessment of Warner–Bratzler shear and sensory panel ratings for beef from US retail and foodservice establishments. *Meat Science*, 77(3), 357-364.
- Ha M., Bekhit A.E.D.A., Carne, A., Hopkins, D.L. (2012). Characterisation of commercial papain, bromelain, actinidin and zingibain protease preparations and their activities toward meat proteins. *Food Chemistry*, 134(1), 95-105.
- Alnoumani, H., Ataman, Z.A., Were, L. (2017). Lipid and protein antioxidant capacity of dried *Agaricus bisporus* in salted cooked ground beef. *Meat Science* 129(3), 9-19.

ACKNOWLEDGEMENTS

This research was conducted as part of the OSU Department of Animal Sciences Undergraduate Research Experience (ASURE), and was partially funded by the OSU Second-Year Transformational Experience Program (STEP).