

antihypertensive activity

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Introduction

The production of foods with antihypertensive activity has particular interest given the important role of diet in the prevention and treatment of hypertension. Enzymatic hydrolysis using one or a combination of food-grade enzymes can be a cheap and reproducible alternative to fermentation for the production of antihypertensive peptides.

Objective:

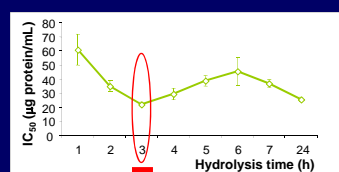
To identify active peptides from a enzymatic hydrolysate of bovine casein with the aim of producing a novel food ingredient based on this hydrolysate.

Laboratory-scale

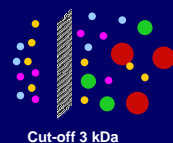
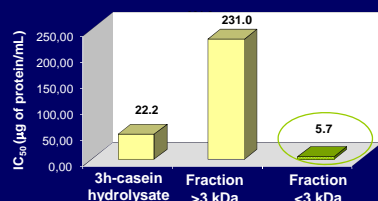
Identification of antihypertensive peptides

Hydrolysis

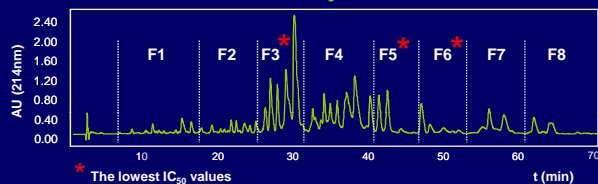
Isoelectric casein (0.5% w/v) $\xrightarrow{\text{Pepsin}}$ T: 37 °C; E/S: 3,7 %, t: 1-24 h



ACE-inhibitory activity after UF



Semipreparative RP-HPLC

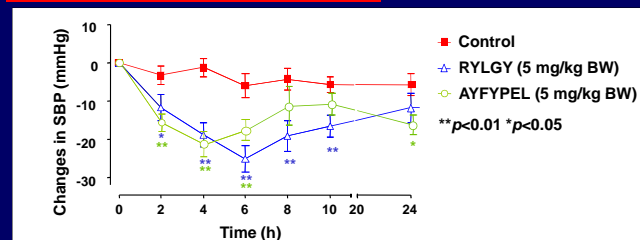


Identification of peptides included in the most active fractions by RP-HPLC-MS and MS/MS

Determination of ACE-inhibitory activity and ORAC-FL

Fraction	Casein fragment	Sequence	IC ₅₀ (µM)	ORAC-FL (µmol Trolox/ µmol peptide)
F3	α _{S1} -CN f(90-94)	RYLGY	0.71	2.83
	α _{S2} -CN f(89-95)	YQKFPQY	20.08	2.03
F5	α _{S1} -CN f(143-149)	AYFYPEL	6.58	3.22
	α _{S1} -CN f(24-31)	FVAPFPEV	475.89	< 0.03
F6	β-CN f(134-140)	HLPLPLL	34.40	0.06
	α _{S1} -CN f(25-32)	VAPFPEV	362.50	0.05

Antihypertensive activity



CONCLUSIONS

Casein hydrolysis with pepsin leads to the formation of antihypertensive peptides.

Peptide RYLYG is one of the most potent ACE-inhibitors identified from food proteins (IC₅₀, 0,71 µM).

RYLYG and AYFYPEL had antihypertensive activity in SHR, and was comparable to VPP.

Hydrolysis was successfully scaled-up, and antihypertensive activity of the casein-ingredient was confirmed in SHR.

The casein-ingredient was well incorporated into yogurt, and antihypertensive peptides remained stable in yogurt during storage at 4 °C.

Production at large-scale

Preparation of casein hydrolysate

Food grade conditions

- Casein
- Water
- Enzyme

Tank 140 L-1500 L

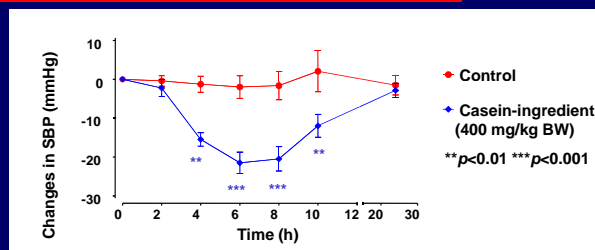
Spray- /Freeze-drying



LOWPEPT®

- High content in antihypertensive peptides
- Comparable IC₅₀ value
- Good reproducibility

Antihypertensive activity in SHR

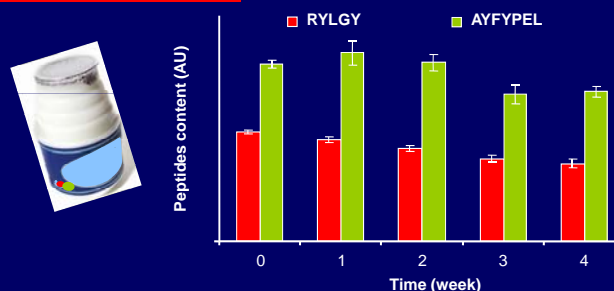


Incorporation into food matrix

Casein-ingredient was incorporated into yogurts and stored at 4 °C for a month.

The peptides content was quantified in WSEs by RP-HPLC-MS.

Shelf life at 4 °C



Clinical studies are being carried out to evaluate the antihypertensive action of casein hydrolysate in humans.

References

- Contreras et al. (2009). *Int. Dairy J.* 19, 566-573
- Miguel et al. (2009). *Food Chem.* 112, 211-214

Acknowledgements

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