Effects of acetylation on dissociation and phosphorylation of actomyosin in ovine muscle during incubation at 4 °C in vitro

Yejun Zhang, Xin Li, Dequan Zhang

Institute of Food Science and Technology, Chinese Academy of Agricultural Sciences/Key Laboratory of Agro-products Quality & Safety in Harvest, Storage, Transportation, Management and Control, Ministry of Agriculture and Rural Affairs, Beijing, China

Introduction: Tenderness is one of the most important meat quality characteristics. A large number of studies have reported that actomyosin dissociation affects meat tenderness (Hopkins et al., 2011). Myosin and actin are the major components of thick and thin filaments respectively. Previous studies have found that phosphorylation of myosin heavy chain (MHC) and actin affect actomyosin dissociation (Cao et al., 2020; Chen et al., 2016). Protein acetylation, which is another important post-translational modification, has been reported to be involved in the regulation of muscle contraction (Jiang et al., 2019). How MHC and actin acetylation regulate muscle contraction and phosphorylation is still unclear. Thus, this study aimed to investigate the influence of MHC and actin acetylation on their phosphorylation and the possible mechanism of protein acetylation and phosphorylation co-regulating actomyosin dissociation in muscle postmortem.

Materials and methods: The longissimus thoracis (LT) muscles were removed from twelve lamb carcasses within 30 min postmortem and then minced and homogenized. The extracted protein samples were diluted to 4 mg/mL and subjected to the following three treatments (n = 12): (1) high acetylation group: adding lysine deacetylase inhibitors; (2) middle acetylation group: adding dimethyl sulfoxide; and (3) low acetylation group: adding lysine acetyltransferase inhibitors. The 1 mM acetyl-CoA was added to each sample to provide acetyl group for acetylation. All three treatments were incubated at 4 ℃ and samples were collected after 0 min, 30 min, 4 h, 12 h, 1 d, 2 d and 3 d incubation.

Results: The acetylation levels of MHC and actin in the high acetylation group were significantly higher than those in middle acetylation and low acetylation groups ($P < 0.05$). The phosphorylation level of MHC in the high acetylation group was significantly lower than that in the middle acetylation and low acetylation groups at 4 h, 1 d and 2 d of incubation ($P < 0.05$). The actin phosphorylation level in the high acetylation group was significantly lower than that in the low acetylation groups at 1 d and 3 d of incubation ($P < 0.05$). Acetylation may decrease/increase the activity of MHC and actin kinase/phosphatase, which in turn reduces its phosphorylation level (Yang & Seto, 2008). The actomyosin dissociation and ATPase activity in the high acetylation group were significantly lower than those in the low acetylation group at 30 min, 4 h, 12 h, 1 d and 3 d of incubation ($P < 0.05$). Previous studies reported that MHC phosphorylation promoted actomyosin dissociation and the phosphorylation of actin tyrosine-53 destroyed the stability of muscle filaments (Liu et al., 2006; Norwood et al., 2018). The results showed that protein acetylation makes the structure of actomyosin more stable by inhibiting phosphorylation of the MHC and actin.

Conclusion: Acetylation of MHC and actin led to a lower actomyosin dissociation. Acetylation of MHC and actin significantly inhibited their phosphorylation in lamb during incubation at 4 ℃. MHC and actin acetylation may inhibit actomyosin dissociation by inhibiting their phosphorylation at 4 ℃ in vitro.

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