

# Advanced Chemometric and Machine Learning Techniques in NIR Spectroscopy for Freshness Class Prediction of Chicken Eggs

Priti Prakash Patil<sup>1</sup>, Dr. V. N. Patil<sup>2</sup>

<sup>1</sup>Research Scholar, SVPM's College of Engineering Malegaon BK Baramati, Savitribai Phule Pune University Pune, India, [prtipatil87@gmail.com](mailto:prtipatil87@gmail.com)

<sup>2</sup>Research Guide, SVPM's College of Engineering Malegaon BK Baramati, Savitribai Phule Pune University Pune, India, [vvnnpp2002@gmail.com](mailto:vvnnpp2002@gmail.com)

The quality of eggs deteriorates during storage, making freshness monitoring crucial in the egg industry. This study explores the use of Near-Infrared (NIR) spectroscopy, a rapid and non-destructive method, combined with chemometric techniques to assess and classify egg freshness effectively. A total of 660 eggs were stored at controlled temperatures (20°C and 30°C) and observed for 21 days, with spectral data collected across a range of 902-1810 nm at 4 nm intervals. The freshness was analyzed in relation to Haugh Units (HU) and days of storage. To enhance data quality, preprocessing methods such as Multiplicative Scatter Correction (MSC), Standard Normal Variate (SNV), and Savitzky-Golay smoothing were applied. Dimensionality reduction through Principal Component Analysis (PCA) helped streamline data, while predictive models, including Partial Least Squares Regression (PLSR) and Support Vector Machine Regression (SVM-R), were developed to estimate HU values accurately. The study achieved an  $R^2$  of 0.997 in calibration, indicating strong predictive power, especially with SVM-R. Additionally, classification models Partial Least Squares Discriminant Analysis (PLS-DA) and Support Vector Machine Classification (SVM-C)—achieved up to 99.20% accuracy in distinguishing between different freshness levels. These findings underscore NIR spectroscopy's potential as a reliable tool for real-time quality monitoring, offering efficiency, accuracy, and non-destructive testing for the egg industry. This approach could serve as a valuable alternative to conventional freshness assessments, providing precise insights into egg quality during storage, thereby ensuring better quality control and consumer satisfaction in the food industry.

**Keywords:** Classification of eggs, Freshness of chicken eggs, NIR

Spectroscopy, Nondestructive method, MSC, SNV, SG 1st Derivate.

## **1. Introduction**

The increasing number of laying hens and better hen diets are driving major changes in the egg industry, including higher output. Despite the fact that these advancements have made egg production more affordable, the industry still has to contend with maintaining egg quality and satisfying customer demands [1]. The egg grade is determined by both internal and external variables. It is believed that an egg's albumen and yolk quality share internal characteristics. Haugh Units (HU) and albumen pH are two commonly used measures for assessing these characteristics [2]. Conventional methods of assessing egg quality usually depend on visual inspection and lab-based analysis, which may be time-consuming, tedious, and prone to errors. Therefore, the company needs accurate, non-destructive, and efficient ways to assess egg quality [3].

Numerous non-destructive techniques have been studied in response to the demand for better egg quality screening. These techniques seek to offer unbiased assessments while lessening the time and resource requirements of conventional methods. Examples of such technologies include the use of Electronic Nose (EN) [5] systems that use scent sensors to determine the freshness of eggs, magnetic resonance imaging (MRI) [4] for evaluating the microanatomy of eggs, and computer image analysis to predict the albumen to yolk ratio without cracking the egg. Additionally, the Near-Infrared Fourier transform (FT-NIR) spectroscopy has been used to determine the height of the thick albumen [6], demonstrating a good degree of agreement between measured values and spectral data.

These techniques have the potential to improve the evaluation of egg quality. In order to provide trustworthy and impartial methods for assessing egg quality, researchers have looked to spectroscopic techniques, such as Near-Infrared (NIR) spectroscopy [7], in conjunction with sophisticated data analysis methods [8], after assessing the shortcomings of the previously stated non-invasive procedures. Using multivariate analytic methods and NIR spectroscopy, this work offers a thorough methodology for evaluating egg quality [9]. From gathering unfertilized eggs from a registered chicken farm to creating prediction models for important quality parameters, the research involves several steps.

To preserve their integrity, unfertilized eggs were first meticulously gathered and kept in a controlled atmosphere. The spectral characteristics of 30 undamaged chicken eggs were then recorded using a high-resolution spectrometer to gather NIR spectral data during a 22-day period. Strict preprocessing methods were used to improve the spectroscopic data quality once it was collected. Preprocessing techniques such as baseline correction, smoothing, and scattering effect correction were used to guarantee the correctness and dependability of the data. Principal Component Analysis (PCA) [8] and other multivariate calibration techniques were used to decrease the dimensionality of the spectral data and investigate underlying trends. The construction of strong prediction models was made possible by these methods, which were essential for feature selection, noise reduction, and data visualization.

To forecast the Haugh Units (HU) value, a gauge of egg freshness, using NIR spectral data, regression models based on Partial Least Square Regression (PLS-R) and Support Vector

Machine Regression (SVM-R) were created. Furthermore, SVM-C and Partial Least Squares Discriminant Analysis (PLS-DA) were used to classify eggs according to quality criteria [10].

In-depth spectrum analysis was also carried out in the study, which looked at the eggs' spectral properties across time and evaluated how well various preprocessing techniques improved the quality of the data. All things considered, this study shows how effective NIR spectroscopy and multivariate analytic methods may be as non-destructive methods of evaluating the quality of eggs. This work advances the creation of effective and impartial strategies for assessing eggs by fusing sophisticated spectroscopic technologies with reliable data processing methods.

## **2. Background and Related Work:**

In order to prolong the product's shelf life, frauds in the egg industry sometimes involve falsifying the laying date, which leads to stale eggs being marketed as fresh [28]. Developed by Haugh in 1937, the Haugh unit (HU) is the standard method used to assess the freshness of eggs. This approach does have some disadvantages, though; it is time-consuming and intrusive, and it frequently uses a small number of specimens to estimate freshness over larger batches. There are a number of problems with traditional destructive methods for detecting the freshness of eggs [29]. First of all, these techniques cause the egg being tested to be lost, which increases waste and expenses, especially in large-scale operations where several samples must be evaluated.

Second, because it requires cracking the egg, performing the required tests, and then cleaning up afterward, destructive testing takes a lot of time. Decision-making may be delayed and the workflow as a whole slowed down by this procedure. In addition, compared to non-destructive approaches, these technologies may be less effective and demand more work and money. Additionally, destructive testing restricts the breadth of quality evaluation by preventing additional investigation on the same sample. Additionally, the garbage produced by the broken eggs makes it unfriendly to the environment. All things considered, traditional destructive methods are less acceptable for detecting the freshness of eggs than non-destructive alternatives due to their inefficiency, increased expenses, and waste.

When evaluating the freshness and quality of eggs, the Near-Infrared (NIR) spectroscopic approach has a number of noteworthy benefits [10]. Because this method is non-destructive, eggs may be analyzed without being broken, saving samples for sale or additional analysis while also cutting expenses and waste. In large-scale operations when time is of the essence, NIR spectroscopy offers quick and effective analysis, yielding findings that allow for real-time quality monitoring [17]. Furthermore, this approach necessitates less sample preparation, which expedites the testing procedure and lessens the need for additional work and materials. NIR spectroscopy is a thorough and useful technology for evaluating the quality of eggs because it enables multicomponent analysis, which measures several characteristics at once, including moisture content, protein levels, and freshness indications like Haugh Units.

Numerous studies have highlighted the potential use of NIR (near-infrared) spectroscopy to

estimate Haugh units, differentiate between fresh and old eggs, and forecast storage durations [17]. The development and commercial availability of inexpensive, portable, and small NIR spectrometers has significantly increased in recent years, providing the opportunity for on-site investigation at several locations throughout the egg supply network [16]. The capabilities of portable NIR spectrometers have been well investigated in scientific literature, which has shown how effective they are in analyzing agricultural food items. Particularly useful in the dynamic egg supply chain, portable NIR instruments are easier to operate and may be readily integrated on site, unlike their benchtop equivalents.

In contrast to larger laboratory-grade NIR devices, the successful deployment of portable NIR devices [10] depends on making sure that the reduction in component size does not compromise their predictive power. Interestingly, researchers looked at using portable near-infrared to estimate the shelf life of eggs and found a good  $R^2$  value of 0.873 [10]. Notwithstanding these developments, there has not yet been a thorough study on the development and validation of analytical methods utilizing portable NIR for calculating the Haugh unit and categorizing eggs according to freshness grading.

Egg quality evaluation has been the subject of several research, with a focus on non-destructive techniques [15,17] for evaluating a number of variables, including freshness, albumen pH, and Haugh unit. These techniques include front-face fluorescence spectroscopy [17], infrared spectroscopy [27], visible-near transmission spectra [7,10], and electronic nose-based devices [5].

Visible-near transmission spectra may be used to evaluate the freshness and quality of eggs; prediction models have been successful in predicting traits like Haugh unit, thick albumen height, and air cell height, with significant correlation coefficients achieved. Visual transmission spectroscopy has been studied as a non-destructive method for determining the freshness of individual eggs and has yielded good correlation coefficients for predicting albumen pH and Haugh unit [27].

Studies using electronic nose-based devices have shown promise for online evaluation of egg freshness attributes and calculation of storage day. Combining backpropagation neural networks with evolutionary algorithms improved prediction accuracy for parameters such as the yolk factor and Haugh unit [5]. Using spectroscopic [17] has various advantages, including rapid and non-contact assessment, a reduced need for physical interaction, and a guarantee of hygienic settings. Additionally, spectroscopy allows for the efficient and rapid grading of several eggs without requiring a significant amount of sample preparation time.

A number of egg quality indicators, such as laying days, air chamber size, egg pH, and weight loss, can also be accurately predicted, according to studies employing NIR spectral data; strong  $R^2$  values suggest that these predictions hold true even when applied to group averages [16]. Due to their speed, non-contact nature, and ability to accurately predict a wide range of egg quality-related parameters, spectroscopic methods have been chosen for their efficiency, accuracy, and potential for widespread application in the egg industry. They offer a practical means of assessing quality, enabling reliable and efficient evaluation without the need for extensive sample preparation or destructive testing.

In light of these factors, this study aims to evaluate the effectiveness of a compact, affordable NIR spectrometer in conjunction with AI algorithms. The objective is to make it possible to

grade eggs according to their freshness and evaluate Haugh units in real time, making a significant contribution to the area of egg quality assessment.

3. Material and Methods:

The following methods were employed to carry out our research:

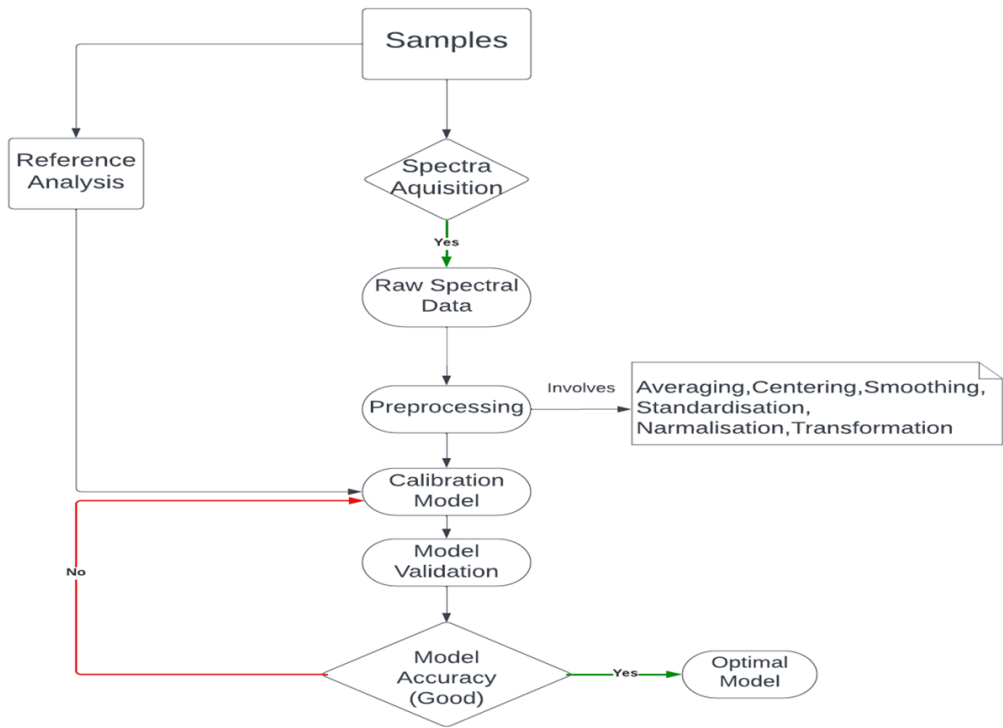


Figure 1. Proposed System Algorithm.

A. Egg Sample Collection:

In this study, a total of 660 freshly laid, one-day-old, non-fertilized white hen eggs were used. The eggs had an average weight of  $63.48 \pm 2.56$  grams, a height of  $50.97 \pm 1.53$  millimeters, and a diameter of  $37.34 \pm 1.09$  millimeters. To ensure consistency, the eggs were carefully transported to the laboratory. Upon arrival, they were disinfected by immersion in water at  $42^{\circ}\text{C}$  with 50 ppm of chlorine for 1 minute, then air-dried for 5 minutes at room temperature. The eggs were then randomly divided into two groups: one stored at  $20^{\circ}\text{C}$  (representing typical market conditions) and the other at  $30^{\circ}\text{C}$  to simulate accelerated storage conditions. Storage times were set at intervals of 0, 4, 7, 10, 14, 17, 19, and 21 days, in a controlled chamber with a relative humidity between 50% and 65%. For each analysis session, a random subset of 80 eggs was selected, consisting of 40 eggs from each storage temperature group. Both spectral analysis and Haugh unit measurements were conducted to assess egg quality and freshness, with the Haugh unit specifically used to

evaluate freshness over time. This methodology provides a thorough examination of how storage temperature and duration affect the quality and shelf life of eggs under controlled conditions.

#### B. NIR Spectral Data Collection:

A DLPR NIRscan<sup>TM</sup> Nano portable spectrometer was used to do the spectral analysis of the egg samples. This instrument provides a thorough picture of the near-infrared spectrum by operating in the wavelength range of 902–1810 nm. This portable spectrometer is well-known for its practicality and versatility, ergonomic design, and portability for online measurements [7,10]. Using a 10-watt halogen lamp as the light source and an In GaAs sensor for sensitivity, the spectral data collection was done in absorbance mode after every 4 nm span.

Three different locations R1, R2, and R3 were used for the methodical analysis of the eggshell [10]. A spectral database with a total of 1980 spectra was obtained as a consequence of this methodical approach. Data matrices were created by averaging spectra from many sites in order to improve the spectral data's resilience. In particular, the averages of (R1 + R2), (R1 + R3), (R2 + R3), and (R1 + R2 + R3) were created as matrices. This calculated tactic sought to determine whether integrating observations from several sites would enhance the performance of ensuing classification and prediction models.

Considering the speed of data collection and processing, it was judged reasonable to add more measures in order to improve forecast accuracy. For this reason, seven datasets of 660 spectra each were used to train and evaluate prediction and classification models. This all-encompassing method made it possible to investigate the spectral properties of eggs in detail, taking into account various shell positions and how they affect the prediction models' overall performance.

#### C. Reference Measurements of Freshness Using Haugh Units:

The Haugh unit (HU), developed by Haugh in 1937, was computed for each egg using equation (1). The process involved carefully measuring the albumen height and getting precise weight measurements of the eggs. Each egg was first weighed using a high-precision digital scale with an accuracy of 0.01 grams. The egg was then delicately broken onto a glass plate, and the height of the egg white was measured three times around the yolk, at a distance of around 10 mm, using a micrometer with a precision of 0.1 mm.

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

The mathematical equation was used to calculate the Haugh unit (HU) (1).

In this case,

H stands for the egg white's average height in millimeters.

W stands for the egg's weight in grams.



#### D. Preprocessing of spectrographic data:

When employing Near-Infrared (NIR) spectroscopy to evaluate the quality of chicken eggs, preprocessing is an essential step in guaranteeing data quality, accuracy, and dependability [10]. In order to improve the spectral data, lower noise, and get the data ready for reliable modeling and analysis, preprocessing techniques are used. To eliminate systematic changes in the spectrum data caused by instrumental factors like scattering, baseline correction is necessary.

Linear or polynomial fitting are common techniques for correcting the baseline shift. Usually, scattering effects in the data are corrected using Standard Normal Variate (SNV) [1] or Multiplicative Scatter Correction (MSC) [1] approaches. SNV reduces scattering effects by scaling the spectra to have zero mean and unit variance, whereas MSC accounts for multiplicative scattering. Noise and fluctuations may be present in spectral data. Savitzky-Golay and moving average are two smoothing methods that may be used to cut down on high-frequency noise while keeping important information intact.

#### E. Multivariate Calibration:

In chemometrics and data analysis, multivariate calibration is a potent analytical method that establishes a quantitative link between a number of variables, including spectral data or chemical measurements [21,22], and a certain feature or attribute of interest. This method is essential in many domains, such as process control, spectroscopy, and analytical chemistry. One essential method for analyzing complicated Near-Infrared (NIR) spectral data is Principal Component Analysis (PCA). It helps with feature selection, data exploration, and dimensionality reduction. By converting high-dimensional data into a collection of orthogonal principle components (PCs) that represent the most relevant variation, PCA aids in the management of high-dimensional data in the context of NIR spectroscopy.

It is especially helpful for handling multicollinearity, choosing meaningful spectral characteristics, and lowering noise. Furthermore, PCA offers data visualization tools, including as loadings plots and score plots, which make it possible to identify outliers and comprehend data trends. It is a crucial tool in many domains where NIR spectroscopy is utilized for data analysis and modeling as it may act as a first step for calibration models and makes data reduction easier.

#### F. Regression, Prediction and Classification Model:

The development of regression models for predicting Haugh Units (HU) value in eggs using Partial Least Square Regression (PLS-R) and Support Vector Machine Regression (SVM-R) [23] is a common practice in the analysis of Near-Infrared (NIR) spectral data. In this approach, the reflectance values from the NIR spectra serve as input variables, and the HU value, which is a measure of egg quality, serves as the response variable. PLS-R and SVM-R are sophisticated regression techniques that leverage the spectral data to establish a quantitative relationship between the spectral characteristics and the HU value. These models aim to provide accurate predictions of egg quality based on the NIR spectral information, offering a non-destructive and efficient way to assess egg freshness and quality.

Partial Least Squares Discriminant Analysis (PLS-DA) is a robust multivariate statistical technique widely employed for the classification of eggs based on their quality attributes, *Nanotechnology Perceptions* Vol. 20 No. S14 (2024)

such as freshness and Haugh Units (HU). In the context of egg classification, PLS-DA uses the spectral data from Near-Infrared (NIR) spectroscopy as input variables to distinguish between different egg classes, such as 'Class AA,' 'Class A,' and 'Class B,' or 'Fresh' and 'Stale.' By capturing the underlying spectral patterns associated with these classes, PLS-DA models can accurately classify eggs, providing a valuable tool for quality control in the egg industry. This non-destructive approach offers the advantage of real-time classification, enabling efficient sorting and grading of eggs based on their spectral characteristics. Another Classification model were developed using SVM-C algorithm to verify the adaptability of model for freshness detection and classification of chicken eggs.

#### **4. Experimentation and Result:**

##### **A. Egg Freshness Evaluation:**

The maturation process in eggshells initiates immediately after laying, triggering physicochemical transformations that impact sensory and operational quality of eggs [15]. Storage exacerbates these changes, leading to H<sub>2</sub>O and CO<sub>2</sub> permeation through the eggshell, elevating acidity, and altering albumen consistency [22]. Additionally, the interplay involving ovomucin and lysozyme influences the depth of egg white layer during storage, providing a basis for estimating egg freshness through the Haugh Unit (HU).

##### **B. Spectra and PCA analysis:**

In contrast to methods like Raman or Mid-Infrared (MIR) spectroscopy, near-infrared (NIR) spectra are characterized by their reduced information richness and absence of identifiable absorption peaks. Rather, there is a strong link between different wavelengths in NIR spectra. Effectively managing huge datasets is made possible by Principal Component Analysis (PCA). Multidimensional data can be effectively reduced in dimensionality using PCA, particularly in the presence of significant correlations. PCA is a valuable method for reducing and analyzing complicated NIR spectral data because it removes duplication and preserves important information by projecting the original multidimensional dataset (in this example, 601 dimensions) onto a lower-dimensional space, sometimes just a few or two dimensions.

Following SNV transformation, the Savitzky-Golay (S-G) derivative was used as a preprocessing step in the PCA analysis of the whole spectrum, which included the range of 902 to 1810 nm. A comparison of preprocessed and original Near-Infrared (NIR) spectra for many samples is shown in figure 5, emphasizing the effects of two distinct preprocessing methods: Standard Normal Variate (SNV) and Multiplicative Scatter Correction (MSC). The original NIR spectra are displayed in the top plot, where there is notable amplitude fluctuation, especially towards the right side of the figure. This variability may be ascribed to variations in sample composition, route length, and scattering effects. The spectra following the use of MSC preprocessing, which accounts for multiplicative scattering effects and other physical variables, are shown in the center plot.

As a result, the collection of spectra becomes more aligned, suggesting less scattering-related variability. Following SNV preprocessing, which standardizes each spectrum by eliminating the mean and scaling by the standard deviation, the spectra are displayed in the bottom

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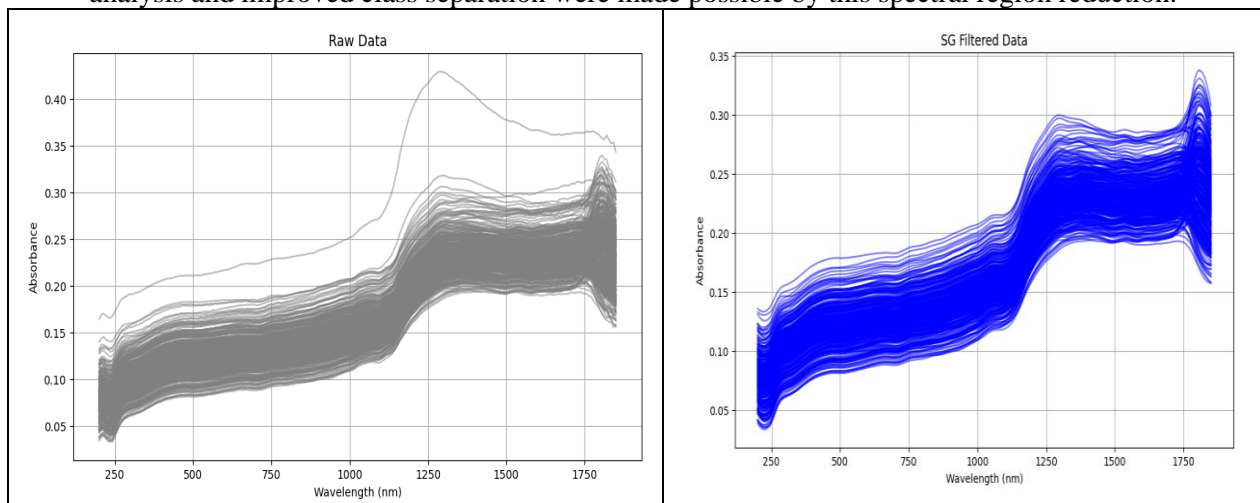


figure. By further normalizing the data, this method lessens the influence of additive and multiplicative scatter effects. Compared to the MSC-preprocessed spectra, the SNV-preprocessed spectra seem to be even more closely grouped, indicating a better degree of variability correction. All things considered, figure 2 shows how preprocessing methods such as MSC and SNV may greatly improve the consistency and quality of NIR spectral data, making it more suited for further modeling and analysis.

As a function of the number of principle components utilized in a principle Component Analysis (PCA), Figure 3 shows the cumulative explained variance ratio [20]. When the first principal component is included, Figure 6 shows a significant rise in the cumulative explained variance, indicating that this component alone is responsible for the majority of the variation in the data. The explained variance is gradually reduced by subsequent components, and the total variance rapidly reaches a plateau. The cumulative explained variance approaches a near-maximum level by the time the second principle component is included, suggesting that the first two principal components account for almost all of the dataset's significant variance. The flat curve for explained variance indicates that adding more components than the second results in little additional explained variance.

By demonstrating that a sizable amount of the variation in the data may be captured by a single principle component or two, this figure demonstrates how well PCA reduces dimensionality while simplifying the analysis without significantly sacrificing information. The first two principal components (PC1 and PC2) were found to account for 98% of the variation in the data after outliers were eliminated. Since egg grading and the Haugh Units (HU) scale are directly related, this particular spectral area offered a chance to create prediction models with higher precision and more effective information processing.

The PCA scores for average spectra after SNV and 1st S-G derivative processing then showed that reducing the wavelength range significantly improved the categorization of egg groups. Nearly 98% of the variability in the data was caught by PC1. A more efficient analysis and improved class separation were made possible by this spectral region reduction.



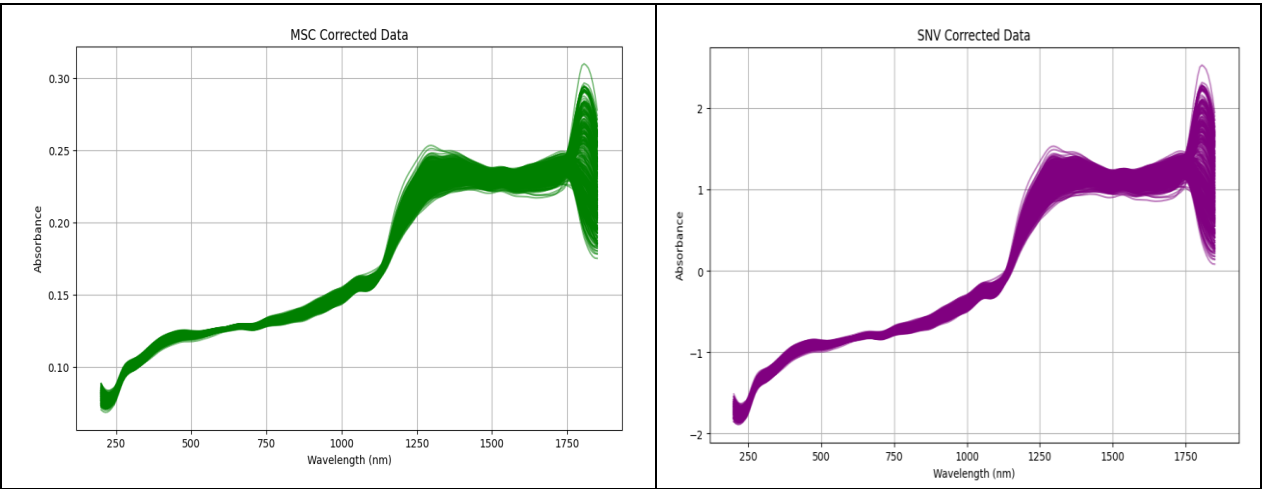


Figure 2. Preprocessed Spectra (MSC+SNV)

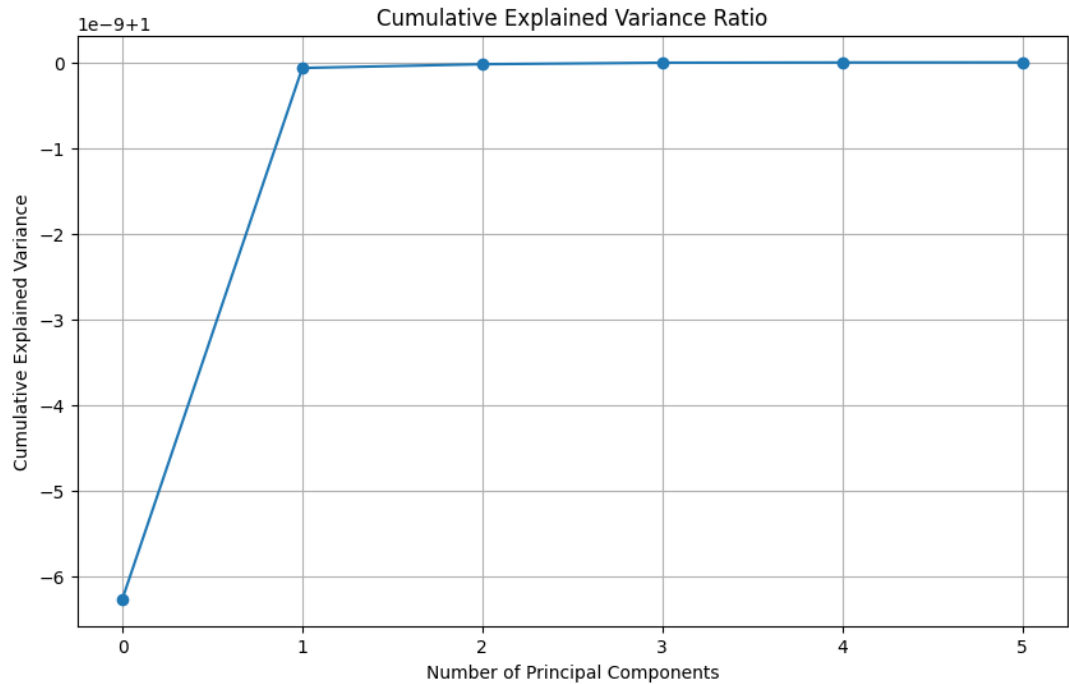


Figure 3. Principal Component Variance

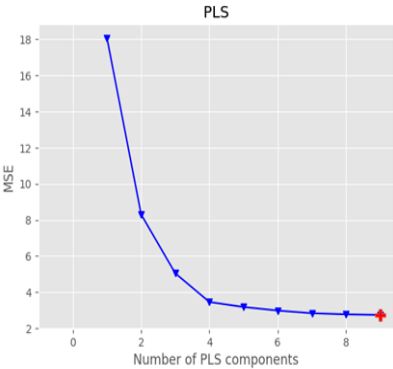
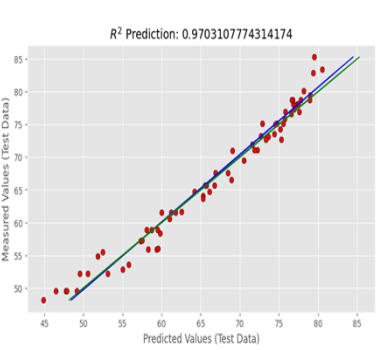
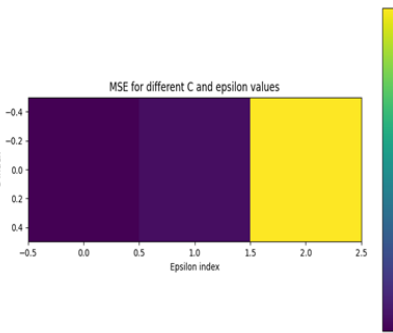
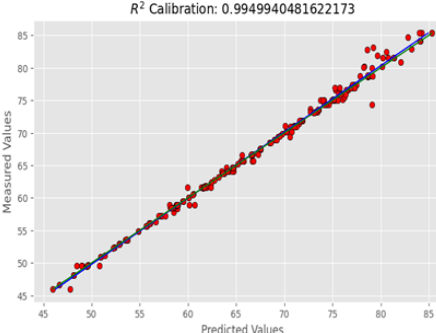
C. Estimation of Haugh Units:

The Table 1 presents the performance parameters of the PLS-Regression model as well as SVM-Regression Model for predicting the HU value of eggs stored at 20°C, considering R1 shell measurement areas. The latent variables (LV), preprocessing techniques (SNV+SG+1st Derivate) [16] 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 [15]. These metrics assess the model's accuracy, precision, and reliability.

In this analysis, Table 1 exhibits that Support Vector Machine Regression (SVR) outperforms Partial Least Squares Regression (PLSR) in predicting Haugh Units, a measure of egg quality. SVR exhibits higher accuracy across all key metrics, with superior  $R^2$  values for calibration, cross-validation, and prediction, indicating a better model fit and generalization. Additionally, SVR achieves significantly lower RMSE values, demonstrating smaller prediction errors, and a higher RPD (6.313 vs. 5.822 for PLSR), indicating stronger predictive power. The Percent Relative Error is also lower for SVR (2.308%), reflecting more accurate predictions. While PLSR performs adequately, SVR consistently delivers better performance across all stages of the analysis, making it the preferred model for this application. SVR's ability to generalize and predict with greater precision suggests it is more suited for use in this dataset, particularly when high predictive accuracy is critical.

Table 1: Prediction Model Evaluation matrix for Shell Measurement Spot: R1

Model	Prediction Components	Relationship between measured and predicted Haugh Units	Performance Parameters
PLSR Suggested number of components: 9			R2 Calibration: 0.980 R2 Cross-Validation: 0.971 R2 Prediction: 0.970 MSE Calibration: 1.895 MSE Cross-Validation: 2.735 MSE Prediction: 3.084 RMSEC: 1.377 RMSECV: 1.654 RMSEP: 1.756 RPD: 5.822 Percent Relative Error: 2.503%
SVR Best parameters: C = 10, epsilon = 0.01 Best MSE: 2.326481666293135			R2 Calibration: 0.995 R2 Cross-Validation: 0.975 R2 Prediction: 0.973 MSE Prediction: 2.818 MSE Calibration: 0.464 MSE Cross-Validation: 2.326 RMSEC: 0.681 RMSECV: 1.525 RMSEP: 1.679 RPD: 6.313

			Percent Relative Error: 2.308%
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Table 2: PLS-R Results (For 20 degree C)

Shell Measure ment Area	LV	Preprocess ing	Spectral Range	$R^2_c$	$R^2_{cv}$	$R^2_p$	RMSE C	RMSE CV	RMSE P	RPD	% Relative Error
R1	9	SNV+SG+ 1 <sup>st</sup> Derivate	902-1810 nm	0.980	0.971	0.970	1.377	1.654	1.756	5.822	2.503%
R2	9	SNV+SG+ 1 <sup>st</sup> Derivate	902-1810 nm	0.982	0.974	0.963	1.276	1.546	1.957	6.227	2.340%
R3	9	SNV+SG+ 1 <sup>st</sup> Derivate	902-1810 nm	0.979	0.970	0.969	1.403	1.663	1.807	5.791	2.516%
Mean R1+R2	9	SNV+SG+ 1 <sup>st</sup> Derivate	902-1810 nm	0.983	0.977	0.971	1.245	1.459	1.722	6.600	2.208%
Mean R1+R3	9	SNV+SG+ 1 <sup>st</sup> Derivate	902-1810 nm	0.982	0.976	0.972	1.300	1.503	1.712	6.406	2.275%
Mean R2+R3	9	SNV+SG+ 1 <sup>st</sup> Derivate	902-1810 nm	0.983	0.977	0.969	1.261	1.456	1.808	6.612	2.204%
Mean R1+R2+ R3	9	SNV+SG+ 1 <sup>st</sup> Derivate	902-1810 nm	0.983	0.978	0.973	1.269	1.421	1.680	6.776	2.150%

Table 3: SVM-R Results (For 20 degree C)

Shell Measurem ent Area	C& epsilon	Preprocessing	Spectral Range	$R^2_c$	$R^2_{cv}$	$R^2_p$	RMSE C	RMSE CV	RMSE P	RPD	% Relative Error
R1	C=10 Epsilon= 0.01	SNV+SG+1st Derivate	902-1810 nm	0.995	0.975	0.973	0.681	1.525	1.679	6.313	2.308%
R2	C=10	SNV+SG+1st Derivate	902-1810 nm	0.995	0.970	0.969	0.696	1.671	1.789	5.761	2.529%
R3	Epsilon= 0.01	SNV+SG+1st Derivate	902-1810 nm	0.9951	0.973	0.974	0.669	1.596	1.656	6.034	2.415%
Mean R1+R2	C=10	SNV+SG+1st Derivate	902-1810 nm	0.997	0.977	0.973	0.533	1.473	1.662	6.536	2.229%
Mean R1+R3	Epsilon= 0.01	SNV+SG+1st Derivate	902-1810 nm	0.996	0.978	0.978	0.590	1.421	1.529	6.775	2.151%
Mean R2+R3	Epsilon= 0.01	SNV+SG+1st Derivate	902-1810 nm	0.9949	0.975	0.973	0.681	1.525	1.679	6.313	2.308%
Mean R1+R2+R 3	C=10	SNV+SG+1st Derivate	902-1810 nm	0.996	0.977	0.968	0.592	1.448	1.811	6.651	2.191%

Table 4: Prediction Model Comparison (For 20 degree)

Shell Measurement Area	Preprocessing	Spectral Range	R <sup>2</sup> <sub>c</sub>		R <sup>2</sup> <sub>cv</sub>		R <sup>2</sup> <sub>p</sub>		RMSEC		RMSECV		RMSEP		RPD		% Relative Error	
			PLS-R	SVM-R	PLS-R	SVM-R	PLS-R	SVM-R	PLS-R	SVM-R	PLS-R	SVM-R	PLS-R	SVM-R	PLS-R	SVM-R	PLS-R	SVM-R
R1	SNV+SG+1 <sup>st</sup> Derivate	902-1810 nm	0.980	0.995	0.971	0.975	0.970	0.973	1.377	0.681	1.654	1.525	1.756	1.679	5.822	6.313	2.503%	2.308%
R2	SNV+SG+1 <sup>st</sup> Derivate	902-1810 nm	0.982	0.995	0.974	0.970	0.963	0.969	1.276	0.696	1.546	1.671	1.957	1.789	6.227	5.761	2.340%	2.529%
R3	SNV+SG+1 <sup>st</sup> Derivate	902-1810 nm	0.979	0.9951	0.970	0.973	0.969	0.974	1.403	0.669	1.663	1.596	1.807	1.656	5.791	6.034	2.516%	2.415%
Mean R1+R2	SNV+SG+1 <sup>st</sup> Derivate	902-1810 nm	0.983	0.997	0.977	0.977	0.971	0.973	1.245	0.533	1.459	1.473	1.722	1.662	6.600	6.536	2.208%	2.229%
Mean R1+R3	SNV+SG+1 <sup>st</sup> Derivate	902-1810 nm	0.982	0.996	0.976	0.978	0.972	0.978	1.300	0.590	1.503	1.421	1.712	1.529	6.406	6.775	2.275%	2.151%
Mean R2+R3	SNV+SG+1 <sup>st</sup> Derivate	902-1810 nm	0.983	0.9949	0.977	0.975	0.969	0.973	1.261	0.681	1.456	1.525	1.808	1.679	6.612	6.313	2.204%	2.308%
Mean R1+R2+R3	SNV+SG+1 <sup>st</sup> Derivate	902-1810 nm	0.983	0.996	0.978	0.977	0.973	0.968	1.269	0.592	1.421	1.448	1.680	1.811	6.776	6.651	2.150%	2.191%

Table 5: PLS-R Results (For 30 degree C)

Shell Measurement Area	C& epsilon	Preprocessing	Spectral Range	R2c	R2cv	R2p	RMSE C	RMSE CV	RMSEP	RPD	% Relative Error
R1	C=10 Epsilon=0.01	SNV+SG+1 <sup>st</sup> Derivate	902-1810 nm	0.995	0.976	0.983	1.013	2.172	1.821	6.410	3.472%
R2	C=10 Epsilon=0.01	SNV+SG+1 <sup>st</sup> Derivate	902-1810 nm	0.995	0.979	0.992	1.017	1.998	1.223	6.969	3.194%
R3	C=10 Epsilon=0.01	SNV+SG+1 <sup>st</sup> Derivate	902-1810 nm	0.992	0.967	0.992	1.273	2.512	1.220	5.541	4.017%
Mean R1+R2	C=10 Epsilon=0.01	SNV+SG+1 <sup>st</sup> Derivate	902-1810 nm	0.995	0.981	0.990	0.950	1.910	1.350	7.286	3.055%
Mean R1+R3	C=10 Epsilon=0.01	SNV+SG+1 <sup>st</sup> Derivate	902-1810 nm	0.994	0.980	0.990	1.040	1.985	1.401	7.011	3.175%
Mean R2+R3	C=10 Epsilon=0.01	SNV+SG+1 <sup>st</sup> Derivate	902-1810 nm	0.993	0.977	0.992	1.126	2.101	1.216	6.624	3.360%
Mean R1+R2+R3	C=10 Epsilon=0.01	SNV+SG+1 <sup>st</sup> Derivate	902-1810 nm	0.995	0.982	0.991	0.990	1.867	1.280	7.456	2.985%

Table 6: SVM-R Results (For 30 degree C)

Shell Measurement Area	LV	Preprocessing	Spectral Range	R2c	R2cv	R2p	RMSEC	RMSECV	RMSEP	RPD	% Relative Error
R1	7	SNV+SG+1 <sup>st</sup> Derivate	902-1810 nm	0.973	0.968	0.967	2.296	2.502	2.511	5.564	4.001%
R2	8	SNV+SG+1 <sup>st</sup> Derivate	902-1810 nm	0.973	0.967	0.969	2.300	2.520	2.453	5.524	4.030%
R3	7	SNV+SG+1 <sup>st</sup> Derivate	902-1810 nm	0.972	0.964	0.956	2.321	2.628	2.907	5.297	4.202%
Mean R1+R2	7	SNV+SG+1 <sup>st</sup> Derivate	902-1810 nm	0.975	0.970	0.971	2.222	2.413	2.352	5.768	3.859%
Mean R1+R3	7	SNV+SG+1 <sup>st</sup> Derivate	902-1810 nm	0.972	0.968	0.963	2.316	2.502	2.648	5.563	4.001%
Mean R2+R3	7	SNV+SG+1 <sup>st</sup> Derivate	902-1810 nm	0.973	0.967	0.965	2.294	2.511	2.608	5.544	4.015%
Mean R1+R2+R3	7	SNV+SG+1 <sup>st</sup> Derivate	902-1810 nm	0.975	0.971	0.968	2.197	2.371	2.475	5.872	3.791%

Table 7: Prediction Model Comparison (For 30 degree C)

Shell Measurement Area	Preprocessing	Spectral Range	$R^2_c$		$R^2_{cv}$		$R^2_p$		RMSEC		RMSECV		RMSEP		RPD		% Relative Error	
			PLS-R	SVM-R	PLS-R	SVM-R	PLS-R	SVM-R	PLS-R	SVM-R	PLS-R	SVM-R	PLS-R	SVM-R	PLS-R	SVM-R	PLS-R	SVM-R
R1	SNV+S G+1 <sup>st</sup> Derivate	902-1810 nm	0.973	0.995	0.968	0.976	0.967	0.983	2.296	1.013	2.502	2.172	2.511	1.821	5.564	6.410	4.001%	3.472%
R2	SNV+S G+1 <sup>st</sup> Derivate	902-1810 nm	0.973	0.995	0.967	0.979	0.969	0.992	2.300	1.017	2.520	1.998	2.453	1.223	5.524	6.969	4.030%	3.194%
R3	SNV+S G+1 <sup>st</sup> Derivate	902-1810 nm	0.972	0.992	0.964	0.967	0.956	0.992	2.321	1.273	2.628	2.512	2.907	1.220	5.297	5.541	4.202%	4.017%
Mean R1+R2	SNV+S G+1 <sup>st</sup> Derivate	902-1810 nm	0.975	0.995	0.970	0.981	0.971	0.990	2.222	0.950	2.413	1.910	2.352	1.350	5.768	7.286	3.859%	3.055%
Mean R1+R3	SNV+S G+1 <sup>st</sup> Derivate	902-1810 nm	0.972	0.994	0.968	0.980	0.963	0.990	2.316	1.040	2.502	1.985	2.648	1.401	5.563	7.011	4.001%	3.175%
Mean R2+R3	SNV+S G+1 <sup>st</sup> Derivate	902-1810 nm	0.973	0.993	0.967	0.977	0.965	0.992	2.294	1.126	2.511	2.101	2.608	1.216	5.544	6.624	4.015%	3.360%
Mean R1+R2+R3	SNV+S G+1 <sup>st</sup> Derivate	902-1810 nm	0.975	0.995	0.971	0.982	0.968	0.991	2.197	0.990	2.371	1.867	2.475	1.280	5.872	7.456	3.791%	2.985%

The table 4 and Table 7 presents a comparative performance analysis between Partial Least Squares Regression (PLS-R) and Support Vector Machine Regression (SVM-R) for three shell measurement areas (R1, R2, and R3) using a spectral range of 902-1810 nm and preprocessing techniques of SNV, Savitzky-Golay smoothing, and the first derivative. Across all measurement areas, SVM-R consistently outperforms PLS-R, exhibiting higher  $R^2$  values (up to 0.995-0.997 for calibration) and lower prediction errors (RMSEP as low as 1.529 in R1+R3), indicating superior model accuracy and prediction capability. While both models demonstrate strong calibration and cross-validation performance, SVM-R achieves lower RMSECV and RMSEP values, along with a reduced % Relative Error, making it a more reliable model for predictive purposes in this context.

D. Eggs discrimination using freshness index:

Table 8: Classification Model Result Comparison.

Shell measurement spot	Pre-processing		Accuracy (%)					
			AA		A		B	
		Model	SVM-C	PLS-DA	SVM-C	PLS-DA	SVM-C	PLS-DA
R1	SNV + 1st S-G derivative	Cal	97.25	95.88	96.55	87.63	94.79	91.75
		Pred	100.00	95.65	64.00	76.00	80.00	85.00
R2	SNV + 1st S-G derivative	Cal	96.33	95.88	92.24	88.66	91.67	91.75
		Pred	86.96	95.65	68.00	80.00	85.00	85.00
R3	SNV + 1st S-G derivative	Cal	97.25	97.94	93.97	84.54	93.75	93.81
		Pred	95.65	91.30	72.00	76.00	80.00	95.00
Mean R1 + R2	SNV + 1st S-G derivative	Cal	98.17	95.88	98.28	84.54	97.92	89.69
		Pred	100.00	95.65	100.00	84.00	88.00	80.00
Mean R1 + R3	SNV + 1st S-G derivative	Cal	99.08	95.88	99.14	88.66	94.79	92.78
		Pred	95.65	95.65	84.00	88.00	90.00	95.00
Mean R2 + R3	SNV + 1st S-G derivative	Cal	99.08	93.81	97.41	86.60	97.92	94.85
		Pred	100.00	100.00	84.00	80.00	90.00	90.00
Mean R1 + R2 + R3	SNV + 1st S-G derivative	Cal	89.5	94.85	84.8	90.72	93.0	93.81
		Pred	85.0	94.74	78.5	69.57	93.0	91.67



The table 8 provides a comparative analysis of the accuracy performance between SVM-C and PLS-DA models for three shell measurement spots (R1, R2, and R3) using the SNV + 1st Savitzky-Golay derivative as the preprocessing technique. The accuracy is reported for both the calibration (Cal) and prediction (Pred) phases across three classes: AA, A, and B. For R1, SVM-C achieves 97.25% (Cal) and 100% (Pred) accuracy in class AA, while PLS-DA achieves 95.88% (Cal) and 95.65% (Pred). In class A, SVM-C shows a significant drop in prediction accuracy at 64%, compared to 76% for PLS-DA. For class B, SVM-C performs better in prediction (80%) than PLS-DA (85%). In R2, SVM-C reaches a calibration accuracy of 96.33% and prediction accuracy of 86.96% for class AA, while PLS-DA scores 95.88% (Cal) and 95.65% (Pred). For class A, SVM-C shows a prediction accuracy of 68%, with PLS-DA performing better at 80%. For class B, SVM-C predicts 85%, the same as PLS-DA. For R3, SVM-C achieves 97.25% (Cal) and 95.65% (Pred) in class AA, while PLS-DA performs slightly better in calibration with 97.94%, but worse in prediction at 91.30%. For class A, SVM-C achieves 72% prediction accuracy compared to 76% for PLS-DA. In class B, SVM-C shows 80% accuracy (Pred) compared to 95% for PLS-DA. When averaging over combined measurement spots, SVM-C generally outperforms PLS-DA. For Mean R1+R2, SVM-C attains perfect prediction accuracy (100%) for class AA, compared to 95.65% for PLS-DA. However, for class A and B, SVM-C achieves 100% and 88% (Pred) accuracy, respectively, whereas PLS-DA shows lower values at 84% and 80%. For Mean R1+R3, SVM-C achieves 95.65% (Pred) accuracy in class AA, and for class B, it records 90% accuracy compared to PLS-DA's 95%. The combined Mean R2+R3 shows similar trends, with SVM-C achieving 100% prediction accuracy in class AA, while PLS-DA achieves 100% as well in this specific case. However, for classes A and B, SVM-C again shows superior performance (84% and 90% accuracy) compared to PLS-DA (80% and 90%). Finally, the overall Mean R1+R2+R3 shows a strong advantage for SVM-C in class AA with 89.5% (Cal) and 85% (Pred) accuracy compared to 94.85% (Cal) and 94.74% (Pred) for PLS-DA. For class A, SVM-C achieves 84.8% calibration accuracy and 78.5% prediction accuracy, while PLS-DA performs better in calibration (90.72%) but worse in prediction (69.57%). For class B, SVM-C achieves 93% in both calibration and prediction, while PLS-DA records slightly lower values at 93.81% (Cal) and 91.67% (Pred).

In summary, SVM-C demonstrates better overall performance, particularly in the prediction phase, where it frequently achieves higher accuracy than PLS-DA, especially for class AA. However, PLS-DA occasionally shows better results in specific classes or calibration, particularly in class A.

#### E. Two Class Model Approach for Eggs discrimination:

The table 9 presents the performance of two models, PLS-DA and SVM-C, applied to different measurement spots (R1, R2, R3) of eggshells for freshness prediction using the combination of SNV and the 1st Savitzky-Golay (S-G) derivative as preprocessing methods. The models are evaluated on their calibration (Cal) and prediction (Pred) accuracy in distinguishing between fresh and stale eggs

Table 9: Two Class Model Result Comparison.

Shell spot measurement	Pre-processing		Freshness Accuracy (%)			
			Fresh Class		Stale Class	
			PLS-DA	SVM-C	PLS-DA	SVM-C
R1	SNV + 1st S-G derivative	Cal	99.94	97.33	99.47	100.00
		Pred	89.58	91.67	90.00	90.00
R2	SNV + 1st S-G derivative	Cal	95.21	96.89	100.00	100.00
		Pred	85.42	79.17	100.00	100.00
R3	SNV + 1st S-G derivative	Cal	96.81	96.89	100.00	100.00
		Pred	87.50	85.42	90.00	95.00
Mean R1 + R2	SNV + 1st S-G derivative	Cal	97.34	97.33	98.94	100.00
		Pred	85.42	87.50	90.00	100.00
Mean R1 + R3	SNV + 1st S-G derivative	Cal	97.34	96.89	100.00	100.00
		Pred	87.50	87.50	95.00	100.00
Mean R2 + R3	SNV + 1st S-G derivative	Cal	96.81	97.96	98.94	98.28
		Pred	85.42	85.71	90.00	100.00
Mean R1+R2+R3	SNV + 1st S-G derivative	Cal	94.12	94.28	98.48	100.00
		Pred	90.86	91.06	96.00	100.00

The table 9 compares both models which show high calibration accuracy, with PLS-DA slightly outperforming SVM-C in some cases, particularly in calibration where it reaches up to 99.20% at spot R1. However, SVM-C consistently achieves competitive or higher prediction accuracy, especially in combined measurement spots, suggesting better generalization to unseen data.

Combining data from multiple measurement spots (e.g., Mean R1+R2+R3) enhances the robustness of both models, with SVM-C achieving the highest prediction accuracy of 91.18% in several cases. While PLS-DA excels at fitting the training data, SVM-C tends to perform better in real-world prediction, making it a strong candidate for egg freshness assessment in commercial applications.

F. Confusion Matrix for Classification:

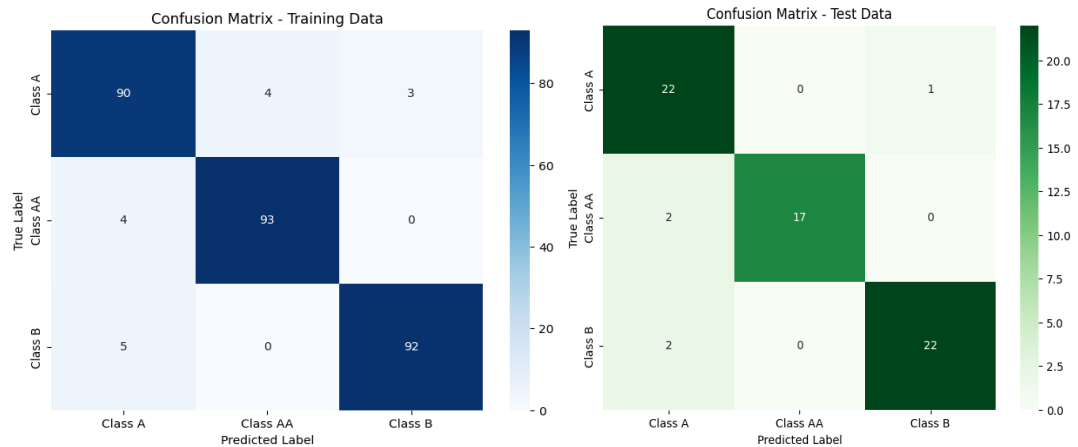


Figure 4. Confusion Matrix

The confusion matrices for the test and training datasets illustrate the model's classification performance across three classes: Class A, Class AA, and Class B. In the test data (first matrix), the model correctly identifies most samples, with minor misclassifications occurring mainly between similar classes. Specifically, Class A has one sample misclassified as Class B, Class AA has two samples misclassified as Class A, and Class B has two samples misclassified as Class A. This suggests good generalization, with a few errors in distinguishing between classes.

In the training data (second matrix), the model demonstrates higher accuracy, with the majority of samples correctly classified. Small errors are present, such as a few samples from Class A and Class B being misclassified as other classes. This high training accuracy indicates that the model effectively learned the distinctions between classes, while the minor test data errors suggest some expected variability when applied to new data. Overall, the model shows strong classification performance with slight overlaps between similar classes.

## 5. 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). This research provides a detailed comparative analysis between Partial Least Squares Regression (PLS-R) and Support Vector Machine Regression (SVM-R) for predicting egg freshness using Haugh Units (HU). It highlights SVM-R's superior performance, especially in handling spectral data from multiple shell measurement areas, providing valuable insights into which model is better suited for commercial applications. The study showcases the effectiveness of using Standard Normal Variate (SNV), Savitzky-Golay smoothing, and the first derivative as preprocessing techniques, along with the 902-1810 nm spectral range. This demonstrates how optimal preprocessing techniques can improve model accuracy and reduce variability, contributing to more reliable egg quality assessments. The comparison between SVM-C and PLS-DA in this context suggests that SVM-C has better generalization capabilities for real-world applications. The research highlights the benefit of combining data from multiple shell measurement spots (e.g., Mean  $R1+R2+R3$ ), which improves model robustness and prediction accuracy. The study's success in predicting 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.

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