The Improving Quality and Shelf Life of Table Eggs

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Abstract

In this review manuscript content, the necessity of high pressure processing, microbial stability, preservation and shelf life of table eggs by high pressure has been described. Rheological properties of table eggs by high pressure and high pressure effects on egg phosvitin, ovalbumin and ovotransferrin has been reported. High pressure effects on foaming properties and on color and texture properties of egg has been executed.

Keywords: *Quality, Shelf Life, Table Egg, HHP*

Introduction to Table Egg and The Necessity of High Pressure Procesing

Whole egg (WE) has excellent nutritional value, especially contains high biological value of protein as compared to any dietary protein sources and egg proteins own all covetable nutritional and functional properties. Liquid egg, homogenized as whole egg or separated into white and yolk, is used as an ingredient and/or as a colorant in a wide variety of processed food products (Tokuşoğlu,2013;ICMSF,1998).

Exclusively, liquid whole egg (LWE) contributes physicochemical characteristics to foods including coagulating, foaming, and emulsifying and gelling (Lee, Heinz & Knorr,1999; Yang & Baldwin, 1995). Due to these important functional properties, LWE can be extensively used as food ingredient and colorant of many foods, such as bakery products, meringues, meat products, chocolate, confectionary products, drinks, infant foods, dressings, noodles and snack food industry. Owing to holding a large quantity of air in the form of fine bubbles, the bubbles of beaten eggs expand in a cake mix and the albumen gives strength to the walls of the air pocket. Egg also contains other nutrients; carbohydrates, vitamins, minerals, phospholipids and other functional lipids. The phospholipids containing yolk confers stability on emulsion of oils and water and utilized in the making mayonnaise, egg phospholipids also used as ingredient of dough and ice cream mix (Tokuşoğlu,2013; ICMSF,1998; Dawson & Martinez-Dawson, 1998; Ahmeda et.al.*,2003).*

The outer eggshell is made almost entirely of calcium carbonate ($CaCO_3$) and is covered with as many as 17,000 tiny pores. It is a semipermeable membrane, that allows air and moisture to pass through its pores. Chalaza parts in opposite directions of egg serve to keep the

yolk centered. and it is stated that the more prominent the chalazae, the fresher the egg (Figure 1).

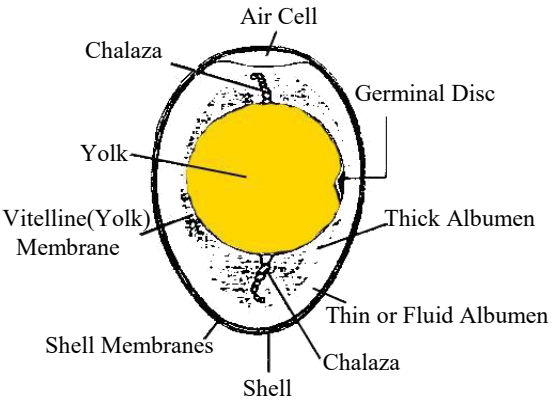

***Figure 1**. TheParts of Egg*

Regrettably, egg and egg products are also responsible for a large number of foodborne illnesses owing to its anatomy and the water, protein and lipid are major components of liquid egg that can support microbial growth at inappropriate storage. Microbial contamination of eggs as well as its economic implications for the poultry industry have been reported (EFSA,2011; Bruce & Drysdal, 1994; Wong & Kitts, 2003).

Traditional thermal treatments used to pasteurize LWE (e.g., 60 °C for 3.5 min in the USA, or 64 °C for 2.5 min in the U.K.) ensure food safety by giving 5-9 Log10 reductions of the most frequent Salmonella *serotypes (Alvarez* et.al.*,2006; Mañas* et.al.*,2003). Eventhough, some heat-resistant microorganisms can survive the above-mentioned pasteurization requirements and spoil the LWE even under refrigerated conditions (Lee* et.al.*,2001). Pasteurisation for LWE is limited to lower pasteurisation temperatures and longer holding times because of the coagulation of its proteins at higher temperatures.*

Salmonella *is the primary important problem in most cases (EFSA,2011). Recently,* Salmonella *in eggs has emerged as a primary concern for public health agencies in Europe and the United States (CDC, 2003, 2004; Schroeder* et al.*,2005). It was reported that pasteurisation of egg products became mandatory in the US in 1966 (Cunningham, 1995) and current regulations in the US require that LWE is heated to at least 60 C for a minimum of 3-5min. The reason may be attributed to the fact that the USDA requires liquid egg to be heated at the above-mentioned temperature and duration time to achieve more than 3.0 log in colony-forming units (CFU)/mL reduction of* Salmonella *(ICMSF, 1998). It is concluded that the functional performance of egg white is impaired when heated for several minutes above 57 C (Ma* et al.*,1997). Owing to the incomplete pasteurization at lower temperatures, the foodborne outbreaks comprising* Salmonella enteridis *have resulted in eggs (Tood,1996; Tauxe,1991). The illness risk is greater when the egg is used as an food ingredient in foods rather*

than when consumed as a individual egg (Todd,2001).

The thermal pasteurization of LWE in most cases leads to protein denaturation and coagulation thence affecting the liquid egg consistency. Therefore, pasteurisation of LWE is limited to lower pasteurisation temperatures and longer holding times owing to the coagulation of its proteins at higher temperatures (Tewari et al., 1999; Tood, 1996; Cheftel, 1995; Tauxe,1991).

High pressure processing (HPP) is an industrially tested technology that offers a more natural, environmentally friendly alternative for pasteurization or shelf life extension of a wide range of food products (Welti-Chanes et al., 2005). The great potential of high pressure processing (HPP) in the food industry has been recently reviewed (Norton and Sun,2008).

Numerously studies including high pressure processing (HHP) technologies have been performed to develop the procedures replacing conventional heat treatment (pasteurization) of liquid egg which is applied at 60-65°C for 5-10 min (San Martín, Barbosa-Cánovas & Swanson,2002; Farr,1990). Using of HHP technology provide the better preservation of native properties of raw foods with similar antimicrobial efficacy as heat treatment. The profitable effects of HHP are demonstrated for many heat sensitive foods and liquid foods are treated in their packing material to avoid potential postinfection of the final product (Oey et al.,2008 ; Seregély et al.,2007).

Previous studies has shown that high pressure processing (HHP) technology is appropriate for destruction of various pathogen microorganisms in liquid whole egg and egg products (Jankowska et al., 2005; Ponce et al.,1999,1998). The viscosity of egg product is related to coagulation of specific egg proteins induced by HHP, thereby, the pressure is effective on the rheological product characteristics. For effectual treatment of LWE, not only achieving the satisfactory microbiological condition but also preserving the beneficial organoleptic and functional features of LWE are necessary (Tokuşoğlu,2013; Ahmed et al., 2003).

Microbial Stability, Preservation and Shelf Life of Table Eggs By High Pressure

HHP processing inactivates microorganisms, denatures proteins and extends shelf life of food products, with minor effects on nutritional value and flavour. For treatment of LWE, not only the achieving the satisfactory microbiological condition, but also the preservation of its beneficial organoleptic and functional properties are important (Guamis et.al.,2005; Ahmed et.al.,2003).

The HHP exposes the foods with pressures in the range of 100-1000 MPa with processing temperatures from below 0 C to 100 C where significant microbial reduction can be achieved (Huang et.al*,2006).*

It has shown that pressure treatments (300 - 450 MPa) at various temperatures (15, 20, or 50°C) for 5-15 min efficiently inactivated Salmonella *Enteritidis inoculated in liquid whole egg (Ponce* et al*., 1999, 1995).*

Bari et.al*.(2008) investigated the using of high pressure pulse treatment to inactivate* Salmonella *Enteritidis inoculated in liquid egg. In that study given by Bari* et.al*.(2008), liquid egg was inoculated with* Salmonella *Enteritidis (8.0 log colony-forming units [CFU]/mL) and exposed to hydrostatic pressures (300-400 MPa) and pressure (350 MPa) pulsing at 25 C, 40 C, and 50 C for up to 40 min to determine the maximum allowable pressure that can inactivate the* Salmonella *with minimal injury. Bari* et.al*.(2008) stated that strains of* Salmonella *SE-2 and SE-3 were the most sensitive strain to 400 MPa (25 C) pressure treatments for 10 min and a 8.0 and 7.0 log10 CFU/mL reduction were obtained for strain SE-2 and SE-3, respectively. Based on these study, strains SE-1 and SE-4 were the least sensitive and a 5.0 and 4.0 log10 CFU/mL of inhibition were achieved, respectively.*

It was shown that the result of HPP treatment of liquid egg inoculated with Salmonella *Enteritidis is shown in Table 1 (Bari* et.al*.,2008).*

Table 1. Populations of *Salmonella* Strain SE-4 Recovered from Liquid Whole Egg Following High Hydrostatic Pressure Treatment (Adopted from Bari *et.al*.,2008)

	Population (log_{10} CFU/mL)[a]	
Treatment	Survival	Reduction
Control	*9.12 0.12*	*0.00*
300 MPa (30 min)	*5.06 0.11*	*4.06 0.11*
350 MPa (30 min)	*4.37 0.10*	*4.75 0.10*
400 MPa (10 min)	*3.16 0.10*	*5.96 0.10*

[a]*Mean SD (n=3). Populations of* Salmonella *were recovered on tryptose soy agar medium. CFU, colony-forming units*

It was reported that the 300 and 350 MPa of pressure treatment for *Salmonella* Enteritidis in liquid eggs gave

the 4.0 and 4.8 log10 CFU/mL of reduction, respectively, whereas 400 MPa of treatment gave the 6.0 log10 CFU/mL of reduction at 25 C for up to 40 min (Bari *et.al*.,2008). The effects of HPP temperatures (25 C, 40 C, 50 C) on inactivation of *Salmonella* Enteritidis in liquid egg was monitored at 350 MPa pressure up to 40 min. It was found the highest inactivation of *Salmonella* in the liquid egg was observed at 50 C which resulted to a 6.0 log10 CFU/mL reduction (Figure 2).

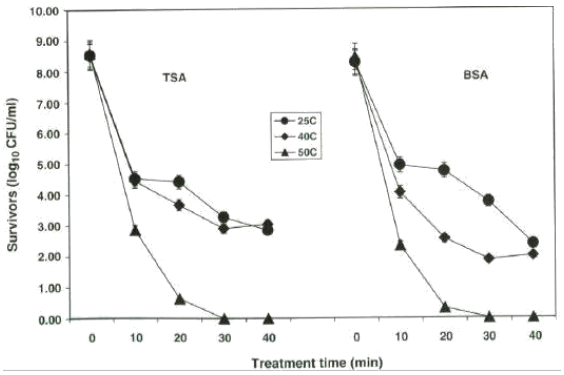

Figure 2*. Theeffects of HPP temperaturesoninactivation of* Salmonella*Enteritidisin LWE at at 350 MPa/ 40 min. Valuesaremeans ± SD of threeexperimentswithduplicatedeterminations (Adoptedfrom Bari* et.al.*,2008)*

It was concluded that when the treated liquid eggs were stored at 4 C, 25 C, and 37 C for 24 hours, no Salmonella *was detected in the samples (Bari* et.al.*,2008)*

HHP treatments have been applied to inactivate different microorganisms inoculated in liquid whole egg (Guamis et al.*, 2005). It was shown that treatments at pressure above 400 MPa combined with temperature of 50°C were able to reduce* Salmonella enteritidis *count by 8 log10 units whereas total bacterial count was also significantly reduced, and 10 cfu/ml of reduction was detected after 15 days of storage at 4 °C (Guamis* et al.*, 2005).*

It was applied 300–450 MPa/ 5–15min at temperatures of -15, 2, and 20 C to liquid whole egg inoculated with Listeria innocua *at a pH of 8.0 by Ponce* et al. *(1998b).* Listeria innocua *inactivation at 400 MPa followed the first-order kinetics for 0–20 min, and exhibited decimal reduction times D of 7.35 min at 2 C while 8.23 min at 20 C. The greatest inactivation (5 log reductions) was obtained at 450 MPa for 15 min at 20 C (Ponce* et al.*,1998b).*

Ponce et.al.*(1998b) stated that the highest reduction of* E.coli *in LWE was obtained at 50 C and it was reported that* E.coli *in LWE was more resistant to pressure at 20 C and -15 C than at 50 C and 2 C (Ponce* et al.*,1998b).*

10^7- 10^8 cfu/ml inoculation of Salmonella Enteritidis *in LWE were subjected to 350 MPa and 450 MPa at 50, 20, 2 and -15 C, with 5,10,15 min of treatmet times as well as cycles of 5-5 and 5-5-5 min treatments (Ponce* et.al.*,1999). It was concluded that inactivation*

increased with pressure and exposure time; the greatest inactivation (8 log cycles) occurred at the severest treatment conditions at 450 MPa/ 50 C whereas the minimal inactivation (1 log reduction) occurred at the lowest temperature and time conditions (Ponce et.al.*,1999).*

Lee et.al.*(2003) performed the effects of various pressures on* Listeria seeligeri *and* E.coli *(10^7 and 10^8 cfu/ml, respectively), in LWE at 5 C;* Listeria *reductions were not detected after 250 MPa and 350 MPa of treatments for 886 s and 200 s of exposure duration,respectively whereas 2 log reductions of* E.coli *were accomplished (Lee* et.al.*,2003). Yuste* et.al.*(2003) reported that 400 MPa/ 5 min of treatments resulted* E-coli *inactivation of 5.5 log cycles in LWE, whereas no* Salmonella typhimurium, Yersinia enterocolitica, Listeria monocyctogenes *were detected (Yuste* et.al.*,2003). Isiker* et.al.*(2003) stated that increasing the pressure had a significant effect on* Salmonella enteridis *inactivation in LWE (Isiker* et.al.*,2003).*

Dong-Un Lee (2002) stated that kinetic studies on the isothermal HHP inactivation of E. coli *in liquid whole egg were performed at 5 and 25 °C in the pressure range of 250 to 400 MPa. and the characteristic tailing inactivation curves were described by a first order biphasic model.*

It was prevailed that the degree of E. coli *inactivation at isothermal pressure condition was independent of applied pressures if the physical characteristics of LWE are considered, i.e., between 2.0 to 3.0 log reductions at 5 °C, and less than 1.0 log reductions at 25 °C in the range of 250 to 400 MPa, so, HHP at 5 °C is more favorable than at 25 °C (Dong-Un Lee, 2002). It was reported that about 3 log reductions of* E. coli *and over 5 log reductions of* Pseudomonas *and* Paenibacillus*, HHP treatment of LWE at 5 °C is regarded to be as effective as conventional thermal pasteurization (Dong-Un Lee, 2002).*

Dong-Un Lee (2002) stated that the HHP processing conditions were fixed to either 250 MPa for 886 s at 5 °C or 300 MPa for 200s at 5 °C which have been indicated as the optimized HP processing conditions. It was put forwarded that the addition of nisin (Figure 3) prior to pressure treatments significantly increased the lethal effects of HHP against Listeria seeligeri*. The individual effects of each nisin and HHP on the* Listeria *reductions were very small, and the increased* Listeria *reductions, up to 5 log cycles were obtained owing to the synergistic action of bactericidal effect of nisin and high pressure effects (Dong-Un Lee,2002).*

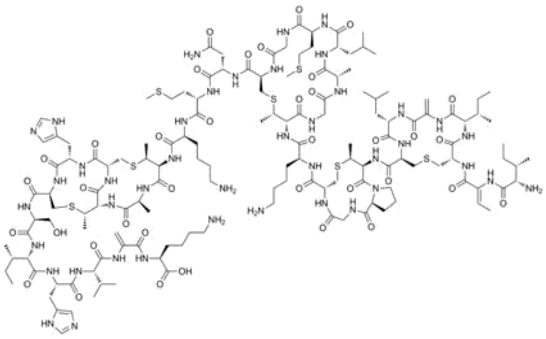

***Figure 3**. TheChemical Formula of Nisin*

The Nisin-HHP combination can be effectively reduced the microbial loads of Gram-negative E. coli. *It was concluded that the marginal effects of Nisin-HHP synergy on* E. coli *reduction in LWE can be expressed by the membrane structure of Gram-negative* E. coli *or by the protective effects of LWE. (Dong-Un Lee,2002).*

Juliano (2006) studied the inactivation of Bacillus stearothermophilus *after different stages in the process: (a) baking of egg mix to form patty, (b) after preheating, (c) after high pressure high temperature (HPHT) processing. In the study described by Juliano (2006),* Bacillus stearothermophilus *spore inoculated in the egg mix showed a one log cycle reduction after baking. It was reported that the inactivation of* B. stearothermophilus *(ATCC 7953) spores in egg patties was accelerated after pressure-assisted thermal processing (PATP) treatment at 700 MPa/105℃ and* B. stearothermophilus *spores was inactivated, rapidly in egg matrix (4 log reductions in 5 min) when compared to thermal treatment at 121 °C (1.5 log reduction in 15 min) (Rajan* et al.*, 2006). Similar results was found by Koutchma* et al.*(2005) and the inactivation of* B. stearothermophilus *in spore strips located between two egg patties can be reduced by at least 6 log cycles at 688 MPa/ 105℃ in 5 min (Koutchma* et al.*, 2005).*

Rheological Properties of Table Eggs By High Pressure

The egg is a low acid food (higher pH) that necessitates preservation by some means to increase the shelf life. It was found that HHP treated egg white at 600 MPa or more gets fully coagulated to form gels (Bridgeman,1914).

It is known that the protein can be denatured, coagulated or gelled, and it depends on several factors such as pH, protein type, temperature, applied pressure and ionic strength. HHP has been focused on food proteins, and its functional properties, modification and texture (Ahmed et.al.*,2003). It is reported that HHP has reduced the alterations of post-process contamination, coagulation and better retention of nutritional qualities of egg as the process is carried out at considerably low temperature and in packed form and also the product could be consumed directly without heat treatment (Ahmed* et.al.*,2003; Knorr, 1996).*

Pressure induce protein denaturation, depending on the protein concentration, pressure level, temperature, and pH (Balny & Masson, 1993).

Egg contains protein of high biological value as compared to any dietary protein. Egg proteins have all the desirable nutritional and functional properties, so egg proteins are widely used in food technology (Lee et al.*, 1999; Hsieh* et.al.*,1993).*

For the food quality control, sensory evaluation, food process and equipment design and also for the new product development; the information of rheological properties of foods is necessary. Based on the origin, chemical and nutritional composition and structure behavior and previous history, the flow behavior of a fluid can be varied from Newtonian to timedependent non-Newtonian in nature (Rao, 1986). It was reported Newtonian and/or time-dependent non-Newtonian flow behavior of egg (Lee, Heinz & Knorr,1999; Comford et.al.,1969) and it was studied the rheology of commercial egg gel white at high temperature using creepand compression measurement (Nagano & Nishinari,2001).

The denaturation or coagulation or structure break down occurs in albumen or egg white protein and the role of protein structure on emulsion and gel rheology is important, so, HHP effects on the rheological properties of egg is also significant (Ahmed et.al.*,2003).*

Ahmet et.al.*(2003) stated that whole liquid egg (WLE) and albumen have been denatured at high hydrostatic pressure (HHP) and it was reported that 100-400 MPa/30 min of HHP application affected the rheological characteristics of WLE, albumen, and yolk (Ahmet* et.al.*,2003). In the study given by Ahmed* et.al.*(2003), an advanced controlled stress rheometer was employed to study the rhelogical properties at a shear rate of 0– 200 s^{-1} using double concentric cylinder for WLE and albumen while parallel plate geometry was used for yolk with shear rate range of 0–500 s^{-1}. It was stated that both WLE and albumen behaved as time dependent fluids (thixotropic) however*

*HHP reduced time dependency substantially. It was also determined that albumen individually exhibited more pressure effect compared to WLE and thixotropy of yolk significantly varied (*p *0:05) during HHP (Ahmet* et.al.*,2003).*

Table 2. *Rate of Thixotropy and Area Under the Curve of Egg Components Obtained from Software (Adapted from Ahmed* et.al.*,2003)*

Sample	*Pressure (MPa)*	*Rate of Thixotropy (Pas^{-1})*	*Area inside the curve ($s^{-1}Pa$)*
WLE	*0.101*	*54.85*	*556.7*
	150	*51.12*	*345.6*
	200	*49.56*	*329.6*
	250	*42.52*	*144.6*
	300	*514.9*	*1618*
	350	*384.7*	*1253*
Albumen	*0.101*	*287.4*	*790*
	150	*34.22*	*111.7*
	200	*11.91*	*63.45*
	250	*7.87*	*53.87*
	300	*81.86*	*423.4*
	350	*ND*	
Yolk	*0.101*	*21,930*	*84,840*
	150	*24,620*	*91,500*
	200	*25,768*	*89,680*
	250	*ND*	
	300	*30,460*	*92,657*
	350	*36,830*	*99,096*

It was studied the effects of high hydrostatic pressure (HHP) on rheological parameters of whole liquid egg (WLE), albumen and yolk as egg components (Table 3 and 4). It was shown that for egg albumen, the magnitude of yield stress and consistency coefficient decreased during pressurization; however, coagulation of protein reversed the trends. It was determined that all egg samples behaved as thixotropic fluid and the structure break down of egg protein enhances with high pressure and it completed at 300MPa/30 min at 20 C (Ahmed et.al.*,2003).*

Table 3. *Effect of Pressure on Rheological Paramaters of Liquid Whole Egg (LWE) Using Herschel-Bulkley Model (Adapted from Ahmed* et.al.*,2003)*

	Pressure (MPa)	*Yield Stress (Pa)*	*Consistency Coefficient ( ), Pas^n*		*Flow Behavior Index (n)*	*Standard Error*
Liquid Whole Egg (LWE) (Shear Rate 0-200 s^{-1})	*0.101 up*	*0.536*	*0.058*		*0.753*	*30.11*
	0.101	*0.171*	*0.023*		*0.959*	*17.07*
	Dn 150	*0.323*	*0.044*		*0.876*	*17.55*
	up	*0.165*	*0.022*		*0.961*	*9.48*
	150 Dn	*0.400*	*0.054*		*0.893*	*15.55*
	200 up	*0.309*	*0.019*		*1.079*	*13.61*
	200 Dn	*0.334*	*0.324*		*0.367*	*19.66*
	250 up	*0.205*	*0.032*		*1.094*	*11.12*
	250 Dn	*0.769*	*0.786*		*0.276*	*16.45*
	300 up	*0.832*	*0.055*		*0.962*	*8.63*
	300 Dn	*1.676*	*0.887*		*0.186*	*18.44*
	350 up	*0.506*	*0.049*		*0.903*	*7.56*
	350 Dn					

Table 4. *Effect of Pressure on Rheological Paramaters of Egg Components Albumen and Yolk Using Herschel-Bulkley Model (Adapted from Ahmed et.al.,2003)*

Egg Components	*Pressure (MPa)*	*Yield Stress (Pa)*	*Consistency Coefficient ( ), Pas^n*	*Flow Behavior Index (n)*	*Standard Error*
Albumen (Shear Rate 0-200 s^{-1})	*0.101 up*	*0.768*	*0.032*	*0.87*	*16.77*
	0.101 Dn	*0.536*	*0.015*	*1.039*	*13.21*
	150 up	*0.096*	*1.43E-3*	*0.954*	*2.57*
	150 Dn	*0.086*	*1.28E-3*	*0.977*	*2.99*
	200 up	*0.053*	*4.016E-3*	*0.914*	*16.13*
	200 Dn	*0.051*	*3.00E-3*	*0.969*	*15.88*
	250 up	*0.033*	*2.41E-2*	*0.564*	*15.22*
	250 Dn	*0.027*	*1.83E-3*	*1.023*	*11.77*
	300 up	*0.220*	*0.642*	*0.248*	*27.33*
	300 Dn	*0.741*	*0.016*	*0.851*	*19.19*
Yolk (Shear Rate 0-500 s^{-1})	*0.101 up*	*10.68*	*21.36*	*0.473*	*53.45*
	0.101 Dn	*9.529*	*7.363*	*0.514*	*6.85*
	150 up	*a*			
	150 Dn	*10.29*	*8.13*	*0.516*	*5.92*
	200 up	*a*			
	200 Dn	*11.26*	*8.52*	*0.509*	*7.068*
	250 up	*a*			
	250 Dn				
	300 up	*a*			
	300 Dn	*21.14*	*9.38*	*0.450*	*6.56*
	350 up				
	350 Dn	*28.61*	*10.09*	*0.448*	*7.23*

High pressure level at 400-600 MPa can cause enough alterations in the viscosity of egg components so that it can become gel with improved quality characteristics than the heat induced gels (Hayashi et.al*,1989). It was expressed that pressure induced gels were softer than untreated samples, more elastic without any cooked taste and flavour and there was no destruction of vitamins and formation of lysinoalanine (Hayashi* et al.*,1989).*

High Pressure Effects on Egg Phosvitin, Ovalbumin and Ovotransferrin

Egg yolk phosvitin represents about 7% of the proteins found in egg yolk and is a highly phosphorylated protein of egg (Samaraweera et.al.*,2011; Abe* et.al.*,1982). Phosvitin, a highly phosphorylated glycoprotein and represents the major fraction of hen egg yolk phosphoproteins (Anonymous,2013). It is known that phosvitin is rich in serine residues and phosphorylated peptides, i.e. phosphopeptides,with antioxidant and mineral-binding ability could be a great source of natural functional biopeptides (Jiang & Mine,2000).*

Volk et.al.*(2012) stated that phosvitin structure maintained overall during high-pressure treatment of 600 MPa applied at an initial temperature of 65 C regardless of the pH and treatment duration, confirming the high structural stability of the phosphoprotein. It was reported that treatment of phosvitin with phosphatase increased the degree of dephosphorylation from 24% to 63%, after 2 and 18 h, respectively. It was also found that angiotensin-converting enzyme (ACE) inhibition and antioxidant activity of dephosphorylated and protease-treated phosvitin was increased by 52% and 39%, respectively, as compared to protease-digested native phosvitin.*

It was showed that pressure treatment of egg white proteins above 450 MPa resulted in a loss of secondary structure (Hayakawa et al.*, 1996). The pressure-induced structural alterations in egg white proteins can also be demonstrated by exposuring of previously buried SH groups and hydrophobic groups (Van der Plancken* et al.*, 2004, 2005ab, 2006 ; Iametti* et al.*, 1999). Iametti* et.al.*(1999) stated that the treated albumen had increased viscosity but retained its foaming and heat-gelling properties (Iametti* et al.*, 1999). It was found that susceptibility of egg albumen proteins to hydrolysis by trypsin increased dramatically after HHP treatment (up to 10 min at 800 MPa).*

As it is known, ovalbumin (OVA) (Figure 7) is the major protein found in egg white, making up 60-65% of the total protein (Huntington & Stein, 2001) and it plays a major role in determining of egg

white behavior on application of HHP (Messens et.al.,1997). High pressure can result in structural modification of egg white that can be correlated to enhancement of functional properties (Messens et al., 1997). The S-form of ovalbumin, the presence of which is an index of egg aging, was not found in any of the pressure-treated samples, that also did not display evidence for covalent protein aggregation (Iametti et al., 1999).

It was reported that the foaming capacity of egg white have been improved owing to the exposure of SH groups that favours foaming stability and capacity (Van Der Plancken et.al.,2007a; Yang et.al.,2010). It was reported that the turbidity, surface hydrophobicity and exposure of sulfhydryl groups in egg white proteins were also increased at pressures over 400 MPa application and the strong increase in surface hydrophobicity was observed between 400 and 700 MPa (Yan et al., 2010 ; Van der Plancken et al.,2005a,2007b). Besides, the decrease and increase of total and exposed SH groups, respectively, were enhanced by pressure above 500 MPa (Van der Plancken et al.,2005a). It was reported that the pressure treatment at 410 MPa induced proteins in egg yolk dispersions to aggregate and undergo a sol-gel transition (Aguilar et al., 2007), while the treatment at 600 MPa resulted in the modification of emulsifying properties without effect on the protein solubility of LDL solutions (Speroni et al., 2005).

It is known that ovotransferrin, accounting for 12–13% of egg white proteins, is a glycosylated protein with an isoelectric point of 6.1 (Huopalahti, López-Fandiño, Anton, & Schade, 2007) and Huopalahti et al. (2007) stated that the ovotransferrin shows 50% homology with mammalian transferrin and lactoferrin, but differs from the other transferrin proteins in its isoelectric point and in the glycosylation pattern (Huopalahti et al., 2007). It was also stated that ovotransferrin is also responsible for the ferric ion transfer from the hen oviduct to the developing embryo (Huopalahti et al., 2007) and it was found that ovotransferrin possessed antifungal activity (Valenti et.al.,1985), immunomodulatory and antiviral activity (Giansanti et al., 2002,2005,2007), antioxidant and anticancer activities (Ibrahim et.al.,2007; Ibrahim & Kiyono,2009). Ovotransferrin is known a rich source of bioactive peptides and recently, there is an great attention regarding the potential of ovotransferrin as functional food and nutraceutical ingredient (Wu & Acero-Lopez, 2011).

Current researches indicated that sonication could affect the exposure of SH groups of ovotransferrin and could release of potent antihypertensive peptides (Lei

et.al.,*2011; Majumder & Wu, 2010). After HHP processing, the conformational and physicochemical alterations are important due to affecting the functional properties of food proteins and also protein bioactivities.*

Acero-Lopez et.al.*(2012) reported the effect of high pressure treatment on ovotransferrin. and it was determined that HHP treatment caused changes in ovotransferrin structure depending on the pH of the sample (Acero-Lopez* et.al.*2012). It was focused that the determination of high pressure effect on the structure and physicochemical properties of ovotransferrin concentrate after processing in an acid (pH 3) and in a basic (pH 8) environments. It was found that, a decrease in total sulfhydryl groups and an increase in surface hydrophobicity were observed along with a partial aggregation at pH 8 and pressures higher than 200 MPa. It was also stated the ovotransferrin adopted a molten globule state at pH 3, and associated with a significant increase in surface hydrophobicity and reactive sulfhydryl content (Acero-Lopez* et.al.,*2012).*

Figure 8 shows the alterations in total sulfhydryl groups and in reactive sulfhydryl content (Figure 8) (Acero-Lopez et.al.,*2012). It was stated that ovotransferrin treated at 200 MPa at pH 8 shows a total SH content of 4 mol SH/g, that is close to the control; whereas further increasing pressure led to considerable decrease in the total SH content to around 2 mol SH/g and 0.9 mol SH/g at 400 and 700 MPa, respectively (Fig.8A).*

In the study decribed by Acero-Lopez et.al.*(2012), the most evidential altaration in reactive SH content was observed at 600 and 700 MPa where it decreased from 1 mol SH/g to about 0.2 mol SH/g (Fig 8B). Van Der Plancken* et.al.*(2005b) revealed that decreasing of the total SH groups was probably due to rearrangement of cysteine residues and oxidation of SH groups (Van Der Plancken* et.al.,*2005b).*

In the study reported by Acero-Lopez et.al.*(2012), it was found the gradual increase in denaturation peak from control up to 400 MPa (Fig. 9). Figure 9 shows differential scanning calorimetry (DSC) thermogram of ovotransferrin samples treated at various pressures between 200-700 MPa at pH 8 (Acero-Lopez* et.al.,*2012).*

High Pressure Effects on Foaming Properties of Egg

Heated liquid eggs coagulate or solidify (as cakes, breads, crackers), whipped egg white produces airier and lighter products (meringues, marshmallow, angel cake), and emulsified egg yolk

phospholipids and lipoproteins produces special products (mayonnaise, salad dressing and sauces) (Davis & Reeves,2002). It is known that food foaming characteristics of egg albumen are quite good. Ferreira et.al.*(1995) stated that foaming properties are evaluated by foaming capacity (*FC*) and foam stability (*FS*). For the determination of* FC *and* FS *the following formulae are used as shown in below (Chang & Chen,2000):*

FC ***(%)*** = *(*FV/ILV*) ×100% ;* **FS** ***(%)*** = *[(*ILV − DV*)*/ILV*] ×100% ;* ***Drainage (ml)*** = LVM – LVS

where: FV – *volume of foam;* ILV – *volume of the initial liquid phase;* DV – *volume of drainage*
LVM – *volume of the liquid phase at* t = *60 min after foaming was finished*
LVS – *volume of the liquid phase at* t = *30 s after foaming*

Lomakina & Míková (2006) reported that various foods are prepared using egg white, most of them being based on the foaming properties of egg white that are owing to the albumen proteins ability to encapsulate and retain air (Lomakina & Míková,2006). Due to the foaming properties of egg white, new methods have been improved the volume and the stability of egg white foam (Lomakina & Míková,2006).

Foaming is affected by water (Baldwin,1986) by temperature, sugar (Stadelman & Cotterill,1994), by egg yolk (Kim & Setser,1982), by oil differency and quantity (Stadelman & Cotterill,1994; Kim & Setser,1982) and also by stabilisers and surfactants (Kim & Setser,1982).

It was expressed that pasteurisation of egg albumen decreased the foaming ability and resulted in the quality reduction and volume of angel cake, this is occurred by ovotransferrin denaturation on pasteurisation at 53°C. Hatta et.al.*(1997) stated that using of metallic ions (Fe, Cu, Al, or other) and salts of phosphoric and citric acids for the increasing of denaturation temperature and the improvement of the foaming properties of egg albumen after pasteurisation (Hatta* et.al.*,1997).*

It was reported that pasteurised egg white required a longer whipping to attain a foam comparable in specific gravity to the foam from unpasteurised egg albumen (Stadelman & Cotterill,1994). Ma et.al.*(1994) stated that effects of chemical modifications on the physicochemical and cakebaking properties of egg white and they reported that the overrun and the foam stability of spray-dried egg white increased significantly by gamma irradiation processing (Ma* et.al.*,1994). It was reported the time for 50% drainage, an index of the foam stability, increased by irradiation with higher dosages indicating improvement in the foam stability whereas decreased in the overrun at 4 kGy but no*

alteration in the foam stability for the frozen egg white (Ma et.al.*,1994).*

It was indicated that the greater increasing of the foaming power observed in the case of ultrasound high pressure combination may be explained by the homogenisation effect of ultrasound (Knorr et.al.*,2004). Knorr* et.al.*(2004) expressed the foaming capacity of liquid egg white by ultrasound processing due to ultrasound dispersed the protein and fat particles in liquid egg white (Knorr* et.al.*,2004). The Table 5 shows the combined processing effects on the foaming capacity of liquid whole egg (Knorr* et.al.*,2004).*

Table 5*. The Combined Processing Effects on the Foaming Capacity of Liquid Whole Egg (Adapted from Knorr* et.al.*,2004).*

Processing	Power	Stability
	Overrun (%)	Stability (%)
Control	479	52
High Pressure (HHP)	490	56
Nisin- HHP	484	55
Ultrasound-HHP	638	50

Hoppe (2010) reported that foaming properties of egg white solutions (10% v/v) were analyzed with varying levels of pressure and pH. It was found that pressure treatment of 10% egg white solutions at pH 9.11 resulted in a homogenous solution with improved foaming capacity over the control (Figure 10) (Hoppe,2010). Figure 10 shows the effect of HPP on foam overrun at pH 9.11 at 0.1 MPa (control), 600 MPa, and 800 MPa (Hoppe,2010). It was shown that increasing pressure resulted in an increase in foam volume and foam overrun increased significantly ($\alpha = 0.05$) at 800 MPa at all time points (Hoppe,2010).

Hoppe (2010) expressed that the foaming properties of egg white solutions were also highly dependent on pH (Hoppe,2010). It was found that the greatest foam overrun was achieved at pH 4.5 wheras foaming ability was significantly decreased at pH 6 (Figure 11). In the study described by Hoppe (2010), it was also reported the increased foam overrun at pH 4.5 could be attributed to major egg white proteins (ovalbumin and ovomucin) important to foaming properties, ovalbumin and ovomucin, which have respective pI of 4.5 and 4.1. (Hoppe,2010).

It was found that foam stability determined the effect of HPP or pH on egg white foaming properties (Hoppe,2010). In the study given by Hoppe (2010), HPP significantly reduced foam stability with the exception of the 800 MPa 0 time point and this data was in

in contrast to the study described by Van der Plancken et. al.*(2007a) and was found the HHP treatment increased overall foam stability (Van der Plancken* et. al.*,2007a).*

Figure 12 shows the effect of HPP (5 min) on 10% egg white solution foam

stability at pH 9.11 and at 0.1 MPa (control), 600 MPa, and 800 MPa (Figure 12) (Hoppe,2010). In the study described by Hoppe (2010), the increased stability of 800 MPa at the 0 time point was attributed to the increased foam volume and incorporation of liquid in the foam. It was found that the liquid drainage was the greatest over the first 5 minutes post-foam (800 MPa) as indicated by the slope and drop in stability as shown in Figure 12 (Hoppe,2011).

High Pressure Effects on Color and Texture Properties of Egg

Singh & Ramaswamy (2010) reported that L ,a ,b values increased with increasing in pressure intensity for whole egg. In eggyolk, L remained mostly stable and a value decreased whereas b values showed great increasing in yellowness. It was also reported that high pressure processing (HPP) induced an increasing in L (lightness) value and a (redness) value of egg white up to 700 MPa while the b value simultaneously decreased indicating decreasing yellowness with increasing treatment intensity (Singh & Ramaswamy,2010).

It was demonstrated that high pressure processing affected the color values of egg white, egg yolk and whole liquid egg respectively and it was found that L value increased linearly at all pressure-time combinations indicating increase in brightness of sample with increasing pressure treatment intensity for egg white (Table 6) (Singh (2012).*

***Table 6**. Hunter L (Lightness) Values of the Egg White and Yolk Subjected to Pressure Level and Treatment Time (Adapted from Singh,2012)*

L* Value				
Egg White				
Pressure	***Time***			
	0	***5***	***10***	***15***
600	*58 ± 0.707*	*63.5 ± 3.5*	*66 ± 4.24*	*80 ± 1.14*
700	*81 ± 1.41*	*86*	*85.5 ± 0.70*	*91.5 ± 0.70*
800	*86 ± 3.53*	*89 ± 4.24*	*91.5 ± 2.12*	*96 ± 1.41*
900	*88 ± 2.82*	*91.5 ± 3.5*	*94 ± 1.41*	*100.5 ± 0.70*
Egg Yolk				
Pressure	***Time***			
	0	***5***	***10***	***15***
600	*57.4 ± 0.84*	*56.8 ± 0.28*	*53.85 ± 0.21*	*61.1 ± 0.14*
700	*57.05 ± 1.34*	*55.7 ± 0.49*	*51.75 ± 0.35*	*44.8 ± 0.070*
800	*55.3 ± 0.98*	*50.6 ± 0.84*	*50.6 ± 0.56*	*51.25 ± 0.07*
900	*57 ± 1.41*	*53.15 ± 0.21*	*55.1 ± 0.28*	*58.5 ± 0.28*

It was found a values increased with increasing pressure treatment*

intensity whereas there was small increase in b value for egg white (Singh,2012). Table 7 shows the Hunter a* and b* values values of the egg white subjected to 600-900 MPa of high pressure at 1-15 min of treatment time (Table 7) (Singh,2012).*

Table 7. *Hunter a (redness) and b (yellowness) Values of the Egg White and Yolk Subjected to Pressure Level and Treatment Time (Adapted from Singh,2012).*

a Value				
Egg White				
Pressure	***Time***			
	0	***5***	***10***	***15***
600	*1.2 ± 0.007*	*2.005 ± 0.021*	*2.22 ± 0.4*	*2.26 ± 0.014*
700	*2.8 ± 0.077*	*2.91 ± 0.01*	*3.125 ± 0.035*	*3.13 ± 0.01*
800	*1.94 ± 0.65*	*2.065 ± 0.021*	*2.005 ± 0.007*	*2.35 ± 0.07*
900	*1.9 ± 0.02*	*2.095 ± 0.021*	*20.025 ±0.007*	*2.25 ± 0.07*
Egg Yolk				
Pressure	***Time***			
	0	***5***	***10***	***15***
600	*9.51 ± 0.014*	*8.46 ± 0.05*	*8.405 ± 0.007*	*61.5 ± 0.07*
700	*8.63 ± 0.04*	*5.84 ± 0.06*	*5.36 ± 0.05*	*4.15 ± 0.07*
800	*4.15 ± 0.07*	*2.95 ± 0.07*	*2.6 ± 0.28*	*1.85 ± 0.07*
900	*3.95 ± 0.07*	*2.765 ± 0.91*	*2.36 ± 0.06*	*2.11 ± 0.014*
b Value				
Egg White				
Pressure	***Time***			
	0	***5***	***10***	***15***
600	*1.9*	*1.61 ± 0.014*	*1.805 ± 0.0007*	*2.25 ± 0.07*
700	*1.5 ± 0.14*	*1.19 ± 0.014*	*1.11 ± 0.014*	*1.25 ± 0.07*
800	*1.3 ± 0.07*	*1.11 ± 0.014*	*0.98 ± 0.0141*	*0.855 ± 0.021*
900	*0.99 ± 0.01*	*1.085 ± 0.021*	*1.615 ± 0.021*	*2.214 ± 0.0021*
Egg Yolk				
Pressure	***Time***			
	0	***5***	***10***	***15***
600	*44.19 ± 15.2*	*56.15 ± 1.20*	*58.1 ± 0.14*	*58.1 ± 0.14*
700	*61.1 ± 0.14*	*62.5 ± 0.70*	*62.25 ± 1.06*	*63.4 ± 1.27*
800	*64.75 ± 0.35*	*66.5 ± 3.53*	*70.4 ± 0.28*	*70.6 ± 0.28*
900	*69.3 ± 1.83*	*70.5 ± 2.12*	*72.5 ± 2.12*	*75.5 ± 0.70*

Singh (2012) reported that egg yolk containing high level of xanthophylls (yellow color) showed different color behavior than that of egg white and egg yolk color changed from pale yellow to

orangish yellow as per visual appearance (Singh,2012). It was found that L value remained constant and a* value decreased from 9.51 to 2.11 indicating diminution in redness of sample whereas b* value increased significantly* ($p < 0.05$) *from 56.5 to 76.5 showing great deal of increasing in yellow color of the egg yolk (Table 6,7) (Singh,2012). It is known that yellower color is desirable from a customer viewpoint.*

It is known that ΔE is the total color difference, it has been represented the color variance of foods during processing (Equation 1). ΔE is obtained as the combined differences in L, a*, and b* values and ΔE is calculated using L*, a* and b* values whereas raw egg components acted as reference (Azarpazhooh and Ramaswamy, 2011).*

$$\Delta E = \sqrt{(L_0^*\text{-}L^*)^2 + (a_0^*\text{-}a^*)^2 + (b_0^*\text{-}b^*)^2}$$

(Equation 1).

Ahmed et al. *(2005) found that ΔE remained constant even after increasing in high pressure processing treatment and this situation indicated the stability of pigments (Ahmed* et.al.*,2005). It was concluded that ΔE increased with increasing in pressure level and treatment time (Singh,2012).*

It is valued that texture is an imperative characteristic of egg and egg based products that affects consumer perception and overall acceptability. Textural alterations in egg constituents are very sensitive to food processing types and utilized parameters (Kilcast & Lewis,1990; Hayashi et.al.*,2000; Tokuşoğlu,2013). Pons & Fiszman (1996) stated that HPP can be used to modify food proteins in controlled manner so as to make egg gels with better quality, uncooked flavor and better textural properties. HPP not only improved the color but also resulted in a more firmer texture than heat coagulated egg products (Singh & Ramaswamy,2010).*

With the increasing in pressure level and treatment time, texture properties including firmness, springiness, cohesiveness, gumminess and chewiness improved for egg white. For egg yolk; firmness, adhesiveness, gumminess, chewiness, resilience were enhanced while cohesiveness decreased with an increasing in pressure level and treatment severity (Singh & Ramaswamy,2010) (Figure 13). Singh (2012) stated that the texture profiles including hardness, adhesiveness, cohesiveness, chewiness, gumminess and springiness of pressure treated egg white, egg yolk and whole liquid egg samples.

It was reported that hardness of whole liquid egg (WLE) increased with increasing in pressure level and treatment time but hardness values were lower than egg yolk and higher than egg white

(Singh,2012). With the high pressure application, the form of egg gels was very adhesive and elastic in the study described by Singh (2012) and these data was accordant with that of reported by Hayashi et al.*(1989). Similarly, it was found that high pressure coagulated egg white gels were more adhesive and elastic than thermally treated gels (Hayashi* et al.*,1989). According to another study, high pressure processed gels have softer structure than that of thermal treatments (Carlez* et al.*,1995). It is known that adhesiveness is related to surface properties. In the study reported by Singh (2012), adhesiveness decreased linearly with increase in pressure level and treatment time for egg white (EW).*

Singh (2012) found that increasing pressure level from 500-900 MPa affected the adhesion properties of egg constituents and WLE followed an increasing trend while adhesiveness value of EW was two fold to that of the WLE. It was also found that EY samples were more adhesive physically than EW and WLE. For adhesiveness, egg components followed EY>EW>WLE pattern, where EY demonstrated maximum hardness (Figure 14). It was interpreted that, highest increase in egg yolk could be the higher amount of fat level in egg yolk matrix, thus increasing adhesiveness (Singh,2012; Singh & Ramaswamy,2010).

It was found that egg white (EW) showed different behavior than those of egg yolk (EY) and whole liquid egg (WLE) because of their high protein content. It was reported that high pressure processing (HPP) caused egg white coagulation and increasing the intensity of pressure level and treatment time caused the egg white gelation (Singh,2012).

It was reported that EW turned to opaque at 600MPa/15 min of HHP treatment and was able to form egg gels that can stand by themselves. EY was able to form gels at 700MPa/15 min while WLE was able to form gels at very short time processing treatment of 700MPa/10 min (Singh,2012).

Singh (2012) reported that egg constituents changed from liquid state to complete gel with coagulation and gelation of egg constituents by increasing pressure application. It was stated that egg gels were formed at high pressure level greater than 600 MPa at temperature well below that required for thermal gel formation (Singh,2012). It was concluded that HHP lead to formation of full set egg gels with improved physicochemical and functional characteristics and without any cooked flavors.

Hoppe (2010) reported that egg gels formed with heat at 95°C had an average hardness value and over twice the value of HP-induce gels at 800 MPa

(Figure 15) while the softest gel was observed at 600 MPa (Hoppe,2010). It was also found that pH reduction decreased the hardness of heat induced gels while it increased the hardness of HHP gels in the study described by Hoppe (2010). Egg gel gumminess was determined and was found with similar pattern to gel hardness with heat induced gels being gummier (Figure 15). Figure 15 shows the effect of heat and HHP treatment on egg white gel hardness and gel gumminess (Hoppe,2010).

Monfort et.al.*(2012) reported that design and evaluation of a high hydrostatic pressure combined process for pasteurization of liquid whole egg (LWE). It was put forwarded the physicochemical and functional properties of non-treated LWE and HHP treated LWE (300 MPa/3 min at 20 °C) followed by heat treatment (52 °C/3.5 min or 55 °C/2 min) in the presence of 2%, and current heat ultrapasteurization treated LWE (Table 8) (Monfort* et.al.,*2012).*

Table 8. *The Physicochemical and Functional Properties of Non-treated LWE, HHP Treated LWE (300 MPa/3 min at 20 °C) Followed by Heat Treatment (52 °C/3.5 min or 55 °C/2 min) in the Presence of 2%, and Current Heat Ultrapasteurization Treated LWE (Adapted from Monfort et.al.,2012)*

		Control	***TC- HHP- HT***		***Ultrapasteurization***
			HHP + 52 C / 3.5 min + 2% TC	***HHP + 52 C / 3.5 min + 2% TC***	***71 C /1.5 min***
Physicochemical Properties					
pH		*7.64 ± 0.06*	*99.9 ± 0.1*	*99.7 ± 0.4*	*102.1 ± 0.7*
L		*35.0 ± 0.4*	*103.8 ± 0.2*	*108.0 ± 0.2*	*118.8 ± 1.7*
a		*11.4 ± 0.3*	*114.0 ± 0.2*	*1143 ± 0.1*	*66.2 ± 1.2*
b		*25.2 ± 0.5*	*93.6 ± 0.1*	*98.2 ± 0.1*	*61.6 ± 1.3*
Viscosity (mPa s)		*12.7 ± 0.1*	*156.4 ± 12.4*	*1323 ± 0.6*	*239.4 ± 2.9*
Soluble Protein		*0.741 ± 0.024*	*88.3 ± 0.1*	*88.3 ± 0.1*	*84.7 ± 0.7*
Functional Properties					
Foaming	*Foaming Capacity (%)*	*504.0 ± 1.6*	*126.5 ± 1.3*	*126.4 ± 0.6*	*31.6 ± 0.8*
	Foaming Stability (min)	*4.68 ± 0.37*	*218.0 ± 28.3*	*186.2 ± 5.9*	*16.0 ± 2.5*
Emulsifying	*Emulsifying Capacity(%)*	*62.1 ± 0.8*	*97.5 ± 2.1*	*95.8 ± 2.5*	*31.2 ± 0.7*
	Emulsifying Stability (min)	*80.6 ± 7.1*	*95.6 ± 2.5*	*89.0 ± 1.7*	*88.4 ± 24.7*
Gelling	*WHC (%)*	*86.6 ± 0.5*	*97.3 ± 1.4*	*99.2 ± 1.4*	*100.3 ± 0.6*
	Hardness (g)	*1041.0 ± 44.5*	*123.2 ± 2.5*	*100.7 ± 3.6*	*126.8 ± 2.0*

In the study reported by Monfort et.al.*(2012), gels from ultrapasteurized LWE were harder than those prepared with LWE treated by HHP, as reported by Van der Plancken* et.al.*(2005) in egg white. It was found that, in the case of gelling properties, gels of treated LWE showed similar values of hardness and water holding capacity (WHC) (Table 14). Marco-Moles* et.al.*(2011) rendered that higher hardness values could indicate that protein-based conformational structures may be irreversibly impaired by high-temperature processes. The designed treatment at 52 °C/3.5 min by Monfort* et.al.*(2012) resulted in harder gels than the treatment at 55 °C/2 min or the non-treated LWE.*

Overall, HPP improved the functional and physicochemical properties of egg white, egg yolk and liquid whole egg. HPP is an emerging technology with the potential to increase new functional properties to food products.

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