

Development of two-band color-mixing technique for identification of broiler carcass conditions [☆]

K. Chao ^{a,*}, Y.-R. Chen ^a, F. Ding ^b, C.-C. Yang ^b, D.E. Chan ^a

^a USDA/ARS/ISL, Beltsville, MD, United States

^b University of Kentucky, Lexington, KY, United States

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Abstract

The development of accurate, rapid, and non-invasive inspection technologies are needed to help poultry processors meet food safety regulations and rising consumer demand while increasing productivity and economic competitiveness. This paper reports on a novel two narrow-band color-mixing technique for identification of broiler chicken carcass conditions. Spectra were collected for samples cut from the breast area of 103 wholesome chicken carcasses, 66 systemically diseased chicken carcasses, and 40 cadaver chicken carcasses using a photodiode array spectrophotometer system. Waveband pairs in the range of 416–715 nm were evaluated for identifying chicken conditions using the two-band color-mixing technique, and the pair of (453 nm, 589 nm) was selected based on color difference index calculations in CIELUV color space. Significant differences in the color characteristics of wholesome, systemically diseased, and cadaver chicken conditions, based on color-mixing using the two selected wavebands, were confirmed by one-way analysis of variance. Decision-tree classification models using the calculated color difference indexes were evaluated first by using the spectral data divided into a validation set and a testing set, and second by 10-fold cross-validation of the entire data set. Classification accuracies achieved for the wholesome, systemically diseased, and cadaver samples were 95.8%, 95.5%, and 100%, respectively, for the validation set; 94.6%, 100%, and 90.6%, respectively, for the testing set; and 98.1%, 97.5%, and 93.9%, respectively, when using 10-fold cross-validation. Published by Elsevier Ltd.

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1. Introduction

The Poultry Products Inspection Act (PPIA) requires Food Safety and Inspection Service (FSIS) inspectors of the United States Department of Agriculture (USDA) to conduct post-mortem inspection for wholesomeness of all chickens intended for sale to US consumers. FSIS has completed the transformation of its traditional inspection

system to a Hazard-Analysis-and-Critical-Control-Point (HACCP) inspection system (USDA, 1996). With more focus on HACCP and HIMP (HACCP-based inspections models project), the FSIS has placed more of the burden of inspection responsibility on the processors. Plants are also responsible for meeting other consumer protection (OCP) issues as determined by the regulatory agency. In essence, processors assume the responsibility for inspection, and the regulatory agency performs oversight and verification to ensure standards are met. HACCP or HIMP helps meet consumer demand for safe, high quality food; however, consumer demand for more food increases the need for, and pressure on, inspectors. Consequently, the development of accurate, rapid, and non-invasive technologies appropriate for operation on high-speed processing lines is of great importance for the poultry industry.

[☆] Mention of trade names or commercial products is solely for the purpose of providing specific information and does not imply endorsement or recommendation by the USDA.

* Corresponding author. Address: USDA/ARS/ISL, Building 303, BARC-East, 10300 Baltimore Avenue, Beltsville, MD 20705-2350; Tel.: +1 301 504 8450; fax: +1 301 504 9466.

E-mail address: chaok@ba.ars.usda.gov (K. Chao).

The USDA's Agricultural Research Service has been conducting research to develop spectral imaging methods suited for the poultry processing industry. In particular, visible/near-infrared (Vis/NIR) spectroscopic technologies have been shown capable of distinguishing between wholesome and unwholesome poultry carcasses and detecting fecal contamination on poultry carcasses due to differences in skin and tissue composition. Chen and Massie (1993) used Vis/NIR measurements taken by a photodiode array spectrophotometer to classify wholesome and unwholesome chicken carcasses, and selected wavelengths at 570 nm, 543 nm, 641 nm, 847 nm based on linear regression for classification. Using Vis/NIR measurements of fecal contamination of poultry carcasses, Windham, Lawrence, Park, and Buhr (2003a) identified four key wavelengths via principal component analysis at 434 nm, 517 nm, 565 nm, and 628 nm. Through single-term linear regression (STLR), an optimal ratio of 574 nm/588 nm was determined and used to achieve 100% detection of contaminants (Windham, Smith, Park, Lawrence, & Feldner, 2003b). Chao, Chen, and Chan (2004) developed an on-line inspection system to measure the reflectance spectra of poultry carcasses in the visible to near-infrared regions between 431 and 944 nm. The instrument measured the spectra of veterinarian-selected carcasses running at speeds of 140 and 180 birds per minute (bpm). Results showed that this Vis/NIR system can be used to differentiate between wholesome and unwholesome poultry carcasses at high speeds. These studies include significant findings for the use of spectral reflectance in the visible region, but have not utilized methods of analysis for sample color as perceived through human vision.

The International Commission for Illumination (CIE) has established a colorimetry system for identifying and specifying colors, and for defining color standards. Following the establishment of the CIE (1924) luminous efficiency function ($V(\lambda)$), the system of colorimetry was developed based on the principles of trichromacy and Grassmann's laws of additive color mixture (Fairchild, 1998). The concept of the colorimetry system is that any color can be matched by an additive mixture of three primary colors, red, green, and blue. Because there are three different types of color receptor cones in the eye, all the colors that humans see can be described by coordinates in a three-dimensional color space which measures the relative stimulations to each type of cone. These coordinates are called tristimulus values and can be measured in color-matching experiments. The tristimulus values are the amounts of the three primary colors which were used to achieve a match.

A system using broad-band primaries was formalized in 1931 by the CIE. Wavelength-by-wavelength measurement of tristimulus values for the visible spectrum produces the color-matching functions. The tristimulus values for a particular color are labeled (X , Y , Z) in the CIE (1931) system. The tristimulus values are extended such that they can be obtained for any given stimulus, defined by a spectral

power distribution (SPD) (Williamson & Cummins, 1983). The SPD can be measured by a spectrophotometer. From the SPD, both the luminance and the chromaticity of a color are derived to precisely describe the color in the CIE system.

Recent research at the USDA Instrumentation and Sensing Laboratory has focused on color-mixing techniques and their implementation for low-cost portable optical devices, such as binoculars, for use by human inspectors in small processing plants where more costly automated vision systems may not be feasible (Ding, Chen, & Chao, 2005, 2006; Chao, Chen, Ding, & Chan, 2005). Ding et al. demonstrated the relationship between color-mixing as perceived by the human eye and the use of band-ratios in multispectral imaging systems; and with visible/near infrared spectral data, conducted a laboratory simulation for identifying six different chicken conditions by human eye using an optical color-mixing device. Using visible/near-infrared spectral data collected for fresh chickens on a 140 bird-per-minute processing line at a commercial processing plant, Chao et al. used calculated values of the color difference index to classify two categories of birds (wholesome and systemically diseased).

The overall objective of this research was to investigate the two-band color-mixing technique for automated poultry inspection. Specific objectives were to select a waveband pair in the visible spectrum region suitable for identifying three chicken conditions (wholesome, systemically diseased, and cadaver) using the two-band color-mixing technique; to compare the chicken color characteristics using two-band color-mixing calculations; and to test decision-tree classification models based on color difference indexes calculated using two-band color-mixing.

2. Materials and methods

2.1. Chicken samples and spectral measurement

A total of 209 chicken carcasses (103 wholesome, 66 systemically diseased, 40 cadaver) were obtained in groups of 10–15 birds from a processing line at a poultry slaughter plant in Cordova, Maryland over a period of four months. The bird conditions were identified at the plant by USDA FSIS inspectors. Chicken carcasses were marked according to condition and placed in plastic bags to minimize dehydration during transport. Then the bags were placed in coolers, covered with ice, and transported to the ISL facility in Beltsville, Maryland, within 2 h for the experiment.

Spectral reflectance data of samples cut from poultry carcasses were collected using a spectrophotometer system, which consisted of a tungsten halogen light source and power supply, a bifurcated fiber-optic probe (diameter 25.4 mm) assembly, a spectrograph, a photodiode array detector and a personal computer with a photodiode array computer interface card installed (Fig. 1). The spectrograph (Spectra-Physics, Model 77400, Stratford, CT) had a focal length of 120 mm, a grating ruling of 400 lines/

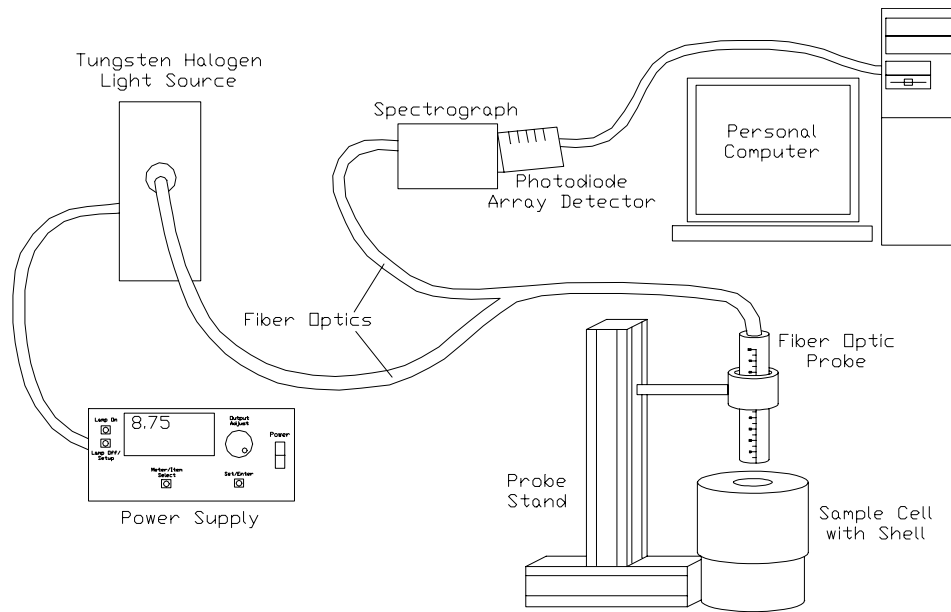


Fig. 1. Schematic of spectrophotometer system.

mm. At the exit of the spectrograph was a 1024-silicon photodiode detector (Spectra-Physics, Model 78220, Stratford, CT) element head. Light from a 100-W quartz tungsten halogen (QTH) light bulb was focused on the light source circular end of a 1.2 m bifurcated fiber-optic bundle (Schott-Fostec, Auburn, NY) with the use of a condensing/imaging lens assembly ($f/1.8$, 33-mm aperture, UV-grade fused silica). A radiometric constant-current DC power supply (Spectra-Physics, Model 68831, Stratford, CT) was used to keep the light intensity constant and minimize color temperature shift as a result of power line fluctuations. This light energy traveled through the fiber-optic cable and exited by means of a concentric ring of optical fibers (diameter 15.8 mm, thickness 1.1 mm), to be focused on the chicken sample surface. After interacting with the chicken sample, the light energy was then collected through a 7.5-mm diameter aperture in the center of the concentric optical probe. The collected energy was transmitted back through the bifurcated fiber-optic cable to a 4-mm high \times 50- μ m wide exit rectangle (slit) on the spectrograph ferrule. The fixed entrance slit of the spectrograph was 3-mm high by 25- μ m wide. The photodiode array detector head was connected via a cable to a personal computer interface controller card (Spectra-Physics, Model CC100, Stratford, CT). This interface card provided a 16-bit analog-to-digital converter that converts the analog signal from each diode to a digitized number and transfers the data to the personal computer.

The sample holder was a cylindrical white Teflon cell with an interior chamber measuring 5 cm in height and 5 cm in diameter. The walls and base of the cylinder were 1.5 cm thick. Each sample was placed flat in the bottom of the cylinder. The probe was mounted in the cylinder so that the distance from the probe to the sample was about 2 cm. For each sample, the right breast was removed

with the skin intact, and from this a 49-mm diameter circular area was cut out. The skin, approximately 4 mm thick, was removed and set aside while the meat was sliced to a thickness of 15 mm. Before sample reflectance measurements were taken, dark background and white reference measurements were collected. A dark background measurement was taken, to compensate for the zero energy signal, by placing the probe 2 cm from the bottom of an empty black Teflon cell (of the same dimensions as the sample holder) with the light source turned off. A white reference measurement was taken, to establish a spectrally flat repeatable high energy reference, by placing the fiber-optic probe 2 cm from the bottom of the empty sample holder with the light source turned on. To take a sample reflectance measurement, the sample (chicken meat with skin overlaid) was placed in the sample holder and the fiber-optic probe was positioned 2 cm above the surface of the sample. Spectra were recorded as relative reflectance based on the white reference measurement, according to the formula:

Relative reflectance

$$= \frac{\text{sample reflectance} - \text{background reflectance}}{\text{reference reflectance} - \text{background reflectance}} \quad (1)$$

The spectrophotometer measured the spectral reflectance of each sample in the wavelength range of 411.33–923.41 nm. Each spectrum was composed of 1024 data points measured at intervals slightly more than 0.5 nm.

2.2. Two-band color mixing

The CIE tristimulus values X , Y , and Z of the color of each chicken sample were obtained by multiplying the spectral irradiance of the QTH light source, S_λ ($\text{W m}^{-2} \text{nm}^{-1}$),

the relative spectral reflectance of the chicken samples, R_λ , and the 1931 CIE color matching functions \bar{x}_λ , \bar{y}_λ , and \bar{z}_λ :

$$\begin{aligned} X &= k \sum_{\lambda} S_{\lambda} R_{\lambda} \bar{x}_{\lambda} \\ Y &= k \sum_{\lambda} S_{\lambda} R_{\lambda} \bar{y}_{\lambda} \\ Z &= k \sum_{\lambda} S_{\lambda} R_{\lambda} \bar{z}_{\lambda} \end{aligned} \quad (2)$$

where k is a normalizing constant. The combined term of $S_{\lambda} R_{\lambda}$ is called the spectral power distribution. In a special case where absolute values for the spectral power distribution is given, it is convenient to use $k = 683$ lumens per watt, from which the calculated value of Y is the luminous flux expressed in lumens.

The additive nature of the tristimulus values, as seen in Eq. (2), means that for the mixing of two colors, the tristimulus values X_m , Y_m , Z_m of the new color can be calculated from the tristimulus values of the colors being mixed:

$$\begin{aligned} X_m &= X_1 + X_2 \\ Y_m &= Y_1 + Y_2 \\ Z_m &= Z_1 + Z_2 \end{aligned} \quad (3)$$

2.3. Wavelength selection using two-band color mixing

Potential wavelength pairs for chicken inspection using two-band color mixing were selected using values of the color difference index, ΔE , calculated for wholesome, systemically diseased, and cadaver chicken samples in CIELUV color space. First, an average wholesome spectrum was calculated using 106 wholesome chicken samples. Second, the tristimulus values for 10-nm wavebands centered at λ were calculated using Eq. (2) for all values of λ in the visible spectrum range of 416–715 nm, resulting in tristimulus values for 300 wavebands. Third, for each of the 90,000 possible pairings of λ_1 and λ_2 from among the 300 wavebands, the tristimulus values for the color-mixing of λ_1 and λ_2 were calculated using Eq. (3). Fourth, the color-mixed tristimulus values were converted into CIE-LUV color space values (Wyszecki & Stiles, 1982) using Eq. (4) as follows:

$$L^* = \begin{cases} 25 \left(\frac{100Y}{Y_n} \right)^{1/3} - 16, & \text{if } Y/Y_n > 0.00856 \\ 903.29 \left(\frac{Y}{Y_n} \right), & \text{otherwise} \end{cases} \quad (4)$$

$$u^* = 13L^*(u' - u'_n)$$

$$v^* = 13L^*(v' - v'_n)$$

with

$$u' = \frac{4X}{X + 15Y + 3Z}$$

$$v' = \frac{9Y}{X + 15Y + 3Z}$$

$$u'_n = \frac{4X_n}{X_n + 15Y_n + 3Z_n}$$

$$v'_n = \frac{9Y_n}{X_n + 15Y_n + 3Z_n}$$

The tristimulus values (X_n , Y_n , Z_n) are those of the nominal white reference, also calculated from 10-nm wavebands. In this study, the spectral reflectance of the white reference, R_λ , was set equal to 1.0 for all wavelengths. The above calculations were repeated using 66 systemically diseased chicken samples and 40 cadaver samples, resulting in color-mixed CIELUV color space values for 90,000 waveband pairings for each of the three categories.

The color difference index, ΔE , takes into account the difference in lightness, hue, and saturation between two samples, and is calculated as the Euclidean distance between two points in this three-dimensional space:

$$\Delta E(L^*u^*v^*) = [(\Delta L^*)^2 + (\Delta u^*)^2 + (\Delta v^*)^2]^{1/2} \quad (5)$$

For all possible pairings of λ_1 and λ_2 , the color difference index ΔE was calculated between all three categories of chicken conditions (i.e. wholesome vs. systemically diseased, wholesome vs. cadaver, and systemically diseased vs. cadaver), for a total of three ΔE values per waveband pair.

A combination of maximum and minimum operations was used to determine the potential waveband pairs for discrimination of chicken conditions by color difference index, ΔE . For example, three values of color difference ΔE were calculated for each of the 90,000 possible waveband combinations of λ_1 and λ_2 . For each combination, the minimum operation selected the lowest ΔE from among the three values. Selecting the smallest difference thus guarantees that all categories can be differentiated using this waveband pair. From the resulting set of 90,000 minimum values, the maximum operation was used to select the waveband pair with the largest ΔE as the potential waveband pair.

2.4. Statistical analysis

Individual L^* , u^* , and v^* values for each of the 106 wholesome chicken samples, 66 systemically diseased samples, and 40 cadaver samples, were calculated for the selected waveband pair. One-way analysis of variance (ANOVA1, MATLAB) was used to determine whether the L^* lightness values were different between the three groups. The analysis of variance requires the assumptions of normality and equality in variance. Prior to performing analysis of variance, the Lilliefors hypothesis test (LILLIETEST, MATLAB) was used to test the normality of each distribution and the Levene's test (LEVENETEST, MATLAB) was used to verify equality in variance. A nonparametric one-way ANOVA using Kruskal–Wallis scores (KRUSKAWALLIS, MATLAB) was also performed if the assumptions of normality and homoscedasticity were not met. Tukey–Kramer's test (MULTCOMPARE, MATLAB) was used for pair-wise comparisons of the mean L^* value between the three groups, where the null hypothesis of no difference was rejected if 0 was included in the

confidence interval. One-way analysis of variance and Tukey–Kramer’s test was also performed for analysis of u^* values and v^* values between the three groups.

2.5. Classification modeling

The classification model was created based on the color difference index, ΔE . The spectral data collected for 103 wholesome chickens, 66 systemically diseased, and 40 cadaver chickens was divided into a validation set (48 wholesome, 34 systemically diseased, and 22 cadaver birds from the first two months of data collection) and a testing set (55 wholesome, 32 systemically diseased, and 18 cadaver birds from the last two months of data collection). Using the selected waveband pair centered at λ_1 and λ_2 , the tristimulus values X_{λ_1} , Y_{λ_1} , Z_{λ_1} , and X_{λ_2} , Y_{λ_2} , and Z_{λ_2} , were calculated from the three mean spectra for the wholesome, systemically diseased, and cadaver validation samples, respectively. The color-mixing sum of these values, $X_{\text{ref}} = X_{\lambda_1} + X_{\lambda_2}$, $Y_{\text{ref}} = Y_{\lambda_1} + Y_{\lambda_2}$, $Z_{\text{ref}} = Z_{\lambda_1} + Z_{\lambda_2}$, were used to obtain the reference L^* , u^* , v^* values for wholesome (L_{wref}^* , u_{wref}^* , v_{wref}^*), systemically diseased (L_{sref}^* , u_{sref}^* , and v_{sref}^*), and cadaver (L_{cref}^* , u_{cref}^* , v_{cref}^*) birds. These coordinates in CIELUV space represent the reference points for wholesome, systemically diseased, and cadaver chicken samples.

The values of L^* , u^* , and v^* for each of the individual 48 wholesome, 34 systemically diseased, and 22 cadaver spectra in the validation set were similarly calculated, for use in calculating the ΔE values between each individual sample and the wholesome and unwholesome reference points. The ΔE values, d_1 , d_2 , and d_3 , between each sample and the wholesome, systemically diseased, and cadaver reference points, respectively, were calculated as follows:

$$d_1 = [(L^* - L_{\text{wref}}^*)^2 + (u^* - u_{\text{wref}}^*)^2 + (v^* - v_{\text{wref}}^*)^2]^{1/2} \quad (6)$$

$$d_2 = [(L^* - L_{\text{sref}}^*)^2 + (u^* - u_{\text{sref}}^*)^2 + (v^* - v_{\text{sref}}^*)^2]^{1/2} \quad (7)$$

$$d_3 = [(L^* - L_{\text{cref}}^*)^2 + (u^* - u_{\text{cref}}^*)^2 + (v^* - v_{\text{cref}}^*)^2]^{1/2} \quad (8)$$

Similarly, for each sample in the testing set, tristimulus values for the mixing of the selected waveband pair were used to obtain the L^* , u^* , v^* values for d_1 , d_2 , and d_3 calculation.

The Classification and Regression Trees (C&RT) decision tree statistic algorithm (Breiman, Friedman, Olshen, & Stone, 1984) was used to generate a series of if-then decision rules for classification of chicken conditions, using the AnswerTree 3.0 program (SPSS, Chicago, IL). To eliminate the risk of model over-fitting, values for a maximum tree level and for minimum node inputs were set as stopping rules. The d_1 , d_2 , and d_3 values for the chicken samples were used as decision tree inputs for model development to classify samples as belonging to the wholesome, systemically diseased, or cadaver categories.

Two sets of tests were performed to develop the decision tree model and evaluate the model performance. For first

test, a decision tree classification model was generated using 48 wholesome, 34 systemically diseased, and 22 cadaver chicken samples in the validation data set. The testing data set (55 wholesome, 32 systemically diseased, and 18 cadaver chicken samples) was then used to measure the performance of the model, independent of the validation data set. For the second test, 10-fold cross-validation was applied to all available chicken samples. This test was performed to evaluate an upper limit on model performance. The total samples, consisting of 103 wholesome, 66 systemically diseased, and 40 cadaver chickens samples, were first randomly split into 10 subgroups. A decision tree was generated with nine of the 10 subgroups and validated by the remaining subgroup. This process was repeated for 10 times, using each subgroup once for validation. The overall successful classification rate was then determined.

3. Results and discussion

3.1. Waveband selection for two-band color mixing application

Fig. 2 shows a plot of the minimum ΔE values for all possible waveband combinations (90,000 waveband pairs) for chicken sample classification. Because the contour plot is symmetric about the diagonal, only the upper half of the plot is discussed here. Four areas of peak ΔE values occur near the (λ_1, λ_2) waveband pairs of (427 nm, 664 nm), (450 nm, 642 nm), (453 nm, 589 nm), and (531 nm, 618 nm), for which the ΔE values are 4.35, 4.47, 4.34, and 3.76, respectively. These values are relatively high and suggest that the human eye can perceive differences shown by these four wavelength pairs; under CIE reference viewing conditions, color differences of 1.0 are considered “noticeable” and color differences greater than 5.0 are considered to be easily differentiable by human eye (CIE,

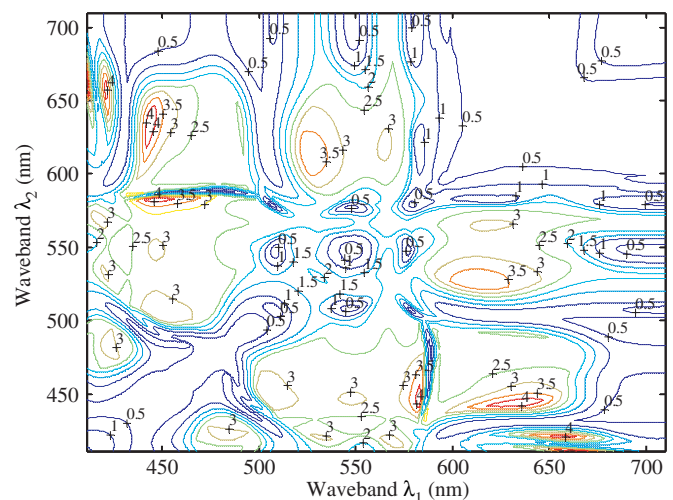


Fig. 2. Contour plot of minimum color difference values ΔE between waveband pairs centered at λ_1 and λ_2 for identification of chicken sample conditions.

1978). Although the highest ΔE value occurs for the second pair (450 nm, 642 nm), the third pair (453 nm, 589 nm) was chosen as being more suitable for differentiating chicken samples by two-band color-mixing due to its similarity to the waveband pair of (454 nm, 578 nm) used by Ding et al. (2005). Using the revised color appearance model (CIECAM 97s) proposed by Fairchild (2001), Ding et al. easily differentiated between six different chicken conditions in a color simulation using the waveband pair of (454 nm, 578 nm).

Fig. 3 shows the average spectra of wholesome, systemically diseased, and cadaver chickens. The visible region contains more useful spectral features for differentiating between conditions than the near-infrared region. For chicken meat, spectral features in the visible region are determined primarily by the relative amounts of three forms of myoglobin, i.e. deoxymyoglobin, metmyoglobin, and oxymyoglobin (Kinsman, Kotula, & Breidemestein, 1994; Swatland, 1989). The deoxymyoglobin and oxymyoglobin components coexist with metmyoglobin in both wholesome and unwholesome chicken meat. These three forms of myoglobin can inter-convert and may degrade through oxygenation, oxidation, and reduction reactions when external processes such as cold storage or cooking are applied (Liu & Chen, 2000). Bands associated with myoglobin include those in the areas of 430 nm, 440 nm, and 455 nm with deoxymyoglobin; 485 nm, 495 nm, and 505 nm with metmyoglobin; and 545 nm, 560 nm, and 575 nm with oxymyoglobin (Liu, Chen, & Ozaki, 2000). In general, wholesome and unwholesome chicken can be characterized by the relative amounts of these myoglobin forms, with wholesome chicken containing more oxymyoglobin and deoxymyoglobin, and unwholesome chicken containing more metmyoglobin (Liu & Chen, 2001).

3.1.1. Analysis of variance for L^* , u^* , and v^*

Table 1 summarizes analysis of variance for L^* in the groups wholesome, systemically diseased, and cadaver

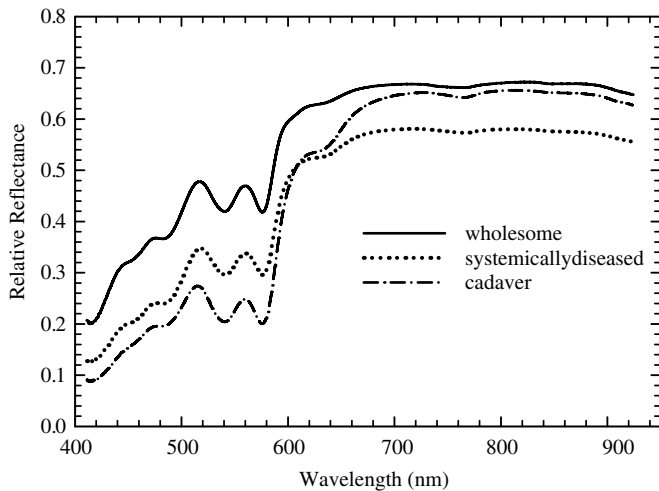


Fig. 3. Mean spectra of wholesome, systemically diseased, and cadaver chicken samples.

chicken samples. The L^* value is significantly different ($P < 0.05$) among the three groups. The Lilliefors test showed that the assumption of normality was valid. The data was normally distributed at a significance level of 0.05. Leven’s test showed statistically significant differences ($P < 0.05$) in variance existed within the data. Thus, the assumption of homoscedasticity was not met. Since not all assumptions for the analysis of variance were met, the Kruskal–Wallis test was performed to see whether results from the one-way analysis of variance would be confirmed or rejected. The Kruskal–Wallis test, which yielded a P value of 0.0, showed that differences existed between groups of wholesome, systemically diseased, and cadaver chickens, confirming the results of the one-way analysis of variance. In addition, Tukey–Kramer’s test (Table 4) for pair-wise comparisons of the mean L^* value showed that significant differences were found between the three groups.

The Lilliefors, Leven’s, and Kruskal–Wallis tests were also performed for analysis of u^* and v^* values for the three chicken categories to confirm results from one-way analysis of variance (Tables 2 and 3), which found that the u^* values among wholesome, systemically diseased, and cadaver samples were significantly different ($P < 0.05$). According to Tukey–Kramer’s test (Table 4), the mean u^* value of the cadaver category was significantly different ($P < 0.05$) from the other two categories but was not significantly different between the wholesome and systemically diseased chicken conditions. One-way analysis of variance found that the v^* values among the three categories were significantly different ($P < 0.05$). According to Tukey–Kramer’s test (Table 4), the mean v^* value of the wholesome category was significantly different ($P < 0.05$) from the other two, but the systemically diseased and cadaver categories were not significantly different from each other.

3.1.2. Classification accuracy

For the validation data set in the first test (Table 5), 95.8% (46 of 48) wholesome chickens, 95.5% (21 of 22)

Table 1
Analysis of variance for L^* values in the groups wholesome, systemically diseased, and cadaver chicken samples

| Source | Sum of squares | DF | Mean square | F | P |
|--------|----------------|-----|-------------|---------|---|
| Groups | 3758.4739 | 2 | 1879.2370 | 94.2851 | 0 |
| Error | 4105.8736 | 206 | 19.9314 | | |
| Total | 7864.3475 | 208 | | | |

Table 2
Analysis of variance for u^* values in the groups wholesome, systemically diseased, and cadaver chicken samples

| Source | Sum of squares | DF | Mean square | F | P |
|--------|----------------|-----|-------------|---------|-----------|
| Groups | 20.2777 | 2 | 10.1389 | 20.6921 | 6.4718E-9 |
| Error | 100.9375 | 206 | 0.48999 | | |
| Total | 121.2152 | 208 | | | |

Table 3
Analysis of variance for v^* values in the groups wholesome, systemically diseased, and cadaver chicken samples

| Source | Sum of squares | DF | Mean square | <i>F</i> | <i>P</i> |
|--------|----------------|-----|-------------|----------|-----------|
| Groups | 106.0634 | 2 | 53.0317 | 12.2585 | 9.3307E–6 |
| Error | 891.1796 | 206 | 4.3261 | | |
| Total | 997.2431 | 208 | | | |

Table 4
CIELUV characteristics of wholesome, systemically diseased, and cadaver chicken samples^a

| Poultry carcass condition | L^* | u^* | v^* |
|------------------------------------|------------------|-----------------|-----------------|
| Wholesome ^b | 76.01a (0.32) | 3.78a (0.06) | 9.86a (0.19) |
| Systemically diseased ^b | 69.47b (0.66) | 3.55a (0.10) | 8.98b (0.31) |
| Cadaver ^b | 65.89c (0.86) | 4.42b (0.12) | 8.05b (0.25) |

^a Means within the same column followed by different letters are statistically different ($P < 0.05$).

^b Values in parenthesis are standard error of the mean (103 wholesome, 66 systemically, and 40 cadaver chicken samples).

Table 5
Classification accuracy for the decision tree model development

| | | Actual chicken condition | | |
|------------------|-----------------------|--------------------------|-----------------------|---------|
| | | Wholesome | Systemically diseased | Cadaver |
| Model prediction | Wholesome | 46 | 0 | 1 |
| | Systemically diseased | 1 | 34 | 0 |
| | Cadaver | 1 | 0 | 21 |

cadaver chickens, and 100% (34 of 34) systemically diseased chickens were successfully classified. The model was then able to successfully classify 94.6% (52 of 55) of wholesome chickens, 100% (18 of 18) of cadaver chickens, and 90.6% (29 of 32) of systemically diseased chickens in the testing data set (Table 6). For the second test using 10-fold cross-validation (Table 7), 98.1% (101 of 103) of wholesome chickens, 97.5% (39 of 40) of cadaver chickens, and 93.9% (62 of 66) of systemically diseased chickens were successfully classified.

Table 6
Classification accuracy for the decision tree model test

| | | Actual chicken condition | | |
|------------------|-----------------------|--------------------------|-----------------------|---------|
| | | Wholesome | Systemically diseased | Cadaver |
| Model prediction | Wholesome | 52 | 0 | 0 |
| | Systemically diseased | 1 | 29 | 0 |
| | Cadaver | 2 | 3 | 18 |

Table 7
Classification accuracy for 10-fold cross-validation of the decision tree model

| | | Actual chicken condition | | |
|------------------|-----------------------|--------------------------|-----------------------|---------|
| | | Wholesome | Systemically diseased | Cadaver |
| Model prediction | Wholesome | 101 | 0 | 1 |
| | Systemically diseased | 1 | 62 | 0 |
| | Cadaver | 1 | 4 | 39 |

It is necessary to evaluate the model by two practical concerns; public health and economic benefit. For the former, the model should not misclassify any unwholesome chickens into the wholesome category. For the latter, the model should eliminate the misclassification of wholesome chickens into any unwholesome category. Misclassification among unwholesome chicken categories, in this study, between cadaver and systemically diseased, may not be very important.

The classification results achieved using the color difference indices based on two-band color-mixing at 453 nm and 589 nm show potential for addressing public health concerns. Among the classification of the validation data set, the testing data set, and testing using 10-fold cross-validation, only 1 unwholesome bird (a cadaver sample) was classified as wholesome.

For economic concerns, the rate of misclassification of wholesome birds was only 2 of 48 for model validation, 3 of 55 for model testing, and 2 of 103 for 10-fold cross validation. This suggests that a low level of misclassified birds could be achieved using two-band color mixing for chicken inspection, to the benefit of poultry processors.

4. Conclusions

A two-band color-mixing technique was demonstrated for identifying wholesome, systemically diseased, and cadaver chicken conditions for samples cut from the breast area of chicken carcasses. All pairwise combinations of 10-nm wavebands in the region of 416–715 nm were examined for differentiating between chicken conditions using the two-band color-mixing technique, and the waveband pair of (453 nm, 589 nm) was found most suitable for identifying the chicken conditions. Significant color differences between wholesome, systemically diseased, and cadaver chicken samples, as indicated by L^* , u^* , and v^* values, were found using one-way analysis of variance. Color difference values calculated from L^* , u^* , and v^* values were used as inputs for decision-tree classification models. For a validation data set containing 48 wholesome, 34 systemically diseased, and 22 cadaver chicken samples, the classification accuracies achieved were 95.8% for wholesome, 95.5% for systemically diseased, and 100% for cadaver. For an independent testing set of 55 wholesome, 32 systemically diseased, and 18 cadaver chicken samples, the classification

accuracies were 94.6%, 100%, and 90.6%, respectively. For model testing using 10-fold cross-validation, classification accuracies achieved were 98.1% for wholesome, 97.5% for systemically diseased, and 93.9% for cadaver. These results show that the two-band color-mixing technique shows potential for use in addressing public health concerns, as only 1 unwholesome (a cadaver sample) bird was misclassified as wholesome.

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