



Scientific Paper

## Egg protein hydrolysates: New culinary textures

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### Abstract

The purpose of this work was to obtain new textures from egg proteins by enzymatic hydrolysis, to diversify their applications in daily and haute cuisine. Pasteurized liquid egg white, yolk and whole egg were hydrolyzed with a food grade aminopeptidase. Before enzyme inactivation, several ingredients were added, such as flavors and colors in liquid or powdered forms (salt, sugars, fruit or vegetal infusions). This yielded novel gels with various textures – from smooth and creamy to rigid – and light foams with a high foaming capacity and ability to re-incorporate air once collapsed, which were characterized by sensory and texture profile analysis. The elaboration process proved simple and fast, allowing an optimum use of the starting material without by-products. It provided the means to improve the techno-functional properties of the egg as an ingredient and to expand its use in new recipes, as well as in the development of new food products, particularly suitable for people with chewing limitations or digestion problems, overweight, obesity, or sensitive to dairy products.

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### Introduction

Texture is a critical attribute that is essential to the overall quality and acceptability of most food products because, in addition to its relevance to mouthfeel, texture is also a property generally related to freshness (Isaksson et al., 2002). During the last decade, gastronomy has become an important feature of the socio-economic landscape of many countries, being the culinary activity a balance between tradition and technology (García-Segovia et al., 2012). New textures are essential to the success of haute cuisine (Adrià et al., 2005; Blumenthal, 2008) and, nowadays, it is usual to find traditional dishes that have been modified by various cooking techniques to present very different textures, such as slush, ice cream, sorbet, crunchy, foam, gelatine spheres and crystals. While it is generally recognized that the development of successful products is a difficult and time-consuming task, these textures are possible

through technological and culinary innovations in a joint work between scientists and cooks.

Egg is one of the highest quality protein sources in our diet and one of the most versatile ingredients used in cooking. It is well known that the ability of egg to form and stabilize different food structures is due to the protein fraction and its physicochemical properties. Egg proteins act in food products as texture improvers and stabilizers, controlling consistency and water retention, by virtue of their foaming, emulsifying, gelling and heat setting properties (Mine, 1995). Therefore, they are widely used as ingredients in the food industry for different purposes.

Enzymatic hydrolysis is a well-known method for increasing the added value of food proteins by modifying their physical and nutritional properties. Breaking of peptide bonds can change these properties in three ways: reducing the molecular weight, increasing the number of ionizable groups and causing the exposure of hydrophobic groups. Enzymatic hydrolysis improves the solubility of food proteins in a wide pH range and modulates their surface or interfacial properties, essential, for instance, for the stabilization of emulsions and foams (Foegeding and Davis, 2011). Thus, hydrolysis modifies the

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Table 1  
Egg-derived products.

	Material	Centrifugation	Post-treatment	Final product
<b>Product 1</b>	Egg white	Yes	No	Cream
<b>Product 2</b>	Egg white	Yes or manual liquid draining	No	Cottage cheese
<b>Product 3</b>	Egg white	No	No	Junket
<b>Product 4</b>	Whole egg	No	No	Custard dessert/crème caramel
<b>Product 5</b>	Yolk	No	No	Custard dessert
<b>Product 6</b>	Egg white	Yes	Whipping	Foam

sensory quality of proteins, their contribution to texture and flavor – as peptides do have a taste and, in fact, cover the entire range of known taste modalities, including bitter tastes that could cause food rejection (Temussi, 2012) –; but also improves digestibility and nutrient bioavailability and provides health benefits through the release of bioactive peptides and the reduction of their allergenic potential (Tavano, 2013).

The physicochemical properties of a culinary preparation are commonly modified using additives, such as agar-agar, gellan, methylcellulose, locust bean gum, lecithin, sucrose ester or gases, such as carbon dioxide or liquid nitrogen, among others, with an enormous range of potential applications to produce foods of novel and interesting consistencies in the kitchen (Barham et al., 2010). However, to the best of our knowledge, the enzymatic hydrolysis of egg proteins has not been explored in depth for obtaining new textures of culinary use. The inclusion of egg hydrolysates with the right techno-functional properties may lead to the modification or generation of food products new attributes of taste, texture, odor, acidity, rheological properties, stability and appearance. Moreover, it may lead to products that can mimic the properties of dairy products of interest for some population groups for health reasons, like allergies, intolerances or specific diseases.

The aim of this work was to obtain new textures from egg proteins through treatment using food grade enzymes. The objectives were to achieve a pleasant appearance, appropriate sensory characteristics and diverse applications in daily and haute cuisine.

## Material and methods

### Products

Commercial pasteurized whole egg, egg yolk and white were purchased from Pitas Agropecuaria S.L. (Guadalajara, Spain). Food grade aminopeptidase from *Aspergillus oryzae* (Flavorpro 750 MDP) was purchased from Biocatalysts (Cardiff, United Kingdom). HCl food grade CODEX was from Sigma-Aldrich (Tres Cantos, Spain) and natural flavoring liquid extracts were provided by Mario Sandoval (Restaurante Coque, Madrid, Spain).

### General protocol of hydrolysis

Pasteurized liquid egg products were hydrolyzed with *Aspergillus* aminopeptidase at an enzyme to substrate ratio

of 2:100 (w:v). Before hydrolyzes, the egg products were acidified to the optimum enzyme pH, 5.5, with concentrated food grade HCl (37%). Hydrolyzes were carried out at 50 °C under constant stirring in a thermostatic water bath for up to 120 min. Enzyme inactivation was achieved by heating the samples at 95 °C for 15 min in a water bath, followed by cooling at 4 °C. Depending on the required final product, several ingredients were added as flavorings and colorings in liquid or powdered forms (salt, sugars, fruit or vegetal infusions). Because the heating step caused the hydrolyzed proteins to gel, and in order to favor a homogeneous mixture, the addition of ingredients was conducted before enzyme inactivation.

As shown in Table 1, five different gel textures (products 1–5) were obtained, depending on the starting material (whole egg, egg yolk or egg white) and the use of salt or sugar, or centrifugation to favor liquid draining from the egg white gels. In the latter case, following separation of the precipitate (product 1), the supernatant was whipped with a Kitchen Aid Ultra Power Mixer (Kitchen Aid, St. Joseph's, MI) coupled with a stationary bowl and rotating beaters, what resulted in a product with foam texture (product 6).

### Sensory analysis

Panelists were recruited among members of the Institute staff who had previously participated in sensory descriptive tests. Criteria for recruitment were: (1) age (between 18 and 55 years-old), (2) absence of allergy to egg and/or dairy products, (3) consumption of dairy and egg-based desserts at least twice per week, and (4) willingness and availability to participate during testing dates. There were 12 female panelists and 9 male panelists.

A five-point hedonic scale (5 – excellent, 3 – good, 1 – unacceptable) with a clear description of each point was used for the assessment of appearance, texture and flavor of the 6 products (UNE-ISO 4121:20036). All panelists were similarly trained to examine these attributes (ISO 8586:2012).

### Texture profile analysis

Texture profile analysis (TPA) was carried out using a TA.XT2i Texture Analyser (Stable Microsystems Ltd., Surrey, England) with a 49 N load cell at a crosshead speed of 1 mm/s and a cylindrical plunger with a flat base of 35 mm of diameter. Samples were cut in pieces of, approximately, 20 mm thick and 20 mm height, which were axially

compressed to 40% of their original height. A time of 5 s was allowed to elapse between both compression cycles. The TPA parameters determined were: hardness, adhesiveness, springiness, cohesiveness, chewiness and resilience.

### Foaming properties

Foam properties were analyzed as described in Rokka et al. (2002) with some modifications. Briefly, 30 ml of either product 6 or egg-white were whipped in a volumetric cylinder with an electric stirrer at its maximum speed. The time required to achieve a stable foam and the volume of the formed foam were measured after whipping. After 24 h, the liquid drained was recovered and whipped again.

## Results

This work describes the production of novel gel (products 1–5, Table 1) and foam textures (product 6, Table 1), with multiple applications in daily and haute cuisine, through the enzymatic hydrolysis of egg proteins (from whole egg, egg yolk or egg white) using a food grade aminopeptidase.

Product 1 was obtained by hydrolysis of egg white. Before enzyme inactivation, sugar and lime–lemon liquid extract were added and, following centrifugation and separation of the supernatant, it gave a very creamy dessert with flavor and texture similar to lemon cream, without including fat or fat-replacing additives. Fig. 1 shows a photograph of this product, with an appearance similar to a custard cream (Fig. 1A), that

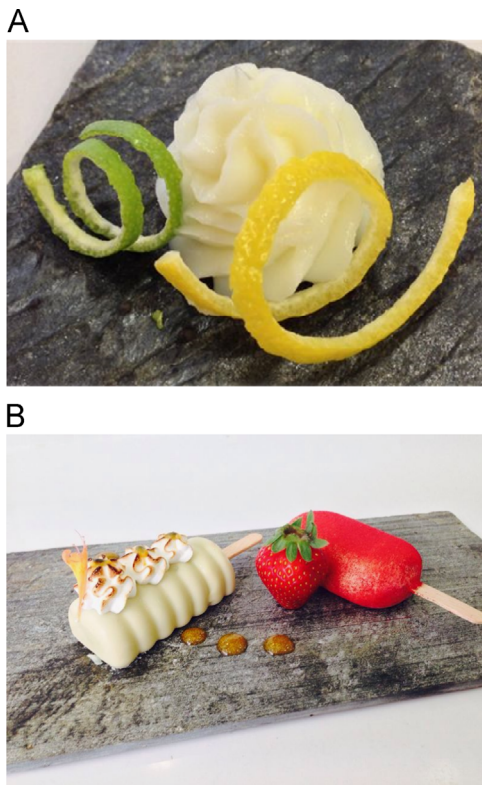


Fig. 1. (A) Cream obtained by hydrolysis and centrifugation of egg white (product 1). (B) Ice-creams produced by freezing this product.

can also be frozen to give appealing ice-creams (Fig. 1B). Product 2 was obtained by hydrolysis of egg white with the addition of salt before enzyme inactivation, giving an egg-based product with a taste and mouthfeel similar to fresh or cottage cheese, but fat-, milk protein- and lactose-free, whose moisture content and resulting texture can be regulated by controlling the extent of centrifugation or manual draining (Fig. 2 and Fig. 3B). In the case of product 3, sugar and a liquid honey extract were added before enzyme inactivation and the gel was not centrifuged, so the resulting product had a flavor and texture comparable to those of junket, a dairy dessert made by enzymatic milk coagulation, but somehow lighter and smoother (Fig. 3A).

Product 4 derived from the hydrolysis of whole egg. Before enzyme inactivation, sugar and a caramel liquid extract were added to obtain a product with flavor and texture similar to those of custard dessert (Fig. 3C), but that compared favorably with commercial custard crème, custard dessert or crème caramel. Product 5 was produced by the hydrolysis of egg yolk with the addition of sugar and a liquid cinnamon extract. This product was similar to the Spanish dessert named “tocino de cielo” but lighter and creamier than the original version (Fig. 3D).

Finally, product 6 was made by whipping the supernatant resulting from centrifugation of the egg white hydrolysate obtained following the process described to prepare product 1. As above, colorings and/or flavorings can be added before enzyme inactivation. As shown in Fig. 4A, this foam (left) was brighter and lighter in texture than the foam produced from raw egg white (right) and a smoother mouthfeel could be perceived after its tasting. This supernatant can also be



Fig. 2. Fresh cheese-like product obtained by hydrolysis and centrifugation of egg white (product 2).



lyophilized and, as result, brilliant flakes or crystals are produced (Fig. 4B).

All the above mentioned egg-derived products may be consumed, fresh or lyophilized, directly or as manageable ingredients in the preparation of sweet or/and salty recipes, allowing innovative dishes. Sensory evaluation scores are shown in Table 2. The attributes investigated were appearance, texture and taste, all essential parameters to meet consumers' expectations. The mean scores for all attributes were well above the midpoint in the 5-point hedonic scale and the mode values indicated that scores above 3 predominated in all cases. It must be emphasized that product 1 obtained scores very close to 4 (very good) for the three attributes, followed by product 6, with high scores as well. It should be noted that a high concentration of intermediate and low molecular weight hydrophobic peptides released after hydrolysis of native proteins can cause bitterness. In this study, only one panelist detected bitterness in product 2 and another one in products 2, 3 and 5. The fact that the enzyme used seldom released bitter peptides is indeed advantageous, since it provides products with neutral taste and scent that can be combined with a wide range of flavorings.

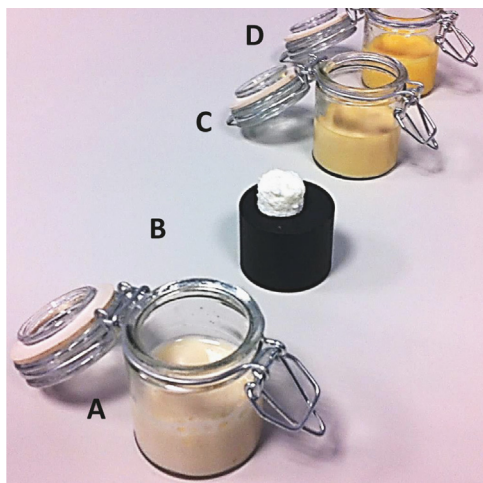


Fig. 3. (A) Junket obtained by hydrolysis of egg white (product 3). (B) Fresh cheese-like product obtained by hydrolysis and centrifugation of egg white (product 2). (C) Custard dessert obtained by hydrolysis of whole egg (product 4). (D) Custard dessert obtained by hydrolysis of egg yolk (product 5).

Sensory analysis provides an ideal opportunity to check consumers' preferences that is now generally recognized as the key for a product acceptance (Stone and Sidel, 2014). In addition to sensory assessment using trained panels and instrumental analysis (Isaksson et al., 2002), mechanical measurement methods allow the evaluation of food texture with the advantage of offering objective results, comparable among different batches or laboratories, at low cost (Ashton et al., 2010). In contrast to sensory analysis, mechanical measurements are little influenced by human factors, so the variation between measurements can be reduced (Casas et al., 2006). In this study, we used TPA for determining the textural properties of the egg-derived gels. During TPA, samples are compressed twice to provide an insight into how they would behave when chewed. As shown in Table 3, products 1–5, were characterized by different TPA parameters, such as hardness (force required to compress the product), springiness (how well the product physically springs back after it has been deformed during the first compression and has been allowed to

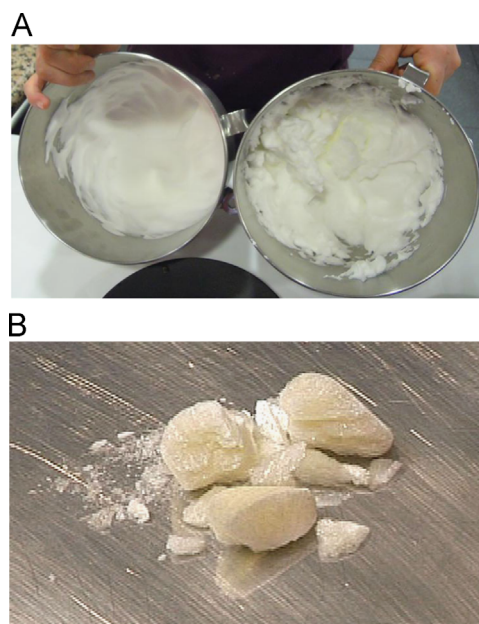


Fig. 4. (A) Foam obtained after whipping the supernatant resulting from hydrolysis and centrifugation of egg white (product 6) (left), compared with the foam obtained after whipping egg white (right). (B) When this supernatant is lyophilized, brilliant flakes or crystals are produced.

Table 2  
Sensory analysis of the egg-derived products.

	Appearance			Texture			Taste		
	Median	Mode	Range	Median	Mode	Range	Median	Mode	Range
<b>Product 1</b>	4.06	5	2–5	3.78	4	1–5	3.28	4	1–5
<b>Product 2</b>	3.92	4	2.5–5	3.56	3	2–5	2.75	3	1–4
<b>Product 3</b>	3.67	3.5	2–5	3.14	3	1–5	3.25	3	1–5
<b>Product 4</b>	3.53	4	2–4	3.18	2	2–5	3.25	3	2–5
<b>Product 5</b>	3.19	3	1.5–5	3.50	3	2–5	3.00	3	1.5–4
<b>Product 6</b>	3.78	4	2.5–4	3.83	4	2–5	3.31	3	2.5–4

Table 3  
Texture profile analysis of the gel-textured egg-derived products.

	Hardness (kg m s <sup>-2</sup> )	Springiness (adimensional)	Adhesiveness (kg m s <sup>-2</sup> )	Cohesiveness (adimensional)	Gumminess (adimensional)	Chewiness (adimensional)	Resilience (adimensional)
Product 1	1.741	0.805	-262.956	0.377	0.655	0.526	0.069
Product 2	4.288	0.776	-0.416	0.35	1.388	1.073	0.132
Product 3	3.778	0.958	-698.912	0.466	1.762	1.688	0.025
Product 4	1.07	0.724	-105.861	0.385	0.414	0.304	0.101
Product 5	0.967	0.947	-370.728	0.773	0.773	0.731	0.023

Table 4  
Foam characteristics of product 6 (Table 1) as compared with egg white.

	Time for optimum whipping (s)	Volume after whipping (ml)	Whipping again after foam drainage	Flavour
Product 6	25	220	Yes	Neutro
Egg white	70	150	No	Salad

wait for the target wait time between strokes), cohesiveness (how well the product withstands a second deformation relative to its resistance under the first deformation), gumminess (energy required to deform the product, calculated as the product of hardness and cohesiveness), chewiness (calculated as the product of gumminess and springiness) and resilience (how well a product regains its original height).

Table 4 shows a comparison between the foam obtained from product 6 and egg white. Product 6 exhibited superior foaming properties, as judged by the observation that it required less than half the time for whipping and it produced a higher foam volume, so it had a higher foaming capacity. Moreover, it was possible to re-stabilize this foam by whipping, even a long time after the original foam had collapsed, which is not possible in the case of egg white. The flavour slightly salted

## Discussion

It is well known that enzymatic hydrolysis of proteins changes food properties such as digestibility, nutritional and sensory quality (texture and flavor) and provides health benefits due to the formation of bioactive peptides and allergen reduction. However, it is not always obvious that enzymatic hydrolysis will lead to an improvement of these properties (Tavano, 2013).

The formation of food gels, that is the transformation of a fluid in a solid through the formation of a macromolecular network, requires native proteins to unfold and re-associate through covalent and non-covalent interactions, which traditionally has been achieved through the application of heat (Totosa et al., 2002). In this respect, the factors influencing the gelling ability of proteins can be divided between those which determine the formation of the gel and those which affect its physical properties. Typically, enzymatic hydrolysis impairs the gelling capacity of proteins because it decreases

their size. Thus, hydrolysis of collagen with papain, which gives rise to intermediate size peptides, produces a jelly with enough firmness and viscosity, while a most exhaustive hydrolysis with Neutrase does not allow gel formation (Damrongsakkul et al., 2008).

On the other hand, the basic properties that make protein a good foaming agent are its ability to quickly adsorb to the air-water interface during whipping; to adopt a rapid conformational change at the interface; and to form a cohesive viscoelastic film through intermolecular interactions (Foegeding et al., 2006). All these positively contribute to the formation and stability of foam containing foods. In regards to the foaming properties, papain (Chen and Chi, 2012) and trypsin (Chen et al., 2012) have recently been used to produce egg white hydrolysates with improved foaming capacity and stability as compared to the original egg white.

Therefore, the length and characteristics of the peptides formed, in terms of their constituent amino acids, the presence of polar and ionizable groups and their hydrophobicity determine the resulting functional properties; which depend, not only the degree of hydrolysis, but also on the specificity of the enzyme and protein used as a substrate (Klompong et al., 2007). In our study, the use of an aminopeptidase to hydrolyze egg proteins yielded novel gels with various textures – from smooth and creamy to rigid –, depending on the starter material and the process conditions, and light foams with a high foaming capacity and ability to re-incorporate air once collapsed.

The products also exhibited a neutral, not-bitter flavor, very convenient for mixing with different flavorings in sugar or salty preparations. Bitter flavor is common in hydrolyzed protein products. It depends on the peptide size, sequence and, mainly, hydrophobicity of the constituent amino acids (the presence of valine, leucine, isoleucine, phenylalanine, tyrosine and tryptophan often causes bitter taste). Unlike the use of other proteases, exopeptidases, that mainly catalyze the cleavage of the terminal peptide bonds, are known for their de-bittering activity (Raksakulthai and Haard, 2003).

The egg-derived products developed in this work are cheap and contain very few additives in their composition. Moreover, the elaboration process is simple, fast and allows an optimum use, not only of the whole egg, but also of its fractions, yolk and white, with the absence of waste or by-products. Therefore, it provides the means to improve the techno-functional properties of the egg as an ingredient and to expand its use in

new recipes, and in the development of new food products. In fact, one of the aims of this work was to diversify and provide alternatives to dairy foods for people sensitive to these compounds. All products are lactose and milk protein-free, while they have an appearance, texture taste and flavor similar to dairy products.

On the other hand, the egg-derived products obtained in this study are a high quality protein source, either low in fat or, in the case of those derived from egg white, fat-free. It is known that protein hydrolysates, as opposed to intact proteins, are more easily digested and absorbed in the gut, which results in a greater amino acid availability and muscle protein synthetic response (Koopman et al., 2009). Therefore, these products can be considered easy to chew and digest, which makes them particularly suitable for people with chewing limitations or digestion problems, in addition to people with weight problems, including overweighted and obese. In this respect, the use in their production of non-nutritive sweeteners that raise the blood sugar slowly, as stevia, can lead to a final product also suitable for diabetic patients.

In conclusion, this study indicates that it is possible to obtain novel gel and foam textures from whole egg, egg white and yolk by enzymatic hydrolysis, providing high added-value and healthy egg products. These results are promising and it is envisaged that they will promote new investigations in the development of foods with novel textures.

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## References

- Adrià, F., Soler, J., Adrià, A., 2005. *El Bulli 1998–2002*. Ecco, London.
- Ashton, T.J., Michie, I., Johnston, I.A., 2010. A novel tensile test method to assess texture and gaping in salmon fillets. *J. Food Sci.* 75 (4), S182–S190.
- Barham, P., Skibsted, L.H., Bredie, W.L.P., Bom Frøst, M., Møller, P., Risbo., J., Snitkjær, P., Mortensen, L., 2010. Molecular gastronomy: a new emerging scientific discipline. *Chem. Rev.* 110 (4), 2313–2365.
- Blumenthal, H., 2008. *The Big Fat Duck Cookbook*. Bloomsbury, London.
- Casas, C., Martínez, O., Guillen, M.D., Pin, C., Salmeron, J., 2006. Textural properties of raw Atlantic salmon (*Salmo salar*) at three points along the fillet, determined by different methods. *Food Control* 17 (7), 511–515.
- Chen, C., Chi, Y.J., 2012. Antioxidant, ACE inhibitory activities and functional properties of egg white protein hydrolysate. *J. Food Biochem.* 36 (4), 383–394.
- Chen, C., Chi, Y.J., Zhao, M.Y., Xu, W., 2012. Influence and degree of hydrolysis on functional properties. antioxidant and ACE inhibitory activities of egg white protein hydrolysate. *Food Sci. Biotechnol.* 21 (1), 27–34.
- Damrongsakkul, S., Ratanathampan, K., Komolpis, K., Tanthapanichakoon, W., 2008. Enzymatic hydrolysis of rawhide using papain and Neutrase. *J. Ind. Eng. Chem.* 14 (2), 202–206.
- Foegeding, E.A., Davis, J.P., 2011. Food protein functionality: a comprehensive approach. *Food Hydrocoll.* 25 (8), 1853–1864.
- Foegeding, E.A., Luck, P.J., Davis, J.P., 2006. Factors determining the physical properties of protein foams. *Food Hydrocoll.* 20, 284–292.
- García-Segovia, P., Barreto-Palacios, V., Iborra-Bernad, C., Andrés-Bello, A., González-Carrascosa, R., Bretón, J., Martínez-Monzó, J., 2012. Improvement of a culinary recipe by applying sensory analysis: design of the new tarte tatin. *Int. J. Gastron. Food Sci.* 1 (1), 54–60.
- Isaksson, T., Swensen, L.P., Taylor, R.G., Fjaera, S.O., Skjervold, P.O., 2002. Non-destructive texture analysis of farmed Atlantic salmon using visual/near-infrared reflectance spectroscopy. *J. Sci. Food Agric.* 82 (1), 53–60.
- Klompong, V., Benjakul, S., Kantachote, D., Shahidi, F., 2007. Antioxidative activity and functional properties of protein hydrolysate of yellow strip trevally (*Selaroides leptolepis*) as influenced by the degree of hydrolysis and enzyme type. *Food Chem.* 102 (4), 1317–1327.
- Koopman, R., Crombach, N., Gijsen, A.P., Walrand, S., Fauquant, J., Kies, A.K., Lemosquet, S., Saris, W.H., Boirie, Y., van Loon, L.J., 2009. Ingestion of a protein hydrolysate is accompanied by an accelerated in vivo digestion and absorption rate when compared with its intact protein. *Am. J. Clin. Nutr.* 90 (1), 106–115.
- Mine, Y., 1995. Recent advances in the understanding of egg white protein functionality. *Trends Food Sci. Technol.* 6 (7), 225–232.
- Raksakulthai, R., Haard, N.F., 2003. Exopeptidases and their application to reduce bitterness in food: a review. *Crit. Rev. Food Sci. Nutr.* 43 (4), 401–445.
- Rokka, T., Ale'n, K., Valaja, J., Ryhänen, E.-L., 2002. The effect of a *Camelina sativa* enriched diet on the composition and sensory quality of hen eggs. *Food Res. Int.* 35, 253–256.
- Stone, H., Sidel, J., 2012. Introduction to Sensory Evaluation. In *Sensory Evaluation Practices* (3rd Edition). Food Science and Technology collection. pp 1–19. Academic Press, ISBN: 978-0123820860.
- Tavano, O.L., 2013. Protein hydrolysis using proteases: an important tool for biotechnology. *J. Mol. Catal. B: Enzym.* 90, 1–11.
- Temussi, P.A., 2012. The good taste of peptides. *J. Pept. Sci.* 18, 73–82.
- Totosaus, A., Montejano, J.G., Salazar, J.A., Guerrero, I., 2002. A review of physical and chemical protein-gel induction. *Int. J. Food Sci. Technol.* 37, 589–601.