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# Comparative study on antioxidant properties of gelatin peptides derived from marine fish species in different in vitro assay systems

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## Introduction

A great interest is developing in the present world to identify new antioxidant compounds from different natural sources. Gelatin peptides contain mainly hydrophobic amino acids and abundance of these amino acids favor to have higher affinity to oil and better emulsifying ability. In the present research, peptides derived from enzymatic hydrolysates of skin gelatin resulted from four different marine fish species were tested for their antioxidant properties in different in vitro assay systems.

## Methods and Materials

### Preparation of gelatin peptides

Skins of Hoki (*Johnius belengerii*), conger eel (*Conger myrister*), Jumbo squid (*Dosidicus gigas*) and chinook salmon (*Oncorhynchus tshawytscha*) were obtained as processing wastes and gelatin was extracted giving hot water treatment as described by Kim et al., (1994). All four gelatin extracts were hydrolyzed using gastrointestinal enzymes, trypsin, pepsin and alpha-chymotrypsin that obtained commercially. Tryptic hydrolysates were further separated using three different ultrafiltration molecular weight cutoffs and separated fractions were lyophilized. Amino acid compositions were identified and quantified using an automatic amino acid analyzer.

### Linoleic acid oxidation inhibition assay

In vitro lipid peroxidation inhibition activity of peptides was determined by assessing their ability to inhibit oxidation of linoleic acid in an emulsified model system (Osawa et al., 1985) and activities were compared with commercial antioxidants, alpha-tocopherol

or butylated hydroxytoluene (BHT).

#### Radical scavenging activity assay

Ability of gelatin peptides to scavenge Hydroxyl (Rosen et al., 1984) and carbon-centered (Hiramoto et al., 1993) radicals were assayed as described previously using ESR spectrometry.

#### Viability enhancement of oxidation induced cells

Human fibroblasts cells were incubated with different nontoxic concentrations of purified peptides (25 to 100 ug/ml) and exposed to t-BHP, and the percentage cell viability was determined using MTT cell viability assay.

### Results and Discussion

Our results report the antioxidant capacity of skin gelatin peptides derived from four different marine fish species in different aspects of oxidation. During research, it was observed that gelatin peptides could act as an antioxidant against linoleic acid peroxidation and the activity was closer to the highly active synthetic antioxidant butylated hydroxytoluene (BHT). They exerted substantial scavenging on both hydroxyl and carbon centered radicals. Viability of radical mediated oxidation induced human lung fibroblasts was enhanced following the treatment of gelatin peptides and it was presumed to be that the peptide involved in maintaining the redox balance in the cell environment. Further, higher scavenging capacities and incapability of ion chelation, fully support the hypothesis that free radical quenching indeed is the main antioxidative mechanism of gelatin peptides derived from skin of marine fish species.

### References

- Hiramoto K., H. Johkoh, K.I. Sako and K. Kikugawa. 1993. DNA breaking activity of the carbon-centered radical generated from 2,2'-azobis(2 amidinopropane) hydrochloride (AAPH). *Free Radic Res Commun* 19:323-332.
- Kim S.K., H.G. Byun, and E.H. Lee. (1994). Optimum extraction conditions of gelatin from fish skins and its physical properties. *J. Korean Ind.Eng. Chem* 5:547-559.
- Osawa T., and M. Namiki. 1985. Natural antioxidants isolated from eucalyptus leaf waxes. *J Agric Food Chem* 33:777-780.
- Rosen G.M., and E.J. Rauckman. 1984. Spin trapping of superoxide and hydroxyl radicals. In: Packer L, (Ed). *Methods in Enzymology*. Orlando: Academic Press. pp.189-209.