Protein hydrolysates from fish by-products: a potential source of bioactive peptides

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State of the art

Fish by-products can be enzymatically transformed into high value-added products such as fish protein hydrolysates (FPH). They are rich in bioactive peptides, essential amino acids, and other nutritional components, making them suitable for use in food, nutraceutical and cosmetic industries. The presence of these compounds is associated with health benefits due to their biological properties (antioxidant, antihypertensive, anticancer, antidiabetic activities, etc.) but they also exhibit high functional properties. With a growing focus on health and well-being, there is an increasing demand for natural and functional ingredients, so FPH can be a high-quality protein source with significant biological properties.

Objectives

- Preparation of FPH from fresh salmon heads (*Salmon salar*) and frozen Cape hake by-products (*Merluccius capensis*) using Alcalase.
- Chemical and nutritional characterization of these hydrolysates.
- Evaluation of FPH antioxidant (DPPH, ABTS and Reducing Power), chelating (Cu^{2+} and Fe^{2+}), antihypertensive and antidiabetic properties.
- Characterization of fatty acid profile of the oil recovered during the hydrolysis.

Raw Material (RM)

	Fresh Salmon heads	Frozen Cape hake frames
Protein (%)	13.8 ± 0.9	14.3 ± 1.4
Moisture (%)	61.2 ± 0.5	81.6 ± 0.0
Ash (%)	2.39 ± 0.19	2.76 ± 0.13
Fat (%)	19.8 ± 0.2	0.5 ± 0.0

Preparation of Fish Protein Hydrolysates

Raw material:H_2O dd (1:2)
↓
Alcalase addition (pH 8.5; 60°C; 1% v/w RM)
↓
Reaction (3 h; pH adjustment)
↓
Enzyme inactivation (95°C; 20 min)
↓
Centrifugation (10000 g; 20 min; 4°C)
↓
Freeze-drying (liquid fraction)

Fish Protein Hydrolysates

	Salmon heads hydrolysates (HPS)	Cape hake hydrolysates (HPP)
Yield (%)	68.1 ± 2.3	75.5 ± 3.4
DH (%)	26.2 ± 0.2	20.3 ± 0.0
Protein (%)	75.6 ± 0.9	76.8 ± 0.5
Moisture (%)	7.6 ± 0.5	4.4 ± 0.1
Ash (%)	13.45 ± 0.31	16.69 ± 0.16
Fat (%)	2.1 ± 0.1	2.7 ± 0.0

* DH = Degree of hydrolysis

Oil recovered **90 %** | **0 %** Oil recovered

HPS Oil recovered

Oleic acid (%)	38.90 ± 0.01
EPA (%)	3.37 ± 0.01
DHA (%)	3.31 ± 0.02
SFA (%)	15.55 ± 0.02
MUFA (%)	52.57 ± 0.02
PUFA (%)	30.78 ± 0.00
ω3/ω6 ratio	0.95 ± 0.00
Atherogenic index (AI)	0.24 ± 0.00
Thrombogenic index (TI)	0.18 ± 0.00
h/H ratio	5.45 ± 0.01

Recoverd oil from HPS with high nutritional potential
- Low AI and TI
- High h/H ratio

EPA=Eicosapentaenoic acid;
DHA=Docosahexaenoic acid;
SFA= total saturated fatty acids;
MUFA=total monounsaturated fatty acids;
PUFA=Total polyunsaturated fatty acids;
h/H=Hypocholesterolemic to hypercholesterolemic ratio

Amino Acids Profile

Essential amino acids (g/100g sample)	RM Salmon	HPS
HIS	0.58 ± 0.10	1.06 ± 0.37
LYS	3.38 ± 0.46	6.09 ± 0.92
LEU	2.22 ± 0.25	4.60 ± 0.02
VAL	1.53 ± 0.12	3.13 ± 0.03
MET	0.47 ± 0.08	1.44 ± 0.11
CYS	0.51 ± 0.06	1.05 ± 0.01
PHE	1.21 ± 0.13	2.47 ± 0.06
TYR	1.05 ± 0.10	2.06 ± 0.06
ILE	1.28 ± 0.12	2.60 ± 0.02
THR	1.93 ± 0.20	3.53 ± 0.13
TAA (g/100g sample)	33.77	70.14
TEAA(g/100 sample)	14.15	28.04
TEAA/TAA (%)	41.91	39.98

* TAA = Total amino acids; TEAA= Total essential amino acids

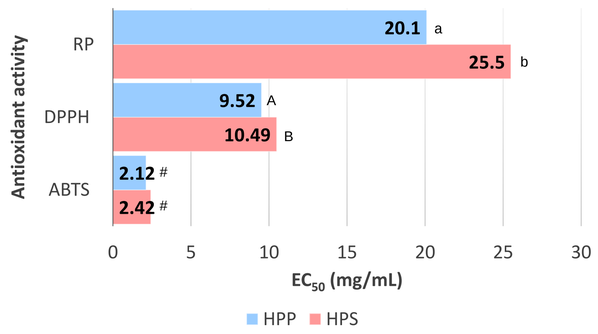
Essential amino acids (g/100g sample)	RM Hake	HPP
HIS	1.33 ± 0.10	0.75 ± 0.15
LYS	5.68 ± 1.67	6.24 ± 0.61
LEU	5.95 ± 0.05	5.42 ± 0.11
VAL	3.75 ± 0.22	3.43 ± 0.03
MET	1.90 ± 0.50	1.56 ± 0.08
CYS	0.79 ± 0.07	0.89 ± 0.03
PHE	3.13 ± 0.17	2.89 ± 0.06
TYR	2.69 ± 0.16	2.42 ± 0.04
ILE	3.40 ± 0.24	2.93 ± 0.07
THR	4.03 ± 0.24	4.02 ± 0.01
TAA (g/100g sample)	74.38	72.57
TEAA (g/100 sample)	32.65	30.53
TEAA/TAA (%)	43.89	42.07

* TAA = Total amino acids; TEAA= Total essential amino acids

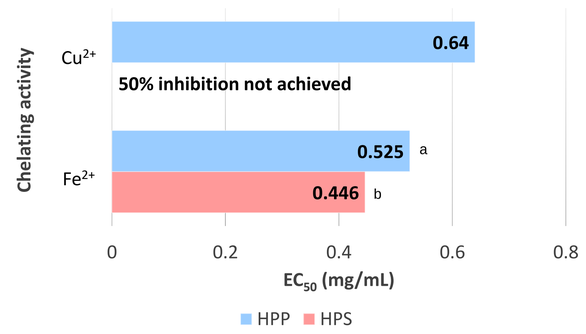
Antioxidant and Biological Activities

Different letters (a,b or A,B) or symbols (#, *) mean significantly different values

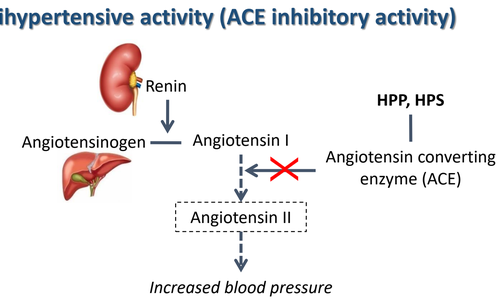
DPPH and ABTS radical scavenging activities and Reducing Power (RP)

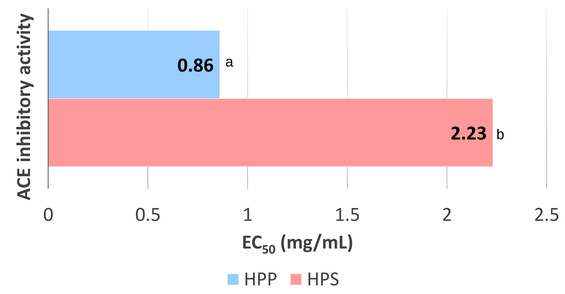


Fe^{2+} and Cu^{2+} chelating activities

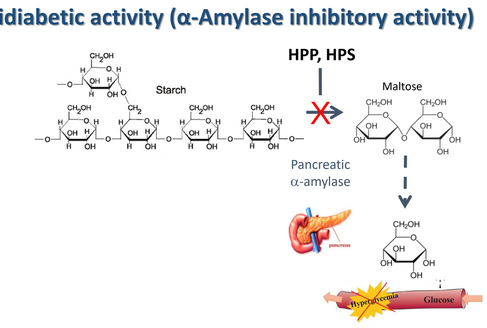


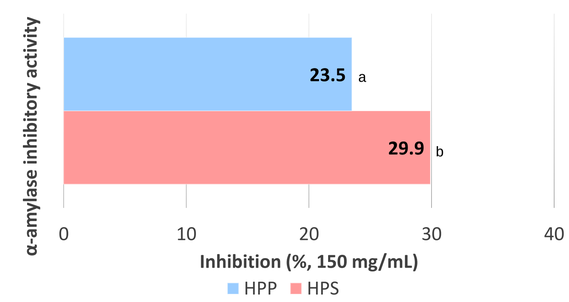
Antihypertensive activity (ACE inhibitory activity)





Antidiabetic activity (α-Amylase inhibitory activity)





Conclusions

- ✓ HPP presented higher yield but lower degree of hydrolysis;
- ✓ HPP and HPS had a similar composition of essential amino acids, with LEU, PHE and TYR having a higher content in HPP;
- ✓ HPP showed the highest DPPH and ABTS radical scavenging activities, reducing power and Fe^{2+} chelating activity;
- ✓ ACE inhibitory activity of HPP was relatively high but α-amylase inhibitory activity of both FPH was very low;
- ✓ EPA + DHA content of oil recovered during the hydrolysis of salmon heads was around 7%;
- ✓ AI, TI and h/H ratio highlighted the good nutritional quality of HPS oil and its high cardio-protective effect;
- ✓ The study reveals the FPH valuable interest as potential food ingredient or nutraceutical, thus enabling the valorisation of these by-products under a circular economy context.

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