

Structural Changes in Albumen Quality during Storage Period

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The egg storage environment influences the nutritional and functional properties of albumen. Changes in albumen quality during storage are characterized by various physical and chemical reactions that are affected by either an increase or a decrease in storage time and temperature. Changes in albumen structure indicate a decline in albumen quality during storage. Albumen traits, such as Haugh unit value (HU) value, albumen pH, and thick albumen height, are often used as indicators of albumen quality during storage and are influenced by storage conditions in a time- and temperature-dependent manner. These traits would invariably influence the albumen's functional, rheological, and biological properties.

Keywords: egg ; albumen quality ; storage time and temperature

1. Egg Weight Loss

During storage, egg weight loss occurs and influences the egg components (albumen and yolk). The rate of egg weight loss is a critical indicator for evaluating the freshness of eggs ^[1] and is associated with the economic value of eggs. Egg weight loss is attributed to the loss of moisture and carbon dioxide through shell pores ^[2]. The rate of gaseous and moisture escape from the shell pores during storage depends on the storage environment (temperature 4 °C or >27 °C, relative humidity, and air flow). Previous studies ^{[3][4]} revealed that egg weight loss is influenced by storage time and temperature. Extended storage time at ambient temperature increases the loss of moisture and gases from the egg to the environment ^[5]. It has been reported that egg weight loss is higher under ambient temperature compared to refrigerated temperatures ^{[6][3][4]}. The lower egg weight loss observed at refrigerated temperatures could be linked to the fast drying and shrinkage of the cuticle plugging air pores in the eggshell. In addition, there is less loss of solvents (moisture and gaseous products) from egg contents, in contrast to storage at ambient temperature, at which the size of air pores increases, facilitating the escape of moisture and carbon dioxide ^[7]. However, Samli et al. ^[8] reported that cold storage did not affect egg weight loss. Reduced rates of egg weight loss during storage helps maintain the internal quality of eggs during extended storage periods. The escape of gases and moisture through shell pores, leading to egg weight loss, also causes changes in albumen pH.

2. Albumen pH

Albumen pH (ApH) is a helpful indicator for evaluating changes in albumen quality over storage time. ApH is about 7.6 in fresh eggs, and the optimum pH of albumen ranges from 7.5–8.50 ^[9]; however, it increases during the storage period and reaches 9.5. Previous studies have shown that ApH increases with extended storage time ^{[10][11]} and storage temperatures ^{[9][12]}. Albumen pH increases more at room temperature than at low temperatures ^{[13][14]}. Škrbić et al. ^[15] reported that refrigerated temperatures lower the rate of increase in albumen pH compared to ambient temperature. However, Altunatmaz et al. ^[16] found no significant effect of temperature on albumen pH after 28 d of storage. The increased pH at room temperature could be due to the increased escape of gases from eggs.

This increase could be due to the dissociation of carbonic acid (H₂CO₃) leading to the formation of water and CO₂ within the albumen and the escape of CO₂ into the environment via the shell pores. This leads to the alkalization of the albumen and a shift in the bicarbonate buffer equilibrium ^[17]. The increased alkalinity of albumen causes a decrease in ovomucin content ^[18], and the highly viscous thick albumen adjacent to the yolk progressively loses its gelatinous structure, leading to albumen liquefaction ^[19]. Albumen pH influences the strength of the vitelline membrane, and increased pH values weaken the vitelline membrane, which may facilitate a high loss of gas and moisture from eggshell pores. Feddern et al. ^[5] reported that increased ApH negatively affects the vitelline membrane and hastens the exchange of albumen alkaline ions with yolk H⁺, leading to protein denaturation. All of these factors cause a decline in albumen quality, and the decline

is more conspicuous at ambient temperature than at low temperatures, irrespective of time; hence, ApH is a function of storage time and temperature. Albumen thinning due to increased pH levels is reflected in albumen height.

3. Albumen Height

Albumen height (AH) is fundamental for calculating the albumen index and Haugh unit values used for albumen quality evaluation. Albumen height is at a maximum in freshly laid eggs and declines with storage time. Various studies [11][20] have demonstrated that AH decreases with storage time and, in one study, AH and albumen index could not be calculated after 18 days of storage owing to increased albumen fluidity [16].

In all 20 studies examined, there was a 100% indication that albumen height decreased significantly during storage relative to the albumen height of fresh eggs. The reduction in albumen height during storage was due to liquefaction of the thick albumen. A decrease in thick albumen height was evident in eggs stored at room temperature compared with those stored at low temperatures [10][11][14]. Albumen proteins, which are fundamental to AH, are decreased quantitatively during storage due to temperature effects [18][21], thereby contributing to the decreased AH observed during storage. The other albumen indices were also affected by storage. Extended storage time decreased albumen weight [10][22] and albumen percentage [23]. This decrease in albumen weight was due to a decrease in the thick albumen weight and an increase in yolk weight, suggesting that water diffused from the albumen through the vitelline membrane into the yolk during storage. Albumen weight loss is higher at room temperature than under low temperatures [16] because of higher water loss from the albumen to the yolk. A drastic reduction in AH over storage time indicates poor albumen quality and adversely influences Haugh unit value (HU) values.

4. Haugh Unit Value

It has been established that albumen quality and egg freshness can be measured based on Haugh unit (HU) values, and HU is determined from thick albumen height and egg weight.

Various studies [5][24][14][25] have reported lower HU values in stored eggs than in fresh eggs. The following are reported comparisons of the HU values of fresh and stored eggs: 82.17 vs. 0 for eggs stored at 21 °C for 30 days [14], 79.21 vs. 62.82 for eggs stored for at 30 °C for 15 days [25], 98.6 vs. 39.36 for eggs stored at 33 °C for 63 days [5], and 83.5 vs. 0 for eggs stored at 25 °C for 56 days [24]. The decline in HU values of stored eggs is due to the disintegration of the ovomucin–lysozyme complex, proteolysis of dense proteins, and consequent reduction in the height of thick albumen [26]. The decrease in HU values was more rapid in eggs stored at ambient temperature than in eggs stored at refrigerated temperatures. The following are reported HU values of eggs at room and refrigerated temperatures when stored for 28 days: 35 vs. 74 [21], 32.66 vs. 71.6 [10], 32.71 vs. 56.41 [27] 38 vs. 64.77 [15], and 0 vs. 74.48 [16]. Although no decrease was observed at a temperature of 5 °C [10], this is in agreement with the reports of [5][27].

The quality of stored eggs is graded according to HU score value: AA grade > 72, A grade = 60–72, B grade < 60, and C grade < 30 [28]. The HU grade of eggs stored at refrigerated temperatures was found to be higher than that of eggs stored at ambient temperature: AA vs. 0 [16], A vs. C [15], and AA vs. 0 [24]. However, Souza et al. [13] reported that, after storage, eggs stored at both temperatures were classified as AA grade, although the HU value of eggs stored at refrigerated temperatures were numerically higher compared to those stored at ambient temperature. The higher HU value and grade of eggs stored at refrigerated temperatures indicates better egg quality, probably because albumen quality is preserved to a greater extent. The lower HU value of eggs stored at ambient temperature could be linked to faster degradation of the ovomucin–lysozyme complex and rapid liquefaction of dense albumen [4]. Hence, albumen thinning occurs more at room temperature than at low temperatures [29]. HU was found to be more stable with less variation under refrigerated conditions than at ambient temperature, suggesting that storage temperature may be a more critical factor influencing HU.

Decreased HU values during storage suggests loss of albumen functional properties. Albumen consistency was lost when the HU score was <70 during storage [30]. Eggs with lower HU values showed a significant reduction in the viscosity and elasticity modulus of ovomucin gel and disaggregation of O-glycoside bonds in ovomucin [31][32]. Therefore, it is crucial to maintain a high HU value during storage. HU values above 70 indicate low protein and lipid peroxidation and, hence, better albumen quality during storage and vice versa.

5. The Effects of Storage Time and Temperature on Functional Properties of the Albumen

The effects of storage time and temperature on albumen traits (ApH, AH, and HU) alter albumen structure and, consequently, influence the biological and functional properties of the albumen.

Albumen viscosity determines its functional properties, such as emulsification, whippability, and gelling properties [33]. Owing to storage time and temperature effects, changes in albumen structure may influence albumen viscosity; consequently, these functional properties are lost when albumen quality declines. The studies of [34][35] demonstrated that albumen viscosity decreases with storage time. Wang et al. [34] reported that, during an extended storage time, beyond a certain point, no changes were observed in viscosity measurement, showing that the gelatinous nature of the albumen had completely disappeared. In another study, eggs were stored at 24 °C for six weeks and the albumen viscosities of fresh and stored eggs was reported as 75.11 vs. 7.72 [42] and 60.46 vs. 5.73 [9]. Reduced albumen viscosity during storage may be attributed to the destabilization of the O-glycoside link between trisaccharides, collapse of the ovomucin gel structure [33], flow behavior index of the albumen over temperature ranges [36], and release of bound water molecules due to the hydrolysis of amino acid chains by the enzymes in the albumen. Reduced albumen viscosity corresponds to an increase in albumen fluidity, reflecting increased total soluble solids [37] and increased liquefaction of the yolk, which subsequently diffuses into the albumen. In addition, decreased viscosity due to storage time reduces the foaming properties of the albumen [9]. When albumen viscosity is lost, the thick albumen becomes less dense and causes albumen liquefaction (albumen thinning).

To a great extent, albumen thinning indicates a decline in egg quality. The reduction in the viscosity of thick albumen gel leads to deterioration in the gelling property and, consequently, albumen thinning [18][38]. Omana et al. [26] demonstrated that albumen thinning was evident at a later time of storage compared to the initial time, suggesting that there is a progressive loss of thick albumen gel with increasing storage time; hence, albumen thinning may be a function of storage time. In addition, albumen thinning has also been associated with the sulfhydryl (SH) groups of ovalbumin, which undergo transition and conversion during storage [39]. S-ovalbumin, a conformational isomer of native ovalbumin, was irreversibly transformed from ovalbumin during prolonged egg storage [40]. This change was due to the configurational inversion of amino acid residues, which is critical for the formation of thermo-stabilized ovalbumin [41]. A higher content of S-ovalbumin increases albumen liquefaction—a claim that is supported by the study of Huang et al. [40], which reported a negative correlation between HU value and S-ovalbumin content. This is in accordance with the study of Fu et al. [42], which revealed that a higher content of S-ovalbumin indicates a lower HU value and reduced freshness of eggs. In addition, albumen thinning may be due to reduced ovomucin–lysozyme interactions and degradation of ovalbumin or clusterin due to proteolysis associated with increased pH [26]. Yuceer and Caner [2] explained that protease enzymes, depolymerized by hydroxyl ions at increasing pHs, destabilize the ovomucin–lysozyme complex, eventually causing thinning of thick albumen and a decrease in HU values. Thus, albumen thinning is an intrinsic self-degrading property that causes changes in the content and structure of albumen during storage. For example, the ovomucin content is higher in dense, thick albumen than in less viscous albumen [34]. Alterations in the structure and composition of albumen proteins may also influence their functional properties.

Albumen protein content and structure tends to change during storage and such changes can reduce related functional properties. For instance, there was a decrease in ovomucin content after six weeks of storage, while no significant variation was observed for ovalbumin and lysozyme [43]. Similarly, Wang et al. [34] reported that increased storage time decreased ovomucin content in albumen, and a corresponding decrease in the viscoelasticity of albumen was observed. This is in agreement with the study of Shan et al. [44], which reported a decrease in rheological properties of albumen due to reduced ovomucin content. Ovalbumin structure (α - and β -sheets) is altered during storage, which reduces foaming and emulsifying properties [45]. Increased ApH during extended storage degrades ovomucin content, making it difficult to extract ovomucin [18]. These findings highlight that albumen proteins are sensitive to storage time and temperature. It is imperative to preserve the content and structure of these proteins to extend the shelf life of albumen without the loss of its functional properties.

The biological functions of albumen, such as antioxidant and antimicrobial activities, may be reduced because of alterations in albumen structure due to storage time and temperature effects. Free amino acids are some of the main contributors to the antioxidant activity of eggs; total antioxidant capacity (TAOC) tends to decline during storage [46]. Antioxidant capacity is more sensitive to storage temperature than storage time. Liang et al. [47] reported that the rate of decline of TAOC was slower at refrigerated temperatures than at ambient temperature. In another study, Nimalartane et al. [46] revealed that antioxidant capacity was very stable during six weeks of storage at refrigerated temperatures. This is likely because free amino acids, responsible for oxidative stability, are more stable at lower temperatures. This could

explain why lipid and protein peroxidation products are often higher in eggs stored at higher temperatures than at lower temperatures, indicating that a low storage temperature does not act as a catalyst for lipid and protein peroxidation.

Structural changes in albumen occur due to storage time and temperature effects, culminating in a decline in albumen quality, which is characteristic of lost functional and technological properties.

In conclusion, eggs are highly susceptible to deterioration during storage, owing to lipid peroxidation and protein denaturation. Strategies that would maintain albumen traits (AH, ApH, HU) in stored eggs similar to those of fresh eggs and help to inhibit biochemical reactions that lead to the collapse of albumen should be adopted. Albumen quality must be maintained during storage to extend its shelf life and increase the potential benefits to consumers and the food and health industries. The use of natural antioxidants that can be easily transferred to eggs and act as a natural shelf-life extender without residue effects is recommended. Investigating natural diets to preserve the internal quality of eggs may offer better chances of consumer acceptance and reduce storage costs.

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