Effects of combined HHP and heat treatment on viscosity attributes and microbiological condition of liquid egg yolk

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ABSTRACT

Minimal processing technologies, like High Hydrostatic Pressure (HHP), heat treatments at low temperatures have an increasing role in food industry. Eggs are considered as functional foods, but for high retention of biological active compounds adequate minimal processing technologies are needed during preservation procedure. In our study, liquid egg yolk (LEY) was examined to meet consumer’s expectations.

Combinations of pasteurization (57–63 °C, 5–7 min) and HHP (350–400 MPa, 5 min) were used to provide microbiological stability of LEY. After these treatments samples were examined for mesophyll aerobes and Enterobacteriaceae cell counts (using Nutrient agar an incubation of 30 °C, 48 h) and viscosity attributes (Anton Paar MCR 92).

Our results show that microbiological stability is significantly influenced by the different parameters of heat treatments and HHP. Heat treatment effected at least 3 orders of magnitude decrease in cell count. Viscosity attributes point out that higher pressure of HHP have a stronger effect on viscosity than the temperature of pasteurization.

The results point out a great opportunity for industrial use of minimal processing technologies for LEY. Microbiological safety is strongly influenced by the order of treatments, but viscosity may be independent from the order of the treatments.

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INTRODUCTION

The poultry industry is one of the fastest growing animal industries globally. The world egg production reached 68.26 Mt in 2013, with an increase of 94.6% from 35.07 Mt in 1990 (Anonymous, n.d.). Eggs are one of the most nutrient dense foods (McNamara & Thesmar, 2005). Hen’s eggs have been reported to be a nutrient-dense food with high-quality protein, which is present in both the egg white and the yolk. Regarding micronutrient composition, one large egg (63–73 g) contains 186 mg cholesterol, 126 mg choline, 0.2 mg riboflavin, 0.5 mg vitamin B₁₂, 24 mg folate, 0.1 mg vitamin B₆, 41 IU vitamin D, 270 IU vitamin A, 0.5 mg vitamin E, 99 mg phosphorus, and 0.9 mg iron. These nutrients are distributed between the egg white and the yolk (McNamara, 2013; Benahmed et al., 2017; Banovic et al., 2018).

Egg yolk is well known as a natural oil-in-water emulsion. Because of its multifunctional properties egg yolk is extensively used in the food, medical, pharmaceutical, and cosmetics industries (Laca et al., 2015; Chalamaiah et al., 2017). Egg yolk is made of approximately 52% dry matter, about 65% of which is fat, 31% proteins, and the remaining 4% carbohydrates, vitamins, and minerals (Guilmineau et al., 2005).

Emulsions are metastable systems that tend to destabilize through a number of mechanisms (e.g., creaming, coalescence, flocculation). In order to increase emulsion stability, which is a key factor for egg yolk’s commercial applications, pH modification or heat treatment stabilization must be used (Bengoechea et al., 2010). Hen egg yolk is an ideal example of natural supramolecular assemblies of lipids and proteins with different organization levels. These assemblies are mainly due to interactions between proteins and phospholipids, and these interactions are essential in understanding and controlling the production of food made with yolk, and particularly emulsions (Anton, 2013).

Food industry prefers to use treated egg products instead of shell eggs. Eggs removed from shell are categorized in several ways, but the most often used categories are liquids, powders, or boiled eggs (Alamprese, 2017; Pelletier, 2017). The use of egg products increases the efficiency of food production and decreases the amount of food waste, while more economic producing is achieved.

In egg product industry, the microbiological safety of liquid products is mainly guaranteed by pasteurization. The United States Department of Agriculture (USDA) requires that liquid whole egg (LWE) is at least heated at 60 °C for no less than 3.5 min, but in the United Kingdom the recommendations are to pasteurize at least at 64 °C for 2.5 min (Rossi et al., 2010; Korver & McMullen, 2017). In France, there is no statutory heat treatment; only microbiological results are determined by regulations. To achieve this, the classically used treatment is to pasteurize whole egg at from 65 to 68 °C for 2–5 min in order to ensure 5–6 decimal reductions of vegetative microorganisms and especially Salmonella and Listeria monocytogenes (Baron et al., 2010). Pasteurization temperatures used in the egg industry are limited by the sensitivity of egg proteins to heat treatment. Thus, pasteurization for 2–10 min from 60 to 68 °C modifies whole egg electrophoretic pattern by especially decreasing ovotransferrin, livetin, ovalbumin,
apovitellenin, lysozyme, and/or ovomucin band intensity (Bartlett & Hawke, 1995; Rossi et al., 2010; Lechevalier et al., 2017).

The new consumer expectations regarding sensorial attributes led the food industry to apply minimal processing technologies aiming at the preservation of bioactive compounds and extending the shelf-life of treated products (Barbosa-Cánovas et al., 2011). One of the most promising technologies is the high hydrostatic pressure (HHP). Nutritionally, it enhances the intake of dietary nutrients in human by converting the complex ones into smaller ones (McInerney et al., 2007). Applications of HHP in food industries includes reduction in spoilage of food, enhancing safety of foods, retaining freshness of food commodities, and improving the shelf life of food items without/with minimal use of preservatives (Considine et al., 2008). As compared to traditional food processing technologies, HHP has less adverse and detrimental effects on quality and nutritional characteristics of food items during processing or preservation. This novel food processing technology is mainly derived from material science as in this technique food is normally treated at >100 MPa (mega Pascal) pressure. This technique is extensively evaluated. During food processing, high pressure applied to food item is equal from all directions, conducted through items uniformly and quickly by the pressure transferring medium which is not dependent upon geometry (Oey et al., 2008; Khan et al., 2018).

The aim of our experiment was to evaluate the changes in microbiological safety and viscosity attributes of liquid egg yolk (LEY) treated with different HHP and heat treatment combinations.

MATERIAL AND METHODS

Sample preparing

Homogenized LEY was taken from the production line of Capriovus Ltd. (Szigetscép, Hungary). Samples were transferred to Szent István University, Dep. of Refrigeration and Livestock Products Technologies under refrigerated conditions (4–6 °C) directly after production. Samples (9 × 250 mL for investigation of techno functional properties and 9 × 3 × 50 mL for microbiological tests) were packaged in high border polyethylene plastic bags.

All samples were first pasteurized than HHP treated. Heat treatments were 57 °C, 10 min or 63 °C, 7 min. After heat treatment samples were cooled to room temperature in melting ice. HHP treatments were carried out in a Resato FPU 100–2,000 HHP equipment at 20 °C. The range of pressure build up was 100 MPa/min, and the unpressurization was immediate after treatment. HHP treatment was carried out for 5 min on 350 or 400 MPa. Samples were cooled down to 4 °C before measuring viscosity and microbiological load. Table 1 summarizes the different treatment parameters and sample coding used in this work.

Inspection of viscosity attributes

Viscosity attributes were investigated with an Anton Paar MCR 92 viscosimeter. The sample temperature was 15 °C and data were collected between 10 and 1,000 1/min share rate. The flow charts were analyzed by Herschel–Bulkley models using the formula:
\[ \tau = \tau_0 + K\gamma^n, \]

The analyzed constants are collected in Table 2.

**Microbiological testing**

In microbiological testing samples (3 repetitions for every sample, 50–50 mL each) were taken in sterile conditions. The storage temperature before measurement was −4 °C. After treatments samples were examined in 24 h for mesophyll aerobes and *Enterobacteriaceae* cell counts (using nutrient agar and usual incubation of 30 °C for 48 h).

**RESULTS AND DISCUSSION**

The evaluation of viscosity attributes is summarized in Table 3. The combined treatments show a difference in viscosity attributes. However, results of the fitted models are not for every sample well acceptable ($R^2 < 0.95$). Similar results are published in case of liquid egg white and liquid whole egg (Wardy et al., 2014). In case of fruits, e.g., mango, HHP and heat treatment modified viscosity as well (Liu et al., 2014). According to the literature, changes are significant almost in every food product, independent from its plant or animal origin (Bengoechea et al., 2010).

**Table 1. Applied treatment parameters of LEY and sample coding**

| Sample | Temperature of heat treatment, °C | Time of heat treatment, min | Pressure of HHP, MPa | Holding time of HHP, min |
|--------|----------------------------------|-----------------------------|----------------------|--------------------------|
| 1      | 0                                | 0                           | 0                    | 0                        |
| 2      | 0                                | 0                           | 350                  | 5                        |
| 3      | 0                                | 0                           | 400                  | 5                        |
| 4      | 57                               | 10                          | 0                    | 0                        |
| 5      | 63                               | 7                           | 0                    | 0                        |
| 6      | 57                               | 10                          | 350                  | 5                        |
| 7      | 63                               | 7                           | 350                  | 5                        |
| 8      | 57                               | 10                          | 400                  | 5                        |
| 9      | 63                               | 7                           | 400                  | 5                        |

Source: (Elgaddafi et al., 2016).

**Table 2. The nomenclature of Herschel–Bulkley model parameters**

| Constant | Nomenclature |
|----------|--------------|
| \( \gamma \) | Shear rate (1/s) |
| \( \tau \) | Yield stress (Pa) |
| \( \tau_0 \) | Yield stress (Pa) |
| \( K \) | Consistency factor (Pas) |
| \( N \) | Flow index, a power law exponent (-) |
| \( R^2 \) | Goodness of fitted model (–) |

Source: (Elgaddafi et al., 2016).
Microbiological spoilage of LEY was sufficiently decreased by combined HHP and heat treatment. *Enterobacteraceae* were not detected in the examined samples. Fig. 1 shows the mesophyll aerobe cell count.

Our results show that first heat treated, then HHP treated LEY samples had a significant lower microbial spoilage. In contrast, single HHP (sample 2, 3), or single heat treatment (sample 4, 5) has a lower effect on microbiota. Other studies point out that minimal processing treatments are sufficient for microbiological safety of egg yolk (Badr, 2006).

The HHP treatment of egg white (Toth et al., 2017) has a high influence on microbiota and rheological properties. Some studies point out that HHP has a higher impact on viscosity of liquid egg products, than heat treatment (Németh et al., 2012).

### CONCLUSIONS

Our results show that HHP and heat treatment influence significantly the viscosity attributes of LEY. The parameters of both treatments have high impact on viscosity of LEY, which has an industrial relevance as well. For reducing rheological changes higher temperature of heat treatment and lower pressure of HHP are proposed.

Microbiological tests highlighted that the order of applied HHP and heat treatment have an important role in microbiological safety of LEY. In aspect of food safety the best choice is using heat treatment at first and then HHP.
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