

Condition-dependent Adenosine Monophosphate Decomposition Pathways by Endogenous Enzyme in Striated Adductor Muscle from Japanese scallop (*Patinopecten yessoensis*)

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[Objective] The post-mortem ATP and related compounds have been extensively studied for quality evaluation of fish or shellfish for over 60 years. "K-value" is based on the ATP breakdown and the subsequent formation of inosine (HxR) and hypoxanthine (Hx). It is well-known that IMP is the main nucleotide present in most fish species such as mackerel, whereas AMP remains the major nucleotide in crustaceans and mollusks. It is controversial whether IMP in scallops generates. Therefore, the aim of this study was to investigate the AMP decomposition pathway in scallop adductor muscle and its influencing factors, such as EDTA or EGTA addition, heating, ions metal concentration change, et al.

[Methods] Crude enzyme solution was made from scallop striated muscle after chopping and homogenization at 4°C, and divided into three groups: control group, with EDTA or EGTA addition. Then, AMP, IMP and AdR were added into the crude enzyme solution as the substrate to detect the decomposition rate at 25°C. In addition, the crude enzyme solution was dialyzed to remove metal ions, and the deionized crude enzyme solution was divided into two groups: the control group (C) and the addition of 0.1% chloramphenicol (CP). Subsequently, AMP was added and changes in its ATP-related compounds was measured by HPLC at 25°C.

[Results] The results showed that IMP accumulated due to AMP decomposition via endogenous enzymes in scallops stored at both 4°C and 20°C. The AMP decomposition rate was highest in the supernatant of homogenized scallop adductor muscle, followed by the suspended solution and precipitate, while IMP could not be decomposed in scallop. AdR decomposed rapidly since high activity of AdR deaminase. The IMP generation rate increased dramatically in scallop crude enzyme solution containing 5 mM EDTA. Moreover, small amount of IMP also generated rapidly in scallop crude enzyme solution containing 5 mM EGTA within 2 h. Moreover, K⁺ was confirmed to promote the decomposition of AMP to IMP, Mg²⁺ promoted the decomposition of AMP to AdR, and Ca²⁺ slightly inhibit the decomposition of AMP. Meanwhile, the protein content and protein composition were also measure during dialysis and incubation. The results demonstrated that, the protein in the scallop enzyme solution of control group gradually denatured due to microbial activity during incubation, while CP group did not. Adenosine deaminase and adenosine kinase were detected in denatured protein fraction (40 kDa) by LC-MS/MS.

[Conclusion]. The decomposition path of AMP in scallops could be changed when concentration of metal ions change. In addition, the production of IMP in scallop was confirmed to be produced by endogenous enzymes via decomposition of AMP, while the generation of HxR and Hx is caused by endogenous enzymes and microbial activities.

Key words: AMP; endogenous enzyme; EDTA; IMP