

Studies on Enhancement of Umami Compounds in Seasoning through Halotolerant Nucleotidases

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Summary

Shewanella species are widely distributed in sea, brackish, and fresh water areas, growing psychrophilically or mesophilically, and piezophilically or piezo-sensitively. Here, membrane-bound 5'-nucleotidases (NTases) from deep-sea *Shewanella violacea* and brackish water *Shewanella amazonensis* were examined from the aspect of NaCl tolerance in order to gain an insight into protein stability against salt. Both NTases were single polypeptides with molecular masses of ~59 kDa, as determined on mass spectroscopy. They similarly required 10 mM MgCl₂ for their activities, and they exhibited the same pH dependency and substrate specificity for 5'-nucleotides. However, *S. violacea* 5'-nucleotidase (SVNTase) was active enough in the presence of 2.5 M NaCl, whereas *S. amazonensis* 5'-nucleotidase (SANTase) exhibited significantly reduced activity with the same concentration of the salt. Although SVNTase and SANTase exhibited high sequence identity (69.7%), differences in the ratio of acidic to basic amino acid residues and the number of potential salt bridges maybe being responsible for the difference in the protein stability against salt. 5'-Nucleotidases from these *Shewanella* species will provide useful information regarding NaCl tolerance, which may be fundamental for understanding bacterial adaptation to growth environments.