Original scientific paper
UDC 637.414.055

EFFECTS OF COMBINED HHP AND HEAT TREATMENT ON VISCOSITY AND MICROBIOLOGICAL SAFETY OF LIQUID EGG YOLK

Adrienn Tóth[1*], Csaba Németh[2], Ildikó Zeke[1], Péter Penksza[1], Karina Hidas[1], László Friedrich[1]

[1]Department of Refrigeration and Livestock Product's Technology, Faculty of Food Science, Szent István University, Ménesi út 43 - 45, 1118 Budapest, Hungary
[2]Capriovus Ltd., Dunasor 073/72 hrsz., 2317 Szigetcsép, Hungary

[*]e-mail: toth.adrienn@etk.szie.hu

Abstract

Minimal processing technologies, like High Hydrostatic Pressure (HHP), heat treatments on law temperatures, ultra-sonication have an increasing tendency 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 was examined to meet consumer's expectations.

Several combinations of law temperature pasteurization (57 - 63 ^{0}C, 5 - 7 min.) and High Hydrostatic Pressure (350 - 400 MPa, 5 min.) were used to provide microbiological stability of liquid egg yolk. After treatments samples were examined for mesophyll aerobes and *Enterobacteriaceae* cell counts (using Nutrient agar and usual incubation of 30 ^{0}C for 48 hours) and viscosity attributes. Viscosity attributes (measured with Anton Paar MCR 92) analyzed by Hershel-Bulkley models.

Our results show that microbiological stability is significant influenced by the different parameters of heat treatments and HHP have a strong effect 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 influence on viscosity than the temperature of pasteurization.

Our results show a great opportunity for industrial use of minimal processing technologies for liquid egg yolk. Microbiological safety is strongly influenced by the order of treatments, but viscosity is independent from order of HHP and heat treatment.

***Key words**: HHP, Minimal processing, Egg products, Liquid egg yolk.*

1. 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 [1]. Eggs are one of the most nutrient dense foods [2]. 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 contains: 186 mg cholesterol, 126 mg choline, 0.2 mg riboflavin, 0.5 mg vitamin B_{12}, 24 mg folate, 0.1 mg vitamin B_6, 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 [3 - 5].

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 because of its multifunctional properties [6, 7]. 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 [8].

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 [9]. 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 [10].

Food industry prefers to use instead of shell eggs, treated egg products. Eggs removed from shell are categorized in several ways, but the most popular is due to the product's texture. These groups are including but not limited liquids, powders, or boiled eggs [11, 12]. 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, microbiological safety of liquid products is mainly guaranteed by pasteurization. The USDA requires that liquid whole egg is at least heated at 60 ^{0}C for no less than 3.5 minutes, but in the United Kingdom the recommendations are to pasteurize at least at 64 ^{0}C for 2.5 minutes [13, 14]. In France, there is no statutory heat treatment; only microbiological results are determined by regulations. To achieve this, the treatments classically used to pasteurize whole egg vary from 65 to 68 ^{0}C for 2 - 5 minutes in order to ensure 5 to 6 decimal reductions of vegetative microorganisms and especially *Salmonella enteritidis* and *Listeria monocytogenes* [15]. Pasteurization temperatures used in the egg industry are limited by the sensitivity of egg proteins to heat treatment. Thus, pasteurization for 2 - 10 minutes from 60 to 68 ^{0}C modifies whole egg electrophoretic pattern by especially decreasing: ovotransferrin, livetin, ovalbumin, apovitellenin, lysozyme and/or ovomucin band intensity [13, 16, and 17].

That led the food industry for application of minimal processing technologies aiming the preservation of bioactive compounds and extending the shelf-life of treated products [18]. 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 [19]. 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 [20]. 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 [21, 22].

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.

2. Material and Methods

2.1 Sample preparation

Homogenized liquid egg yolk (LEY) was taken from the production line of Capriovus Ltd. (Szigetcsép, Hungary). Samples were transferred to Szent István University, Dep. of Refrigeration and Livestock Products Technologies under refrigerated conditions (4 - 6 ^{0}C) directly after production. Samples were packaged in high border polyethylene plastic bags.

All samples were first pasteurized than HHP treated. Temperatures of heat treatments were 57 and 63 ^{0}C, the time of heat penetration was 10 and 7 min. respectively. After heat treatment samples were cooled to room temperature in melting ice. HHP treatments were carried out in a Resato FPU 100 - 2000 HHP equipment in room temperature. The range of pressure build up was 100 MPa/min, and the unpressurization was immediate after treatment. Samples was cooled down to 4 ^{0}C before measuring viscosity and microbiological load. Table 1 summarizes the different treatment parameters and sample coding used in this work.

Table 1. Applied treatment parameters of LWE and sample coding

Sample	Temperature of heat treatment, ^{0}C	Time of heat treatment, s	Pressure of HHP, MPa	Holding time of HHP, s
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

2.2 Inspection of viscosity attributes

Viscosity attributes were investigated with an Anton Paar MCR 82 viscosimeter. The sample temperature was 15 ^{0}C and data were collected between 10 and 1000 1/min. share rate. The flow charts were analysed by Herschel-Bukley models. The analysed constants are collected in Table 2.

Table 2. The nomenclature of Herschel-Bukley model parameters

Constant	Nomenclature
a	Empirical parameter 1
b	Empirical parameter 2
p	Fluid behaviour index
R^2	Goodness of fitted model

source: [23]

2.3 Microbiological testing

In microbiological testing samples were taken in sterile conditions. The storage temperature before measurement was 4 - 6 ^{0}C. After treatments samples were examined in 24 hours for mesophyll aerobes and *Enterobacteriaceae* cell counts (using Nutrient agar and usual incubation of 30 ^{0}C for 48 hours).

3. Results and Discussion

The evaluation of viscosity attributes are summarized in Table 3. The combined treatments show a difference in viscosity attributes. However, results of the fitted models are not in every case well acceptable (R^2 values). Similar results are published in case of liquid egg white and liquid whole egg [24]. In case of fruits e.g. mango HHP and heat treatment modified viscosity as well [25]. According to the literatures, changes are significant almost in every food product, independent from its plant, or animal origin [26].

Table 3 summarizes the results of Herschel-Bukley models. Results of statistical models are well fitting to measured results (R^2 values). Fluid behavior index (p) is highly influenced by HHP and heat treatment. The higher pressure range was applied, the lower p values were calculated, which shows an increasing pseudoplastic behavior of liquid egg yolk.

Microbiological spoilage of LEY was sufficient decreased by combined HHP and heat treatment. *Enterobacteraceae* were not detected in examined samples. Figure 1 shows the mesophyll aerobe cell count.

Our results show that first heat treated, than HHP treated LEY samples had a significant lower microbial spoilage. In contrast, single HHP, or single heat treatment has a lower effect on microbiota. Other studies point out, that minimal processing treatments are sufficient for microbiological safety of egg yolk [27].

The HHP treatment of egg white [28] 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 [29].

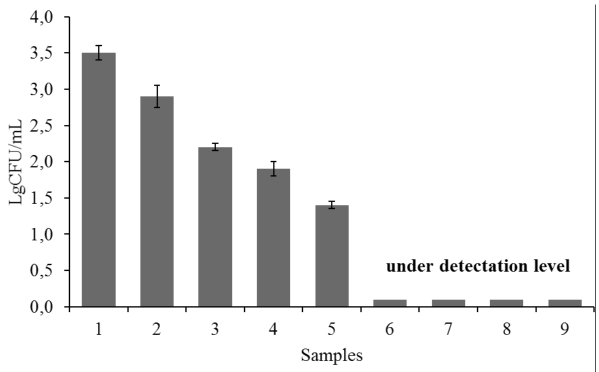

Figure 1. Mesophyll aerobe cell count of HHP and heat treated LEY

4. Conclusions

- Our results show that HHP and heat treatment influence significantly the viscosity attributes of liquid egg yolk. The parameters of both treatment 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 first heat treatment and then HHP.

Acknowledgement

Our research is supported by the ÚNKP-17-3 New National Excellence Program of The Ministry of Human Capacities and by EFOP-3.6.3.- VEKOP-16-2017-00005. We are also very thankful for the kindly cooperation of Capriovus Ltd. and Szent István U-niversity, Dep. of Refrigeration and Livestock Product's Technologies.

Table 3. Calculated constants of Herschel-Bukley models of treated LEY samples

Constant	Sample								
	1	2	3	4	5	6	7	8	9
a	-0.72	-1.09	-3.4	-5.64	-1.06	-4.35	3.64	0.26	-1.22
b	0.92	1.35	13.24	23.17	16.41	26.46	47.27	41	31.6
p	0.85	0.81	0.53	0.46	0.5	0.44	0.39	0.4	0.39
R^2	0.99	0.96	0.94	0.93	0.94	0.95	0.96	0.97	0.96

5. References

[1] World Agriculture. *Towards 2015/2030 - An FAO perspective*. URL: http://www.fao.org/docrep/005/y4252e/y4252e07.htm. Accessed 09 Jun 2017.

[2] McNamara J. D., and Thesmar S. H. (2005). Eggs. In: Caballero B. (Ed.), Encyclopedia of Human Nutrition (2nd Ed.), Elsevier, Netherlands, pp. 86-92.

[3] McNamara J. D., and Thesmar S. H. (2013). Eggs. In: Caballero B. (Ed.), Encyclopedia of Human Nutrition (3rd Ed.), Academic Press, Cambridge, USA, pp. 132-138.

[4] Benahmed F., Wang H., Beaubrun J. J., Gopinath G. R., Cheng C. M., Hanes D. E., Hammack T. S., Rasmussen M., Davidson M. K. (2017). *Detection of Salmonella enterica subsp. enterica serovar cubana from naturally contaminated chick feed*. Journal of Food Protection, 80, 11, pp. 1815-1820.

[5] Banovic M., Arvola A., Pennanen K., Duta D. E., Brückner-Gühmann M., Lähteenmäki L., Grunert K. G.. (2018). *Foods with increased protein content: A qualitative study on European consumer preferences and perceptions*. Appetite, 125, pp. 233-243.

[6] Laca A., Paredes B., Rendueles M., and Díaz M. (2015). *Egg yolk plasma: Separation, characteristics and future prospects*. LWT - Food Science and Technology, 62, 1, Part 1, pp. 7-10.

[7] Chalamaiah M., Esparza Y., Temelli F., and Wu J. (2017). *Physicochemical and functional properties of livetins fraction from hen egg yolk*. Food Bioscience, 18, pp. 38-45.

[8] Guilmineau F., Krause I., and Kulozik U. (2005). *Efficient analysis of egg yolk proteins and their thermal sensitivity using sodium dodecyl sulfate polyacrylamide gel electrophoresis under reducing and nonreducing conditions*. Journal of Agricultural and Food Chemistry, 53, 24, pp. 9329-9336.

[9] Bengoechea C., Romero A., Aguilar M. J., Cordobés F., and Guerrero A. (2010). *Temperature and pH as factors influencing droplet size distribution and linear viscoelasticity of O/W emulsions stabilised by soy and gluten proteins*. Food Hydrocolloids, 24, 8, pp. 783-791.

[10] Anton M. (2013). *Egg yolk: Structures, functionalities and processes*. Journal of the Science of Food and Agriculture, 93, 12, pp. 2871-2880.

[11] Alamprese C. (2017). *Chapter 24 - The Use of Egg and Egg Products in Pasta Production*. In: Hester Y. P. (Ed.), Egg Innovations and Strategies for Improvements, Academic Press, San Diego, USA, pp. 251-259.

[12] Pelletier N. (2017). *Life cycle assessment of Canadian egg products, with differentiation by hen housing system type*. Journal of Cleaner Production, 152, pp. 167-180.

[13] Rossi M., Casiraghi E., Primavesi L., Pompei C., and Hidalgo A. (2010). *Functional properties of pasteurised liquid whole egg products as affected by the hygienic quality of the raw eggs*. LWT - Food Science and Technology, 43, 3, pp. 436-441.

[14] Korver D., and McMullen L. (2017). Chapter 4 - Egg Production Systems and Salmonella in Canada. In: Ricke S., Gast R. (Eds.), Producing Safe Eggs, Academic Press, San Diego, USA, pp. 59-69.

[15] Baron F., Jan S., and Jeantet R. (2010). *Microbiological quality of egg products* (in French). Sciences et technologie de l'œuf: De l'œuf aux ovoproduits, pp. 321-349.

[16] Bartlett M. F., and Hawke E. A. (1995). *Heat Resistance of Listeria monocytogenes Scott A and HAL 957E1 in Various Liquid Egg Products*. Journal of Food Protection, 58, 11, pp. 1211-1214.

[17] Lechevalier V., Guérin-Dubiard C., Anton M., Beaumal V., Briand D. E., Gillard A., Le Gouar Y., Musikaphun N., Tanguy G., Pasco M., Dupont D., Nau F. (2017). *Pasteurisation of liquid whole egg: Optimal heat treatments in relation to its functional, nutritional and allergenic properties*. Journal of Food Engineering, 195, pp. 137-149.

[18] Barbosa-Cánovas V. G., Ghani A., Juliano P., and Knoerzer K. (2011). *Introduction to Innovative Food Processing Technologies: Background, Advantages, Issues, and Need for Multiphysics Modeling*. Innovative Food Processing Technologies: Advances in Multiphysics Simulation, pp. 3-21.

[19] McInerney K. J., Seccafien A. C., Stewart M. C., and Bird R. A. (2007). *Effects of high pressure processing on antioxidant activity, and total carotenoid content and availability, in vegetables*. Innovative Food Science and Emerging Technologies, 8, 4, pp. 543-548.

[20] Considine M. K., Kelly L. A., Fitzgerald F. G., Hill C., and Sleator D. R. (2008). *High-pressure processing - Effects on microbial food safety and food quality*. FEMS Microbiology Letters, 281, 1, pp. 1-9.

[21] Oey I., Lille M., Van L., and Hendrickx M. (2008). *Effect of high-pressure processing on colour, texture and flavour of fruit- and vegetable-based food products: A review*. Trends in Food Science and Technology, 19, 6, pp. 320-328.

[22] Khan K. M., Ahmad K., Hassan S., Imran M., Ahmad N., and Xu C. (2018). *Effect of novel technologies on polyphenols during food processing*. Innovative Food Science & Emerging Technologies, 45, pp. 361-381.

[23] Elgaddafi R., Ahmed R., and Growcock F. (2016). *Settling behavior of particles in fiber-containing Herschel Bulkley fluid*. Powder Technology, 301, pp. 782-793.

[24] Wardy W., Pujols Martínez D. K., Xu Z., No K. H., and Prinyawiwatkul W. (2014). *Viscosity changes of chitosan solution affect physico-functional properties and consumer perception of coated eggs during storage*. LWT - Food Science and Technology, 55, 1, pp. 67-73.

[25] Liu F., Li R., Wang Y., Bi X., and Liao X. (2014). *Effects of high hydrostatic pressure and high-temperature short-time on mango nectars: Changes in microorganisms, acid invertase, 5-hydroxymethylfurfural, sugars, viscosity, and cloud*. Innovative Food Science & Emerging Technologies, 22, pp. 22-30.

[26] Bengoechea C., Romero A., Aguilar M. J., Cordobés F., and Guerrero A. (2010). *Temperature and pH as factors influencing droplet size distribution and linear viscoelasticity of O/W emulsions stabilised by soy and gluten proteins*. Food Hydrocolloids, 24, 8, pp. 783-791.

[27] Badr M. H. (2006). *Effect of gamma radiation and cold storage on chemical and organoleptic properties and microbiological status of liquid egg white and yolk*. Food Chemistry, 97, 2, pp. 285-293.

[28] Toth A., Nemeth C., Horváth F., Zeke I., and Friedrich L. (2017). *Impact of HHP on microbiota and rheological properties of liquid egg white, a kinetic study*. Journal of Biotechnology, 256, Supplement, pp. 93.

[29] Németh C., Balla C., Pásztor-Huszár K., Zeke I. (2012). Effect of high pressure treatment on liquid whole egg. High Pressure Research, 32, 2, pp. 330-336.