

NIR Spectroscopy for Rapid Freshness Assessment and Quality Classification of Chicken Eggs

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ABSTRACT

Eggs undergo significant alterations during storage, which results in a loss of quality. To keep an eye on the freshness and quality of the eggs, it is essential to predict these changes. This study's objective was to assess the use of visible Near infrared (NIR) spectroscopy, which is a quick, non-destructive, online method for evaluating the quality of eggs. During the study six hundred sixty whole fresh eggs with white shells produced by the same group of hens fed a typical feed was acquired. They were placed in temperature-controlled environments whose temperature was 20°C and 30°C respectively and observed their spectra for 25 days of storage. The spectra of 40 eggs were collected for 0, 4,7,10,14,17,19,21,25 days within the NIR spectral range of 902 to 1810 nm; the absorption spectrum data was found to be collected for every 4nm span. The spectral non-destructive data was contrasted with the Haugh Units (HU) of the egg sample in terms of freshness and to the quantity of storage days in terms of quality. This study explores the potential of Near-Infrared (NIR) spectroscopy combined with chemometric analysis for non-destructive egg quality assessment, focusing on predicting Haugh Units (HU) and storage duration. The research involved systematic data collection, preprocessing of NIR spectra, and developing predictive models using Partial Least Squares (PLS) regression. Results demonstrated a high accuracy in predicting HU values and storage duration, with an R² value of 0.986 for calibration. Eggs stored at 20°C maintained higher HU values than those stored at 30°C, significantly impacting freshness assessment. Principal Component Analysis (PCA) effectively reduced data dimensionality, enhancing model precision. Combining shell measurement areas and preprocessing techniques improved PLS-DA model performance, achieving up to 95.75% accuracy in classifying egg freshness. The findings highlight the practical application of NIR spectroscopy and chemometric analysis in the food industry for ensuring egg quality and freshness.

Keywords: Classification of eggs, Freshness of chicken eggs, NIR Spectroscopy, Non-destructive method, MSC, SNV, SG 1st Derivate, PLS-R, SVM-R, PLS-DA, SVM-C.

INTRODUCTION

Eggs constitute a crucial component of human diets, rich in essential nutrients such as protein, vitamins, omega-3 fatty acids, lutein, and selenium (Suktanarak and Teerachaichayut, 2017). India stands as a significant

player in global agricultural production, particularly in the domains of egg and meat production. According to the latest data from the Food and Agriculture Organization Corporate Statistical Database (FAOSTAT) for the year 2020, India secures the 3rd position in global egg production and the 8th position in meat production. Over the years, there has been a substantial growth in egg

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production within the country, witnessing a notable increase from 78.48 billion in the fiscal year 2014-15 to a substantial 129.60 billion in the fiscal year 2021-22. Due to their affordability and being a readily accessible source of animal-based protein, eggs boast a significant consumption rate of 7.77 kg/capita as of 2017.

The economic feasibility and nutritional value of eggs, however, have led to a concerning rise in fraudulent activities (Hosen, S. Z., Swati, P., & Dibyajyoti, S., 2013), notably the deceptive alteration of information pertaining to egg quality. Instances of mislabeling, substitution, and the sale of stale eggs as fresh have been reported with increasing frequency. The advent of the COVID-19 pandemic has further complicated matters, disrupting supply chains, amplifying demand, and subsequently causing a surge in costing. This volatile situation establishes a setting ripe for fraudulent activities, even within the context of relatively low-cost products like eggs (Puertas, G., Cazón, P., & Vázquez, M., 2023).

To combat these challenges, there is a pressing need for the establishment of authenticity and the implementation of risk assessment protocols throughout the egg supply chain, from farms to supermarkets. Such measures necessitate the incorporation of advanced technologies that are not only sensitive but also rapid and cost-effective (Cruz-Tirado, J.P., et al, 2021).

Frauds in the egg industry commonly revolve around misrepresenting the laying date to extend the product's shelf life, resulting in stale eggs being sold as fresh (R. Mota-Grajales, J.C. Torres-Peña, J.L. Camas-Anzueto, M. Pérez-Patricio, R. Grajales Coutiño, F.R. López-Estrada, E.N. Escobar-Gómez, H. Guerra-Crespo, 2018). The conventional way of determining the freshness of eggs involves the Haugh unit (HU), introduced by Haugh in 1937. However, this method is not without its drawbacks, it is both invasive and time-absorbing, often relying on a limited quantity of specimens to extrapolate freshness across larger batches.

Traditional destructive methods of egg freshness detection have several drawbacks (Fu, Dandan, et al, 2022). Firstly, these methods result in the loss of the egg being tested, which leads to waste and increased costs, particularly in large-scale operations where many samples need to be tested. Secondly, destructive testing is time-consuming, as it involves breaking the egg, conducting the necessary tests, and cleaning up afterward. This process can slow down the overall workflow and delay decision-making. Additionally, these methods can be less efficient and require more labor and resources compared to non-destructive techniques. Destructive testing also prevents the possibility of further analysis on the same sample, limiting the scope of quality assessment. Furthermore, it is not environmentally friendly due to the waste generated from the broken eggs. Overall, the inefficiency, higher costs, and waste associated with traditional destructive methods make them less desirable for egg freshness detection compared to non-destructive alternatives.

The Near-Infrared (NIR) spectroscopic method offers several notable advantages for assessing egg freshness and quality (Cruz-Tirado, J.P., et al, 2021). This technique is non-destructive, allowing eggs to be tested without breaking them, thus preserving the samples for sale or further analysis and reducing waste and costs. NIR spectroscopy provides rapid and efficient analysis, delivering quick results that enable real-time quality assessment, which is especially beneficial in large-scale operations where time is critical (Loffredi, Eleonora, et al, 2021). Additionally, this method requires minimal sample preparation, streamlining the testing process and reducing the need for extra labor and resources. NIR spectroscopy also allows for multicomponent analysis, simultaneously measuring multiple attributes such as moisture content, protein levels, and freshness indicators like Haugh Units, making it a comprehensive and practical tool for egg quality assessment.

The possible application of NIR (near-infrared) spectroscopy to Haugh unit estimation, distinguish between fresh and stale eggs, and predict storage times has been underscored by various studies (Loffredi, Eleonora, et al, 2021). Notably, recent years have witnessed a surge in the development and commercial availability of affordable, transportable, and compact NIR spectrometers, offering the potential for on-site analysis at various points along the egg supply network (Dong, Xiaoguang, Xiuying Tang, et al, 2018).

Scientific literature has extensively explored the capabilities of portable NIR spectrometers, demonstrating their efficacy in analyzing agricultural food products. Unlike their benchtop counterparts, portable NIR devices can be seamlessly integrated on site and easily used for on-site analysis, proving particularly valuable in the dynamic egg supply chain.

However, the effective deployment of handheld Near-Infrared devices (Cruz-Tirado, J.P., et al, 2021) hinges on ensuring that the scaling down their components does not undermine their predictive capabilities compared to larger laboratory-grade NIR devices. Notably, researchers investigated the application of handheld Near-Infrared in estimating egg shelf life, achieving a promising R^2 value of 0.873 (Cruz-Tirado, J.P., et al, 2021). Despite this progress, the establishment and verification of analytical techniques leveraging handheld NIR for estimating the Haugh unit and classifying eggs based on freshness grading have yet to be comprehensively reported.

Numerous studies have been conducted on the evaluation of egg quality, with an emphasis on non-destructive methods (J. Zhang et al., 2023; E. Loffredi et al, 2021) for assessing several factors such as freshness, albumen pH, and Haugh unit. These methods include visible-near transmission spectra (H. Guo et al, 2022; J. P. Cruz-Tirado 2021), infrared spectroscopy (D. Fu et al, 2022), front-face fluorescence spectroscopy (E. Loffredi et al, 2021), and electronic nose-based devices (S. M. Yimenu, J. Y. Kim, and B. S. Kim, 2017).

Egg freshness and quality may be assessed using visible-near transmission spectra; prediction models have been successful in predicting characteristics such as air cell height, thick albumen height, and Haugh unit, with substantial correlation coefficients obtained. Like this, (D. Fu et al, 2022), research on visual transmission spectroscopy has produced strong correlation coefficients for predicting albumen pH and Haugh unit and has been investigated as a non-destructive technique for assessing individual egg freshness.

Research employing electronic nose-based devices has demonstrated potential for online assessment of egg freshness characteristics and storage day estimation. Prediction accuracy for metrics like the yolk factor and Haugh unit was increased by combining evolutionary algorithms with backpropagation neural networks (S. M. Yimenu, J. Y. Kim, and B. S. Kim, 2017).

There are several benefits to using spectroscopic (E. Loffredi et al, 2021) including quick and non-contact assessment, less physical touch required, and sanitary conditions guaranteed. Spectroscopy also makes it possible to grade many eggs quickly and effectively without requiring a lot of sample preparation time.

Furthermore, studies using NIR spectral data have shown that a number of egg quality indicators, including as laying days, air chamber size, egg pH, and weight loss, can be accurately predicted; strong R^2 values indicate that these predictions remain accurate even when applied to group averages (X. Dong et al., 2018). The spectroscopic techniques have been selected for their efficiency, accuracy, and potential for broad use in the egg business due to their quick and non-contact nature, as well as their capacity to precisely anticipate many factors related to egg quality. They provide a useful way to evaluate quality, making it possible to do so effectively and dependably without requiring destructive testing or a lot of sample preparation.

Considering these considerations, this investigation endeavors to assess the efficacy of an Economical,

Compact NIR spectrometer when coupled with artificial intelligence algorithms. The goal is to enable the real-time evaluation of Haugh units, and the grading eggs based on freshness, providing a valuable contribution to the field of egg quality assessment.

Material and Methodology

The following methods would become employed to carry out my research:

Egg Sample Collection:

A total of six hundred and sixty (660) freshly laid eggs, precisely one day old, were sourced for this study. These eggs exhibited a mean weight of 63.48 ± 2.56 grams, a height of 50.97 ± 1.53 millimeters, and a diameter of 37.34 ± 1.09 millimeters. The eggs, all non-fertilized and of white hen variety, were carefully procured from two distinct farms. Each farm contributed an equal share of 330 eggs, and these were sourced from the chickens of comparable years, brought up in identical feeding practices, and within similar circumstances around the environment. To maintain consistency, the eggs were transported with great care from the farms to the laboratory. Upon arrival, they underwent a meticulous disinfection process involving immersion in water at 42°C containing 50 parts per million (ppm) of chlorine for a duration of 1 minute. Subsequently, the eggs were allowed to air-dry for 5 minutes at room temperature. The collected egg samples were then randomly divided into two groups. The first group was stored at a temperature of 20°C , representing typical market conditions, while the second group was subjected to an accelerated storage temperature of 30°C . The storage durations included 0, 4, 7, 10, 14, 17, 19, or 21 days within a controlled chamber with a relative humidity ranging from 50% to 65%. For each analysis session, a subset of 80 eggs was randomly selected. This subset comprised 40 eggs from each storage temperature. Spectra acquisition and Haugh unit measurements were performed on these selected eggs.

The Haugh unit, a critical parameter for assessing egg freshness, was carefully measured during each analysis session. This comprehensive methodology ensures a detailed investigation into the implications of temperature during storage and duration on the quality attributes of the eggs, providing valuable insights into their shelf life and freshness under varied conditions.

NIR Spectral Data Collection:

The spectral analysis of the egg samples was conducted utilizing a DLPR NIRscan™ Nano portable spectrometer. This device operates within the wavelength range of 902–1810 nm, providing a comprehensive view of the near-infrared spectrum. Known for its ergonomic design, ease of transport, and adaptability for online measurements, this portable spectrometer offers practicality and versatility (H. Guo et al, 2022; J. P. Cruz-Tirado2021). The gathering of spectral data was carried out in absorbance mode after every 4nm span, utilizing a halogen lamp of 10-watt as the light origin and an In GaAs sensor for sensitivity.

The eggshell was systematically analyzed at three distinct locations: R1, R2 and R3 (J. P. Cruz-Tirado2021). This meticulous approach resulted in the acquisition of a spectral database comprising a total of 1980 spectra.

To enhance the robustness of the spectral data, data matrices were constructed by averaging spectra from different locations. Specifically, matrix creation took place for the averages of (R1 + R2), (R1 + R3), (R2 + R3), and (R1 + R2 + R3). This strategic approach aimed to assess whether combining measurements from multiple locations would yield improved results in subsequent classification and prediction models.

Given the expediency of both data acquisition and processing, the inclusion of additional measurements was deemed justifiable to enhance prediction accuracy. Consequently, a total of seven databases (each containing 660 spectra) were employed for the training and testing of prediction and categorization models. This

comprehensive approach allowed for a thorough exploration of the spectral characteristics of eggs, considering different shell locations and their collective impact on the predictive models' performance.

Evaluation of Haugh Units.:

Equation (1) was used to calculate each egg's Haugh unit (HU), which was defined by Haugh in 1937. The procedure included taking exact weight readings of the eggs and closely examining the albumen height. Initially, a high-precision digital scale with a 0.01-gram accuracy was used to weigh each egg. Then, using a micrometer with a resolution of 0.1 mm, the egg was carefully cracked onto a glass plate, and the height of the egg white was measured three times around the yolk, about 10 mm from the yolk.

The Haugh unit (HU) was determined through the application of the formula:

$$HU = 100 \log (h + 7.6 - 1.7w^{0.37}) \quad (1)$$

Here,

h represents the average height of the egg white in millimeters, and

W denotes the weight of the egg in grams.

Statistical Examination:

Spectra Preprocessing:

The preprocessing of spectroscopic Near-Infrared (NIR) data for chicken eggs is a critical phase in ensuring the quality, accuracy, and reliability of data when employing NIR spectroscopy for assessing egg quality. Various preprocessing techniques are employed to enhance spectral data, mitigate noise, and ready the data for robust modeling and analysis (Coronel-Reyes, J., Ramirez-Morales, I., Fernandez-Blanco, E., Rivero, D., Pazos, A., 2018). One key preprocessing step is baseline correction, vital for eliminating systematic variations in spectral information produced by instrumental consequences like scattering. Common methods for

baseline correction involve linear or polynomial fitting to rectify baseline shifts.

To address scattering effects, Multiplicative Scatter Correction (MSC) or Standard Normal Variate (SNV) (Brasil, Yasmin Lima, et al., 2022) techniques are often utilized. MSC is effective in compensating for multiplicative scattering, while SNV scales the spectra to achieve zero mean and unit variance, effectively reducing the impact of scattering effects.

Spectral data can inherently contain noise and fluctuations. To address this, smoothing techniques such as Savitzky-Golay or moving averages are applied (Guo, Hairong, et al, 2022). These techniques effectively reduce high-frequency noise while retaining essential information, ensuring that the processed data is more amenable to subsequent modeling and analysis. The careful application of these preprocessing techniques contributes significantly to the overall success and accuracy of NIR spectroscopy in evaluating chicken egg quality.

Estimation of HU:

Estimation models were constructed employing PLS regression. The objective was the Haugh Unit (HU) forecast values, serving as the response variables, for each individual egg according to the absorbance values derived from Near-Infrared (NIR) spectra used as input variables. The utilization of these regression techniques aimed to establish robust and accurate models for predicting the HU values, thereby contributing to the comprehensive assessment of egg quality using spectral data (Zhang, W., Pan, L., Tu, S., Zhan, G., & Tu, K., 2015).

Egg Categorization based on Freshness Ratings

In the classification of chicken eggs PLS-DA, or partial least squares discriminant analysis, was used in the investigation, a chemometric approach widely used for sample classification based on spectroscopic data. The theoretical analysis involves several key components

inherent to PLS-DA. Latent Variables (LV) play a crucial role in this method, representing the maximum covariance between the spectral data (predictor variables) and the class labels (response variables). Loading plots and score plots are instrumental in understanding the relationships between original variables, particularly wavelengths, and the derived latent variables. Regression coefficients guide the contribution of each variable to the prediction of class membership. The classification process begins with data preprocessing, where raw spectral data undergoes transformations like Standard Normal Variate (SNV) (Fu, Dandan, et al., 2022) and Savitzky-Golay smoothing. Calibration of the model follows, utilizing a dataset with known class labels, and the quantity of principal components is optimized through cross-validation. Validation and evaluation assess the model's robustness and performance metrics such as sensitivity, specificity, accuracy, and error rates.

The theoretical considerations in the context of egg classification involve understanding how latent variables capture spectral variations relevant to discriminating between fresh and stale eggs (Liu, Chengkang, et al., 2022). Loading plots aid in identifying key spectral features contributing significantly to the classification, highlighting regions where molecular changes associated with egg freshness occur. Decision boundaries established by PLS-DA in the latent variable space delineate regions corresponding to different classes, facilitating the assignment of samples to their respective categories. This theoretical analysis underscores the interpretative aspects of PLS-DA, unveiling spectral characteristics crucial for non-destructive quality assessment and effective egg classification (Akowuah, Thomas O., et al., 2020).

The dataset was split up into two groups: the training dataset, constituting 70% of the spectra, was utilized for model development and internal cross-validation, while the remaining 30% formed the prediction (external test) set. The PLS Regression model for the classification and

estimation of egg quality demonstrated robust performance across various metrics (Akowuah, Thomas O., et al., 2020). The models were assessed using R^2 for calibration, cross-validation, and prediction, where high values indicated strong fitting and predictive capabilities. RMSE values for model assessment stages were low, signifying accurate model predictions. Relative error and the Ratio of Performance to Deviation (RPD) provided normalized measures of prediction accuracy, with RPD values suggesting satisfactory predictive ability. Sensitivity, specificity, and overall accuracy were considered in classification tasks, showing the models' proficiency in discriminating between freshness classes. The incorporation of specific spectral regions and preprocessing techniques further enhanced model performance. Overall, the PLS-R models exhibited promising results, indicating their potential as effective tools for the non-invasive evaluation of egg quality utilizing Near-Infrared spectroscopy.

Experimentation and Result:

Egg Freshness Evaluation:

The maturation process in eggshells initiates immediately after laying, triggering physicochemical transformations that impact the sensory and operational quality of eggs (Zhang, Jingwei, et al, and 2023). Storage exacerbates these changes, leading to H_2O and CO_2 permeation through the eggshell, elevating acidity, and altering albumen consistency (Guo, Hairong, et al, 2023). Additionally, the interplay involving ovomucin and lysozyme influences the depth of the egg white layer during storage, providing a basis for estimating egg freshness through the Haugh Unit (HU).

Fig. 1 illustrates the changes in Haugh Unit (HU) values and egg freshness classes over the storage period at temperatures of 20 °C and 30 °C. In Fig. 1 a significant decrease in HU value over time is evident, with eggs stored at 30 °C exhibiting a more pronounced decline compared to those at 20 °C, particularly noticeable after

14 days. This suggests that eggs stored at 20 °C can maintain freshness (HU > 60) for up to 14 days, while those stored at 30 °C remain fresh for only 10 days.

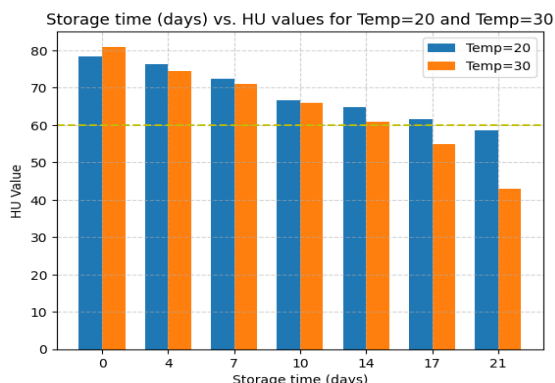


Fig.1: Storage Time vs Haugh Unit (at 20°C and 30°C)

However, eggs maintain a notably elevated Haugh Unit (HU) value when stored at 20 °C even on day 21 (Fig.1). Consequently, storing eggs at higher temperatures, especially in tropical climates, introduces greater variability in samples for regression models. This increased variation in response data may contribute to more accurate models, enhancing the reliability of RPD and RER measurements.

Spectra and PCA analysis:

Near-Infrared (NIR) spectra, unlike techniques such as Raman or Mid-Infrared (MIR) spectroscopy, are known for their lower information content, lacking distinct absorption peaks. Instead, NIR spectra exhibit a high correlation among various wavelengths. Principal Component Analysis (PCA) proves to be exceptionally useful in handling such datasets efficiently. PCA is adept at reducing the dimensionality of multidimensional data, especially when strong correlations are present. By projecting the original multidimensional dataset (in this case, 601 dimensions) into a lower-dimensional space, often just a few or two dimensions, PCA eliminates redundancy and retains essential information, making it a powerful tool for simplifying and interpreting complex NIR spectral data.

The PCA analysis conducted on the full spectra range of 902 to 1810 nm involved preprocessing using the Savitzky-Golay (S-G) derivative after SNV transformation. Fig Fig. 2 presents a comparison between original and preprocessed Near-Infrared (NIR) spectra for multiple samples, highlighting the impact of two different preprocessing techniques: Multiplicative Scatter Correction (MSC) and Standard Normal Variate (SNV). The top plot shows the original NIR spectra, characterized by significant variability in amplitude, particularly towards the right side of the plot, which can be attributed to differences in sample composition, path length, and scattering effects. The middle plot depicts the spectra after applying MSC preprocessing, which is designed to correct for multiplicative scattering effects and other physical variations. This results in a more aligned set of spectra, indicating reduced variability due to scattering. The bottom plot shows the spectra after SNV preprocessing, which standardizes each spectrum by removing the mean and scaling by the standard deviation. This technique further normalizes the data, reducing the impact of both multiplicative and additive scatter effects. The SNV-preprocessed spectra appear even more tightly clustered than the MSC-preprocessed spectra, suggesting a higher level of correction for variability. Overall, fig.2 demonstrates how preprocessing techniques like MSC and SNV can significantly enhance the quality and consistency of NIR spectral data, making it more suitable for subsequent analysis and modeling. Fig.3 illustrates the cumulative explained variance ratio as a function of the number of principal components used in a Principal Component Analysis (PCA) (Saman Abdanan Mehdizadeh, Saeid Minaei, Nigel H. Hancock, Mohamad Amir Karimi Torshizi, 2014). Fig. 3 reveals a sharp increase in the cumulative explained variance with the addition of the first principal component, suggesting that this component alone accounts for most of the variance in the data. Subsequent components contribute progressively smaller amounts to the explained variance,

with the cumulative variance quickly plateauing. By the time the second principal component is included, the cumulative explained variance reaches a near-maximum level, indicating that the first two principal components capture almost all the significant variance in the dataset. Adding more components beyond the second yields minimal additional explained variance, as evidenced by the flat curve for the remaining components. This plot highlights the effectiveness of PCA in reducing dimensionality, showing that a significant portion of the data's variance can be captured with just one or two principal components, simplifying the analysis without substantial loss of information. After the removal of outliers, the initial two principal components (PC1 and PC2) were found to capture 98% of the data's variance. Given the direct association between egg grading and the Haugh Units (HU) scale, this specific spectral region presented an opportunity to develop enhanced precision in predictive models, enhancing information analysis efficiency. Subsequently, narrowing down the wavelength range considerably enhanced the grouping of egg categories, as demonstrated by the PCA scores for average spectra following SNV and 1st S-G derivative processing. PC1 captured nearly 98% of the data's variability. This reduction in the spectral region contributed to enhanced class separation and more effective analysis.

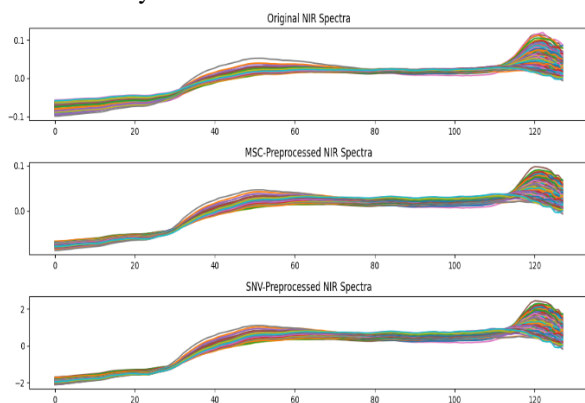


Fig.2: Preprocessed Spectra (MSC+SNV)

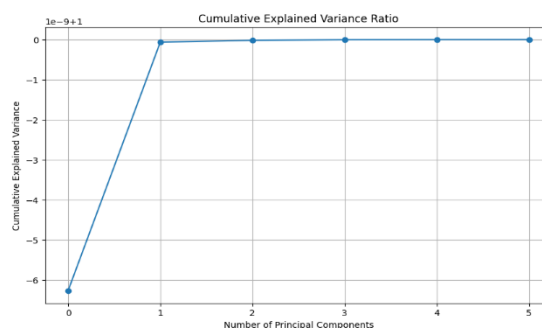


Fig.3: Principal Component Variance

Estimation of Haugh Units:

Table 1 presents the performance parameters of the PLS-Regression model for predicting the HU value of eggs stored at 20°C, considering different shell measurement areas. The latent variables (LV), preprocessing techniques (SNV+SG+1st Derivate) (Dong, Xiaoguang, Xiuying Tang, et al, 2018), and spectral range (902-1810 nm) are varied to evaluate their impact on the model's predictive capabilities. For each shell measurement area, the table includes key metrics such as R^2_c (coefficient of determination for calibration), R^2_{cv} (coefficient of determination for cross-validation), RMSEC (root mean square error for calibration), RMSECV (root mean square error for cross-validation), RPD (ratio of performance to deviation), and % Relative Error (Zhang, Jingwei, et al, 2023). These metrics assess the model's accuracy, precision, and reliability. Generally, the models exhibit high coefficients of determination (R^2_c and R^2_{cv}), low root mean square errors (RMSEC and RMSECV), and satisfactory RPD values, indicating their effectiveness in predicting HU values for eggs at 20°C. The Mean R1+R2+R3 model stands out with the highest R^2_c (0.986) and RPD (5.743), demonstrating superior predictive performance. Fig.4 is indicated by a red cross, highlighting the optimal number of components that minimize the MSE. Beyond this point, the MSE starts to increase, suggesting that adding more components leads to overfitting and a degradation in model performance. The plot helps in selecting the

optimal number of components for the PLS model to achieve the best predictive performance. In Fig. 5 the high R^2 value from cross-validation (0.9625) indicates that the model has a strong predictive capability, explaining approximately 96.25% of the variance in the measured HU values based on the predicted HU values. The closeness of the data points to the blue line and the high R^2 value suggests that the model predictions are highly

accurate. These results suggest that the combination of specific shell measurement areas, preprocessing methods, and spectral ranges significantly influences the PLS-R (Barker, M., & Rayens, W. 2003) model's ability to predict HU values, with potential applications in assessing egg quality.

Table 1: Evaluation Metrics for PLS-R Model Predicting HU in Eggs at 20°C

Shell Measurement Area	LV	Preprocessing	Spectral Range	R^2_c	R^2_{cv}	RMSEC	RMSECV	RPD	% Relative Error
R1	8	SNV+SG+1 st Derivate	902-1810 nm	0.984	0.963	1.282	1.953	5.165	3.091
R2	6	SNV+SG+1 st Derivate	902-1810 nm	0.979	0.961	1.455	1.991	5.066	3.152
R3	7	SNV+SG+1 st Derivate	902-1810 nm	0.982	0.964	1.370	1.919	5.258	3.037
Mean R1+R2	8	SNV+SG+1 st Derivate	902-1810 nm	0.985	0.969	1.223	1.788	5.641	2.830
Mean R1+R3	7	SNV+SG+1 st Derivate	902-1810 nm	0.985	0.967	1.256	1.835	5.438	2.904
Mean R1+R2+R3	8	SNV+SG+1 st Derivate	902-1810 nm	0.986	0.970	1.196	1.756	5.743	2.780
Mean R2+R3	7	SNV+SG+1 st Derivate	902-1810 nm	0.983	0.968	1.310	1.795	5.621	2.841

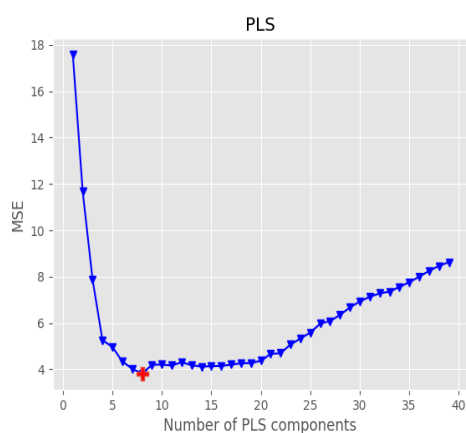


Fig.4: Number of PLS components (LV) for minimum MSE

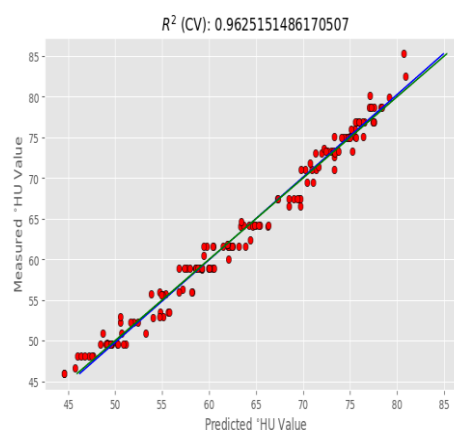


Fig.5: Predicted HU value vs Measured HU value graph.

Eggs discrimination using freshness index:

The PLS-DA models developed for the classification of hen's eggs based on freshness utilized different shell measurement areas, latent variables (LV), preprocessing techniques, and spectral ranges. In region R1, with 8 LV (Table 1), employing Standard Normal Variate (SNV) transformation, Savitzky-Golay (SG) smoothing, and first derivative preprocessing over the spectral range of 902-1810 nm, the model exhibited an accuracy of 93.75% in distinguishing freshness classes. Similarly, regions R2 and R3, with 6 and 7 LV, respectively, applying the same preprocessing methods and spectral range, demonstrated accuracies of 90.86% and 93.06% (Table 2). Combining measurement regions, such as Mean R1+R2, Mean R1+R3, Mean R2+R3, and Mean R1+R2+R3, further enhanced accuracy, reaching 95.7%, 89.00%, 91.25%, and 95.75%, respectively (Table 2).

Table 2: Parameters of the PLS-DA Model for Hen's Egg Classification Based on Shell Measurement Location:

Shell Measurement Area	LV	Preprocessing	Spectral Range	Freshness Accuracy
R1	8	SNV+SG+1 st Derivate	902-1810 nm	93.75%
R2	6	SNV+SG+1 st Derivate	902-1810 nm	90.86%
R3	7	SNV+SG+1 st Derivate	902-1810 nm	93.06%
Mean R1+R2	8	SNV+SG+1 st Derivate	902-1810 nm	95.7%
Mean R1+R3	7	SNV+SG+1 st Derivate	902-1810 nm	89.00%
Mean R1+R2+R3	8	SNV+SG+1 st Derivate	902-1810 nm	95.75%
Mean R2+R3	7	SNV+SG+1 st Derivate	902-1810 nm	91.25%

Discussion:

The scalability of NIR spectroscopy for commercial use in egg quality assessment offers substantial advantages, but it also faces several potential barriers that need to be addressed. From the experiments and results discussed, the non-destructive nature of NIR spectroscopy emerges as a key advantage. This method allows for the assessment of egg quality without damaging the product, which is crucial for maintaining the integrity of commercially processed eggs. The rapid analysis capability of NIR spectroscopy, as evidenced by the significant detection of changes in Haugh Units (HU) over different storage periods, is particularly beneficial for high-throughput environments such as egg processing facilities. This speed can significantly enhance productivity by enabling the quick sorting and grading of eggs.

Moreover, the potential for automation with NIR spectroscopy is notable. NIR spectrometers can be integrated into automated systems, including conveyor belts and robotic arms, to provide continuous analysis of eggs on production lines. This integration ensures consistent and efficient quality control. The minimal sample preparation required for NIR spectroscopy also adds to its convenience in commercial settings, where time and labor efficiency are critical. Preprocessing techniques such as Multiplicative Scatter Correction (MSC) and Standard Normal Variate (SNV), as used in the experiments, help to normalize data and reduce variability, streamlining the process further.

The predictive accuracy of NIR spectroscopy models is another significant advantage. The experiments demonstrated high coefficients of determination (R^2_c and R^2_{cv}), low root mean square errors (RMSEC and RMSECV), and satisfactory ratio of performance to deviation (RPD) values. For instance, the Mean R1+R2+R3 model achieved an R^2_c of 0.986 and an RPD of 5.743, the outcomes were just more reliable than earlier

findings using tabletop NIR spectrometers. Dong et al. 2018 predicted the HU using the VIS/NIR range (340–1030 nm), with an R^2 of RMSEP = 7.72 to 12.15 and smaller $p = 0.717$ to 0.881 . Similar results were found by Suktanarak et al. 2018 when they examined the prediction of the HU value using VIS/NIR (588–1091 nm) and FT-NIR (1000–2500 nm) spectrometers, with R^2 $p = 0.91$ (RMSEP = 5.64) and R^2 $p = 0.83$ (RMSEP = 7.11), respectively. In their evaluation of ANN models based on NIR spectra in the 1000–2500 nm region (Lin et al. 2011) found that the models performed well for predicting egg freshness, with R^2 $p = 0.879$ and RMSEP = 2.44. Conversely, Kempes et al. 2007 and Abdel-Nour et al. 2011 investigated VIS/NIR spectrometers to create PLSR models. Indicating superior predictive performance. Such high predictive accuracy enhances decision-making in quality control processes, allowing for reliable assessment of egg freshness and quality.

However, several barriers to the implementation of NIR spectroscopy in commercial settings exist. The initial cost of purchasing and installing NIR spectroscopy equipment can be substantial. This includes costs for spectrometers, integration with existing production lines, and necessary software for data analysis and model calibration. Additionally, implementing and maintaining NIR spectroscopy systems requires specialized knowledge, which means companies may need to invest in training personnel or hiring experts, thereby increasing operational costs.

Developing accurate calibration models is crucial for reliable results, and this process can be time-consuming and data-intensive. Accurate calibration requires substantial data from various egg samples to ensure the robustness and generalizability of the models. Furthermore, natural variability in eggs, such as differences in shell thickness, color, and internal composition, can affect the accuracy of NIR spectroscopy. Advanced preprocessing and calibration

techniques are necessary to address this variability, which adds complexity to the implementation process.

Environmental factors, such as temperature, humidity, and lighting conditions, can also impact the performance of NIR spectroscopy. Consistent environmental conditions must be maintained in a commercial setting to ensure accurate measurements. For example, the experiments showed that eggs stored at 30°C exhibited more pronounced declines in HU values compared to those stored at 20°C , highlighting the importance of controlled storage conditions.

To overcome these barriers, companies can implement NIR spectroscopy in phases, starting with pilot projects to evaluate feasibility and refine calibration models before scaling up to full production. Collaborating with technology providers, research institutions, and industry experts can help address technical challenges and reduce the learning curve associated with adopting new technology. Investing in training programs for staff can build the necessary expertise in-house, reducing reliance on external consultants and ensuring smooth operation and maintenance of the systems. Developing robust and adaptive calibration models that account for variability in egg characteristics and environmental conditions is crucial, with continuous updating and validation of these models enhancing accuracy and reliability. Engaging with regulatory bodies early in the implementation process can help ensure that NIR spectroscopy systems meet all necessary standards and requirements, facilitating smoother approval and compliance.

Conclusion:

In conclusion, this study has demonstrated the potential of Near-Infrared (NIR) spectroscopy combined with chemometric modeling as a powerful tool for the non-destructive assessment of egg quality, with a particular focus on predicting Haugh Units (HU) and storage duration. The research encompassed a systematic approach to data collection, preprocessing of NIR spectra,

and the development of predictive models for these essential egg quality attributes. The results indicated that the models, particularly the Partial Least Squares (PLS) regression, could predict storage duration with a high degree of accuracy, showcasing an R^2 value of 0.986 for the calibration dataset. The results suggest that NIR spectroscopy, in conjunction with chemometric modeling, offers a promising and practical solution for assessing egg quality attributes. The study's success in predicting storage duration and Haugh Units underscores the potential of this approach for applications in the food industry, ensuring the quality and freshness of eggs in various production processes. The Mean R1+R2+R3 area

shows consistently high freshness accuracy at 95.75%, indicating that combining measurements from the bottom, middle, and top areas provides an effective approach for predicting egg freshness. R1 (Bottom) and R3 (Top) areas individually exhibit high accuracy, emphasizing their significance in assessing egg quality. The use of eight latent variables consistently appears in the analysis, suggesting that this number optimally captures the relevant information for the predictive models. The specified preprocessing techniques (SNV, SG, and the first derivative) contribute to improving the accuracy of freshness predictions.

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التحليل الطيفي بالأشعة تحت الحمراء القريبية لتقييم سريع لنضارة البيض وتصنيف جودته

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ملخص

يتعرض البيض لتغيرات كبيرة أثناء التخزين، مما يؤدي إلى فقدان الجودة. لمراقبة نضارة البيض وجودته، من الضروري التنبؤ بهذه التغيرات. كان هدف هذه الدراسة تقييم استخدام التحليل الطيفي بالأشعة تحت الحمراء القريبية المرئية، وهي طريقة سريعة وغير مدمرة عبر الإنترنت لتقييم جودة البيض. خلال الدراسة، تم الحصول على ستمائة وستين بيضة طازجة كاملة بقرشور بيضاء أنتجت نفس مجموعة الدجاج التي تتغذى على علف نموذجي. تم وضعها في بيئات يتم التحكم في درجة حرارتها حيث كانت درجة الحرارة 20 درجة مئوية و30 درجة مئوية على التوالي وتم ملاحظة أطياها لمدة 25 يومًا من التخزين. تم جمع أطياها 40 بيضة لمدة 0 و4 و7 و10 و14 و17 و19 و21 و25 يومًا ضمن النطاق الطيفي للأشعة تحت الحمراء القريبية من 902 إلى 1810 نانومتر؛ ووجد أن بيانات طيف الامتصاص تم جمعها لكل فترة 4 نانومتر. تمت مقارنة البيانات الطيفية غير المدمرة بوحدات هاو (HU) لعينة البيض من حيث النضارة وكمية أيام التخزين من حيث الجودة. تستكشف هذه الدراسة إمكانات التحليل الطيفي للأشعة تحت الحمراء القريبية (NIR) جذبًا إلى جنب مع التحليل الكيميائي لتقييم جودة البيض غير المدمرة، مع التركيز على التنبؤ بوحدات هاو (HU) ومدة التخزين. تضمن البحث جمع البيانات بشكل منهجي، والمعالجة المسبقة لأطياها الأشعة تحت الحمراء القريبية، وتطوير نماذج تنبؤية باستخدام الانحدار الجزئي للمربعات الصغرى (PLS). أظهرت النتائج دقة عالية في التنبؤ بقيم وحدات هاو ومدة التخزين، مع قيمة R^2 تبلغ 0.986 للمعايرة. احتفظت البيض المخزنة عند درجة حرارة 20 درجة مئوية بقيم HU أعلى من تلك المخزنة عند درجة حرارة 30 درجة مئوية، مما أثر بشكل كبير على تقييم النضارة. قلل تحليل المكونات الرئيسية (PCA) بشكل فعال من أبعاد البيانات، مما عزز دقة النموذج. أدى الجمع بين مناطق قياس القشرة وتقنيات المعالجة المسبقة إلى تحسين أداء نموذج PLS-DA، حيث حقق دقة تصل إلى 95.75% في تصنيف نضارة البيض. تسلط النتائج الضوء على التطبيق العملي لتحليل الطيف بالأشعة تحت الحمراء القريبية والتحليل الكيميائي في صناعة الأغذية لضمان جودة البيض ونضارته.

الكلمات الدالة: تصنيف البيض، نضارة بيض الدجاج، التحليل الطيفي بالأشعة تحت الحمراء القريبية، الطريقة غير المدمرة، SVM-C، PLS-DA، SVM-R، PLS-R، الأول، SG، مشتق، SNV، MSC.

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