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# Scientific Paper

# Egg yolk fractions as basic ingredient in the development of new snack products

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#### **Abstract**

In this work, a new type of snack has been developed. Egg yolk and its fractions (plasma and granules) have been employed as main ingredients for the development of three base formulations. These products were characterized by means of colour, microstructure, textural and sensorial analyses. In addition, the base recipes were interpreted by a trained chef in order to give a culinary point of view. Egg yolk derivatives proved to be a key ingredient for the development of these snacks. Additionally, yolk derivatives also enhanced the organoleptic characteristics of the snacks. Sensory evaluation results showed that, in general, products were favourably assessed. In addition to their low fat (and specifically cholesterol) content and, hence, their dietetic value, granule snacks obtained the highest score in the sensory evaluation.

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Keywords: Egg yolk; Egg yolk fractions; Snacks; Texture; Sensory evaluation

# Introduction

Egg industry has seen a considerable evolution in the last 50 years, indeed, in parallel with changes in egg-processing technology, there has been a continuing growth of further processed egg products. Specifically, nowadays, approximately 30% of the total consumption of eggs is in the form of further processed eggs (Froning, 2008).

As one of the most versatile product, and on account of its multifunctional properties, egg yolk is widely used, not only in the food industry, but also in the biotechnological field (Laca et al., 2015). Egg yolk is an oil-in-water emulsion containing 52% dry matter, about 65% of which is fat, 31% proteins and the remaining 4% carbohydrates, vitamins and minerals (Guilmineau et al., 2005). In natural conditions, yolk is constituted by a continuous aqueous phase referred to as plasma, and insoluble denser structures

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(0.3–2 µm) referred to as granules (Guilmineau and Kulozik, 2006). Thus, yolk can be easily separated at an industrial scale into this two fractions by centrifugation (the supernatant represents the plasma and the pellet is made up of granules) (Laca et al., 2014). Plasma contains LDL and livetins, while granules are mainly constituted by high-density lipoproteins (HDL), phosvitin, and low-density lipoproteins (Anton, 2013). Granules represent about 22% of yolk dry matter, accounting for about 50% of yolk proteins and 7% of yolk lipids; whereas plasma corresponds to about 78% of yolk dry matter and it accounts for about 90% of yolk lipids and 50% of yolk proteins (Anton, 2007), hence separate usage of each egg yolk fraction in food processing could possibly offer new approaches (Strixner and Kulozik, 2013).

According to Opazo (2012), in the last two decades, the knowledge and practices promoted by the avant-garde movement in the culinary industry have led to radical innovations in high-end cuisine, even beyond the gastronomic field. In this context, international gastronomy and food science are in search for appealing ingredients, new foodstuffs and new technology and methods for food preparation (Krigas et al.,

2015). Consequently, since nowadays industry is looking for new uses of egg components, it is important to explore innovative applications taking into account the benefits of the interaction of science and gastronomy.

Thus, two different products (plasma and granules) can be simply obtained from the egg yolk fractionation and, until now, these raw materials have scarcely been employed in the culinary field (García et al., 2015; Laca et al., 2010a, 2014, 2015). So, this paper addresses the exploitation of these new egg yolk products as food ingredients in the development of a new type of snacks. Additionally, these fractions can supply to the developed products interesting characteristics regarding nutrient profile and also physical properties. The final aim is to expand the uses of egg yolk, a product used as food ingredient since a long time ago, in the gastronomic field.

Egg yolk and egg yolk fractions (plasma and granules) have been employed in this work as key ingredient for the development of a new type of snacks with added value from a nutritional and sensorial point of view. These products have been characterized by means of colour, microstructure, textural and sensorial analyses. In addition, the base recipes were interpreted by a trained chef in order to give a culinary perspective.

#### Materials and methods

#### Extraction of egg yolk and egg yolk fractions

Egg yolks were prepared from fresh eggs. The shelling of the eggs and the separation of the yolk from the albumen were performed manually. The albumen residuals were eliminated from the yolk using a blotting paper, and the removal of the vitelline membrane was achieved using tweezers. The fractionation method was conducted according to Laca et al. (2010b). The egg volk material is mixed with distilled water (1:1.5 v/v)and pH is adjusted to 7 by the addition of NaOH (1 N) (PanReac AppliChem, Barcelona, Spain). Then, diluted volk is kept overnight at 4 °C before centrifuging at 4 °C and 10,000g for 45 min to separate into plasma (supernatant) and granule (precipitate) fractions (KUBOTA 6500, Kubota Corporation, Tokyo, Japan). Egg yolk and egg yolk fractions (plasma and granules) were freeze-dried at  $-70\,^{\circ}\text{C}$  and  $0.1\,\text{mBa}$  in a Telstar Cryodos Lyophilizator (Telstar Group, Terrassa, Spain). Samples were frozen at -80 °C prior to lyophilization.

# Formulations and development of snacks

After preliminary assays, the final formulation of snacks was as follows. The snacks contained 4.5% (w/v) of egg yolk, plasma or granules, 2.5% (w/v) of carrageenan GPI 200 (Gum Products International, Ontario, Canada) and 1.5% (w/v) of locust beam gum E-415 (Innovative Cooking S.L., Madrid, Spain) in distilled water. Snacks of 20 g were prepared by mixing the corresponding quantity of ingredients, and then the mixture was blended with a Heidolph SilentCrusher Homogenizer (Heidolph Instruments, Schwabach, Germany) during 15 s at 17,500 rpm. The homogenize dough was cooked at

105 °C during 20 min in a UNB 400 heater (Memmert GmbH+Co. KG, Schwabach, Germany), with hand mixing during 3 s at 10 min of cooking. Afterwards, the mixture was gelled by cooling at room temperature. Once the product was gelled, it was taken out from the mould and it was cut into slices of 2 mm of thickness. Then, these slices were dried in a heater at 105 °C during 7 h. In addition, a sample without egg derivatives was prepared following the same steps with the same conditions in order to be employed as control product.

This elaboration procedure was slightly modified as follows by a trained chef with the aim to obtain different culinary products. The initial mixture of ingredients was seasoned with salt and garlic, and, once the products were gelled, they were dried in a food dehydrator 245 W (Lacor, Álava, Spain); finally, they were deep fried in hot olive oil. In addition, the initial mixture of ingredients was gelled in a mould in order to get edible recipients. These edible recipients were filled with a blend of beaten egg, cream, bacon and mushroom flavoured with salt, black pepper and basil, and cooked in a kitchen oven.

# Microstructure analysis

Snacks were analyzed employing scanning electron microscopy (SEM). Samples were fractured and torn with a blade; fragments were mounted on aluminium SEM stubs and coated with gold in a Sputtering Balzers SCD 004 (Optics Balzers AG, Balzers, Liechtenstein). The microscope used was a JEOL-6610LV SEM (Jeol Ltd., Tokyo, Japan).

#### Colour measurement

The colour measurements were carried out using an Ultra-Scan VIS spectrophotometer (Hunter Associates Laboratory Inc., Reston, Virginia, USA). It was standardized with a light trap and white title, and a green title was used to verify the instrument long-term performance. Analyses were carried out at least in duplicate and were conducted in specular exclusion mode; this mode includes the effects of gloss and texture, so the evaluation of colour is similar to human-eye perception. The colour was measured in terms of CIE-Lab parameters:  $L^*$  (whiteness or brightness),  $a^*$  (redness or greenness) and  $b^*$  (yellowness or blueness) (Wei et al., 2012).

# Moisture content

Moisture content was determined by gravimetry, in triplicate. Twenty grams of thick grain sea sand (PanReac Applichem, Barcelona, Spain) were weighted in a stainless steel mortar with its pestle and it was dried in a UNB 400 heater (Memmert GmbH+Co. KG Schwabach, Germany) at  $165\pm1$  °C during 1 h. The mortar was then kept in a vacuum desiccator for 30 min until its weight value was constant. Subsequently, approximately 3 g of sample were weighed into the mortar and the sea sand and snack sample were mixed with the pestle. The mortar was placed in a UNB 400 heater (Memmert GmbH+Co. KG, Schwabach, Germany) at  $105\pm1$  °C for 5 h. The mortars were then allowed to cool

in a vacuum desiccator at room temperature for 30 min before being finally weighed.

#### Thickness determination

Snack thickness was taken with a 0.01 mm Vernier Caliper Series 532 (Mitutoyo, Illinois, USA). Measurements were carried out in triplicate.

#### Microorganism count

Total plate counts of samples were determined according to the European Standard ISO 4833, 2003. PCA culture medium was supplied by Biokar Diagnostics (Allone, Francia). The snacks were prepared one day before the analyses and were packaged until their use.

#### Sensory evaluation

Following the procedure described in Formulations and development of snacks section, snacks were prepared, packaged and stored in the laboratory at room temperature until its use one day before the sensory evaluation. The procedures used in this preliminary sensory evaluation were based on UNE-ISO 6658:2008 Standards.

A semi-trained panel was a properly instructed about the methodology to be used in the organoleptic test and how to avoid physiological and environmental factors that might condition the results. The tests were carried out during the early afternoon in a room with artificial lightning, at a room temperature of 20 °C and in isolated work stations free from noise. The panel was made up of ten healthy subjects between 20 and 30 years of age without any sensorial defects. None of them took any medication. Samples were encoded and presented to the panel, and two kinds of trials were then developed, a first one concerning the acoustic sensations and a second one with regard to other sensory attributes.

Three attributes can be used to define the acoustic sensations, *crispness*, *crunchiness* and *crackliness*. In this work, *crunchiness* has not been assessed as this attribute is mainly related with fresh products. Although these attributes are difficult to define (Chauvin et al., 2008), *crispness* can be defined as the sound intensity that is produced by the product when it is bite with incisors one time maintaining the lips open (Seymour and Hamann, 1988; Dacremont, 1995); whereas

crackliness is the sound intensity produced by the product when it is bite, without being grinded, between 3 and 4 times with molars (Vickers, 1981; Dacremont, 1995). Hence, the evaluation of these attributes is very interesting as the product sound is different when it is entire and dry (crispness) and when it is fragmented and insalivated (crackliness). In the first trial, panellists evaluated bite sounds and chew sounds in terms of crispness and crackliness, additionally a general assessment of sound was also included in this trial. In this evaluation, panel was provide with a reference snack (a commercial standard); the value of crispness and crackliness of the standard was settled as 3 in a 1–5 scale (value 1 reflects low intensity and value 5 reflects high intensity respect to the reference snack).

Secondly, panellists scored colour, flavour, odour and hardness attributes for each product, and the products were also evaluated globally in a 1 (totally unpleasant) – 5 (totally pleasant) scale. Final scores were averaged from all of the obtained data.

#### Statistical analysis

Data were analyzed running an analysis of variance (ANOVA) with a confidence level of 95%. Previously it was checked that the data came from a normal distribution and that their variance have the same value (Levene's test). The statistical software employed was Statgraphics Centurion XVI.

## Results and discussion

General characteristics and nutritional profile

As can be seen in Fig. 1, the control snack (elaborated only with the texturing agents and without adding egg yolk) is thinner and more brittle compared with snacks developed employing egg yolk derivatives.

The average thickness value of snacks formulated employing egg derivatives as ingredients were  $1.0 \pm 0.2$  mm (similar to the value showed by the commercial standard employed as reference in sensory evaluation). Since each slide of snack weighted  $0.5 \pm 0.1$  g, then the weight per unit area is approximately  $1 \text{ kg/m}^2$ , and the density around  $1.05 \text{ g/cm}^3$ .

Table 1 shows the composition and nutritional value of the developed base snacks. Composition values were calculated according to data reported by Laca et al. (2014, 2015) and the

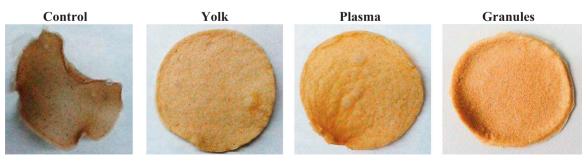


Fig. 1. Snacks developed with egg yolk derivatives and control (without egg, containing only carrageenan and locust beam gum).

carrageenan and the locust beam gum technical datasheets. Total energy value for each formulation was obtained from energy equivalents for available carbohydrate, fat, and protein, 4 kcal/g, 9 kcal/g, and 4 kcal/g, respectively (FAO, 2003; Komatsu et al., 2013). As the amount of texturing agents employed is always the same in all formulations, the composition and the nutritional value differences between snacks are determined by the egg ingredient employed in each product and also by the final product moisture content. As it was already mentioned in Introduction section, plasma contains the largest amount of egg yolk lipids, whereas granules are constituted mainly by proteins. It should be pointed out that, respect to similar commercial reference snacks, the developed products have lower caloric content, lower lipid content (and therefore lower cholesterol content) and higher protein content. Furthermore, it is remarkable that granule snack showed the lowest lipid content and the lowest total energy value too. Finally, it is also important to remark that the nutritional profile of egg snacks is better, as egg yolk contains a high proportion

Table 1 Composition and nutritional value of developed snacks per 100 g of product.

	Yolk snack	Granule snack	Plasma snack
Energy (kcal)	440.0	353.4	422.2
Dry extract (g)	98.1	98.4	97.4
Carbohydrates (g)	28.5	29.3	26.8
Proteins (g)	14.0	26.2	9.9
Total fat (g)	30.0	14.6	30.6

of n-6 PUFA and its proteins are highly digestible, containing the most important essential amino acids; additionally, eggs also supply various minerals and contain major vitamins (Surai and Sparks, 2001).

#### Microstructure

In Fig. 2 the microphotographs of snacks are shown. Regarding the control snack (without egg derivatives), SEM observation revealed that the microstructure was quite homogeneous, although particle agglomerates can be observed. In this context, it is important to remark that the synergistic interaction of carrageenan with locust bean gum is determined by the mixture ratio of both hydrocolloids (Santacruz et al., 2002), in this case the ratio was 3:1. Thus, when the egg yolk derivatives are added to the formulation, then the interaction between all the compounds generates a new microstructure. In fact, the firmer structure of snacks developed with egg yolk and its fractions is probably originated by this interaction.

Granule snacks exhibited a coarse very homogeneous structure, whereas in yolk and plasma snacks the microstructure was more heterogeneous with the surface covered by small aggregates that are possible originated by fat accumulation.

#### Colour

As it is shown in Table 2, and as it was expected, control snack exhibited the lowest values of all parameters in

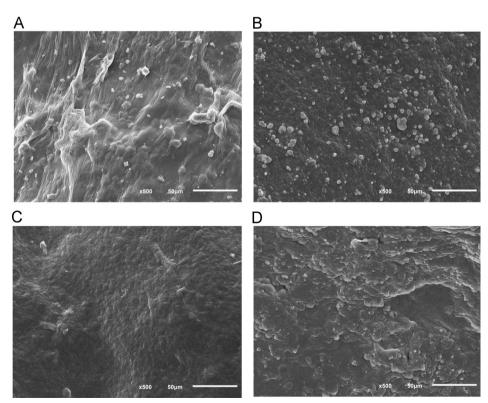


Fig. 2. Scanning electron micrographs (SEM) of snacks: (A) control snack, (B) yolk snack, (C) granule snack and (D) plasma snack ( $500 \times \text{magnification}$ ) (bar:50  $\mu \text{m}$ ).

Table 2 Values of  $L^*$ ,  $a^*$  and  $b^*$  colour parameters for snacks. Average values  $\pm$  SD are reported.

	$L^*$	a*	<i>b</i> *
Control snack	$40.2 \pm 0.3$	$-0.6 \pm 0.1$	$4.4 \pm 0.6$
Yolk snack	$54.1 \pm 4.4$	$6.2 \pm 1.0$	$23.9 \pm 2.5$
Granule snack	$55.3 \pm 1.6$	$4.1 \pm 1.7$	$22.1 \pm 1.7$
Plasma snack	$50.9 \pm 1.2$	$6.9 \pm 0.1$	$22.9 \pm 0.6$

Table 3 Sound evaluation for *crispness* and *crackliness* (scale 1–5, reference product value: 3) and general assessment of sound (1–5 scale). Average values  $\pm$  SD are reported.

	Yolk snack	Granule snack	Plasma snack	Reference
Crispness	$1.9 \pm 0.8^{b}$	2.9 ± 1.2 <sup>a</sup>	$1.7 \pm 0.7^{b}$	3.0
Crackliness	$1.8 \pm 0.7^{\rm b}$	$2.7 \pm 0.4^{a}$	$1.6 \pm 0.7^{b}$	3.0
Global evaluation	$3.0 \pm 0.5^{\mathrm{a}}$	$3.9 \pm 0.5^{b}$	$2.3 \pm 1.0^{\circ}$	$4.6 \pm 0.5^{b}$
of sound				

Values with the same superscript do not differ significantly from each other (confidence level: 95%).

comparison with the other samples. This is due to the fact that the use of egg yolk derivatives as ingredients in snack development supplies pigments (mainly carotenoids) to the final products.

Plasma snack showed the higher value of  $a^*$  (redness) and yolk snack showed the higher value of  $b^*$  (yellowness) parameters, respectively, whereas granule snack presented the higher  $L^*$  (whiteness) value. These higher values of redness and yellowness in yolk and plasma snacks are determined by the egg yolk liposoluble pigments (granule total lipid content is much lower than in case of yolk and plasma).

#### Total mesophilic count

In all samples, the total mesophilic count was below  $10^2$  cfu/g, value that remarks the correct and suitable conditions during the snack development process, as it is much lower than the typical values of total plate count of other foodstuffs such as, for example, frozen milk-based products ( $10^5$  cfu/g) (Directive 92/46/EEC).

# Sensory evaluation

Results of sensory evaluation are shown in Tables 3 and 4. As it has been reported previously by other authors (Vickers, 1985; Spence, 2015), what we hear while eating plays an important role in our perception of the textural properties of food. Indeed, the sounds that are generated while biting into or chewing food provide a rich source of information about the textural properties of that which is being consumed (Spence, 2015). In case of dry products (like the snacks developed in

Table 4 Moisture content and general assessment of sound (1–5 scale) of developed snacks and reference product. Average values  $\pm$  SD are reported.

	Moisture (%)	Global evaluation of sound
Yolk snack	$1.9 \pm 0.4$	$3.0 \pm 0.5$
Granule snack	$1.6 \pm 0.1$	$3.9 \pm 0.5$
Plasma snack	$2.6 \pm 0.4$	$2.3 \pm 1.0$
Reference snack	$3.6 \pm 0.3$	$4.6 \pm 0.5$

Table 5 Sensorial evaluation of selected attributes and global evaluation of these attributes (scale 1–5). Average values  $\pm$  SD are reported.

	Yolk snack	Granule snack	Plasma snack
Colour	$2.7 \pm 0.9^{a}$	$2.3 \pm 0.7^{a}$	$3.1 \pm 1.3^{a}$
Flavour	$2.2 \pm 0.8^{a}$	$2.6 \pm 0.9^{a}$	$2.7 \pm 0.9^{a}$
Odour	$2.5 \pm 0.7^{a}$	$2.6 \pm 0.7^{a}$	$3.1 \pm 0.6^{a}$
Hardness	$2.7 + 0.7^{a}$	$3.6 \pm 0.9^{b}$	$2.4 + 0.9^{a}$
General evaluation	$2.5 \pm 1.1^{a}$	$2.8 \pm 1.2^{a}$	$2.6 \pm 1.0^{a}$

Values with the same superscript do not differ significantly from each other (confidence level: 95%).

this work), the sound come from the rupture of void cells and the air release.

As can be seen in Table 3, granule snack obtained the highest score in *crispness* and *crackliness*, additionally the global evaluation value of this sample was not significantly different from the reference product. Values obtained for yolk and plasma snacks did not differ significantly between them, whereas the scores of these samples were significantly different from those of granule snack. As it is shown in Table 4, a link between sound score and moisture content can be observed, in fact, the more the amount of water content the lower the score that was obtained in the global evaluation of sound. Hence, it is important to obtain sufficiently dried snacks in order to contribute positively to its organoleptic properties.

Regarding the evaluation of the other different attributes and, as can be seen in Table 5, plasma snack obtained the best score in colour, flavour and odour, whereas in hardness it was evaluated with the lowest score. Furthermore it should be pointed out that granule snack hardness was the only attribute that differed significantly from the attributes of the other samples. In all cases, general evaluation of the products was near to 3 (the medium value of the scale), additionally there were not significant differences between the samples in this evaluation. Thus, the developed products, mainly in the case of granule snacks, have shown to be an interesting base product for the development of different suitable applications.

## **Applications**

The base snacks developed in this work are shown in Fig. 1. The advantages of this snack are mainly that it is a nutritive crunchy product with an appealing flavour. In addition, its low water content (less than 3%) favours a long self-life. The

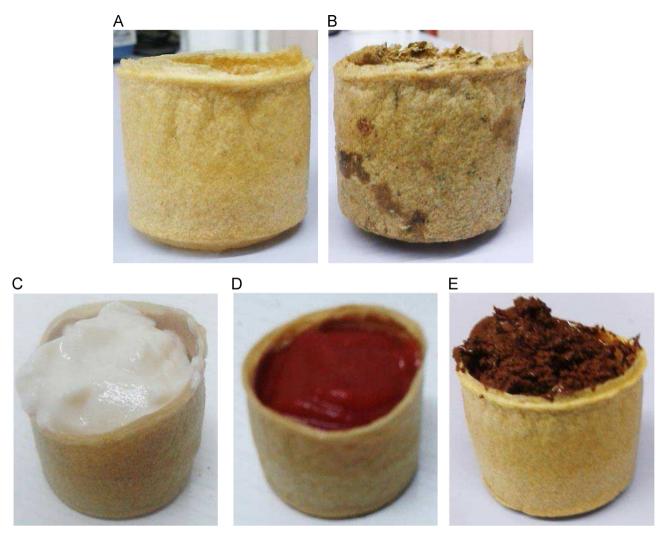


Fig. 3. Examples of applications of granule recipients. Recipients developed including in the recipe different spices (A:0.25% w/v vanilla and B:0.75% w/v oregano) and containing different foodstuff (C:yogurt, D:tomato sauce and E:chocolate).



Fig. 4. Tartlets with mushrooms and basil developed employing granule recipients.



Fig. 5. Granule snacks seasoned with salt and garlic and fried in olive oil.

formulation and also the elaboration process of these products can be modified with the aim to be employed in different applications. Indeed, the base formulation can be modified including different additives (colourants and/or flavouring), whereas the elaboration procedure can be adapted in order to obtain, for example, edible recipients with the same advantages

of those showed by the snacks (Figs. 3 and 4). Moreover, snacks and edible recipients can be used by chefs for the development of different culinary creations such as those showed in Figs. 4 and 5.

#### **Conclusions**

Egg yolk derivatives have shown to be a key ingredient for the development of these new snack products. Furthermore, according to data obtained in this work, it has been proved that the use of yolk derivatives improved not only the organoleptic characteristics of the snacks, but also its mechanical properties and nutritional profile. In fact, although in sensory evaluation, all base products were favourably assessed, it is important to remark that granule snacks obtained the highest score. Additionally, some potential applications of these base products, such as edible recipients or fried snacks, have been successfully assayed as examples.

To sum up, it should be pointed out that, after years of utilization in food field, this study expands the use of yolk, showing this raw material still as a suitable ingredient for culinary experimentation.

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