Rapid Softness Prediction and Microbial Spoilage Visualization of Whole Tomatoes by Using Hyper/Multispectral Imaging

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Abstract: The choice of selecting fruit for canned whole tomatoes is driven by several quality attributes including sweetness, acidity, and softness of tomatoes. Moreover, tomatoes can be contaminated with a variety of molds during cultivation, harvest, and transportation. Conventional evaluation operations for tomato softness and microbial spoilage are usually time-consuming, destructive, and high-cost. One strategy for rapid tomato sorting is to utilize hyper/multispectral imaging. This paper proposes to improve on traditional broad-band infrared imaging of existing color and dirt sorters by increasing the spectral resolution of the information collected. The findings of this study will characterize the potential of the technology in terms of predicting tomato softness and identification of tomato microbial spoilage for further development by the industry.

Keywords: tomato; spectral imaging; softness; microbial spoilage

1. Introduction

Tomatoes are the second most grown and consumed vegetable in the United States. Tomatoes typically ripen gradually from the inside out, involving dramatic changes in color, flavor, texture, and composition (such as chlorophyll replaced by carotenoids, mainly lycopene) [1]. The majority of tomatoes consumed are thermally treated and processed into various products, such as canned whole tomatoes and tomato paste. The choice of selecting fruit for canned whole tomatoes is driven by several quality parameters, such as the acidity and softness of the tomatoes [2]. Soft tomatoes, which are not suitable for canned foods, are usually detected by handling to feel the texture in the food industry. When soft tomatoes are detected, they can be delivered to the tomato paste production line instead of the canned tomato line. Tomato softening is a consequence of loss of turgor, or degradation of polymer constituents of the cell wall [3]. The breakdown of cell walls and the structural components of tomatoes has a great impact on the texture of a tomato.

Currently, the online detection of the softness and microbial spoilage of the whole tomato is still full of grand challenges. Buyers, processors, and scientists who focus on tomato breeding and quality control strategies need a rapid technology to improve the efficiency of tomato quality evaluation. For tomato processors, having a method to automatically determine the quality of tomatoes is essential to establish a purchase price, and it must be performed very quickly. However, the mechanical texture analyzer with a destructive nature (puncture testing) and low efficiency (flat plate compression) is not the ideal solution for online whole fruit softness detection [1,4–6]. Tomato mold contamination can occur at different times from the farm to the table [7]. These infected fruits are discarded as waste or organic fertilizer for the soil. In a previous study, fungus in infected tomato fruit was accurately detected by using enzyme-linked immunosorbent assay (ELISA) [8]. However, this kind of technique requires laborious steps, such as extraction and derivatization, and is destructive and time-consuming [9]. It takes a long time to obtain the results, and the analysis of the sample is of high-cost. In the future,
the sustainable development of advanced agriculture will definitely require the novel development and in-depth application of smart agricultural technology [10–12].

Rapid methods for the assessment of the safety and quality of tomato products have made advances during the last 15 years [13]. Infrared spectroscopy was used as an efficient method to measure sugars, acids, and soluble solids content (SSC) in processing tomatoes [14,15]. The cost of point spectral instruments is generally much lower than that of spectral imaging systems. Nevertheless, the drawback of portable spectroscopic methods is that spectral data are collected from a single point or from a small portion of tested samples which may not guarantee data accuracy and representativeness [16–18]. The point spectroscopy would provide a mean spectrum of several single points (average measurement) of a sample, irrespective of the area of the sample scanned. As the spectra collected are averaged to provide a single spectrum, the heterogeneous information on spatial distribution (such as spoilage defect) within the sample is thus lost for point spectroscopy.

Hyper/multispectral imaging can integrate the characteristics of spectroscopy and imaging into one system, and provide heterogeneous information captured from one end of a sample to the other, which has been acknowledged as a more advanced means to meet the demand in food industries [19–23]. In a recent study, hyperspectral imaging (HSI) was successfully used to detect and quantify fish microbial spoilage, yielding determination coefficients in prediction ($R^2_P$) of more than 0.90 [24]. The cracking defects on cherry tomatoes were identified by HSI coupled with ultraviolet (UV) (320–400 nm) light as an excitation source for producing fluorescence emissions in 400 to 700 nm [25]. It was indicated that the fluorescence emission could be a significant attribute for the discrimination of defective sites from the sound surface of the cherry tomatoes. Due to the destruction of tissues by fungal contamination (alternaria or stachybotrys), the chemical compositions including protein and lipid content, and physical parameters including density, volume and weight of tomatoes will change, and this change can be detected in spectral features [26]. Autofluorescence analysis either using steady-state or time-resolved fluorescence emission has been employed as a tool for the evaluation of organoleptic/nutraceutical content of many foods [27,28]. Hyperspectral fluorescence based on a blue laser (408 nm) was associated with fruit firmness [29]. Multispectral imaging (MSI) has also been effectively applied to detect other quality traits of plant foods such as color, SSC, pH, and antioxidants [30,31]. The results obtained using such methods are comparable to those using reference methods. However, there are rare studies on rapid assessments of whole tomato softness due to ripening and senescence, and microbial spoilage (i.e., fungal infection) and the discoloration that it causes based on hyper/multispectral imaging. The study was designed to improve on traditional broad-band infrared imaging of existing color and dirt sorters by increasing the spectral resolution of the information collected.

2. Objective of the Project

The specific objectives of this project are to:

(a) Establish both HSI and MSI systems for whole-tomato softness and microbial spoilage assessments
(b) Acquire the reflectance and fluorescence spectral images of both unpeeled and peeled fruit from three tomato cultivars
(c) Develop statistical models to predict the softness of measured tomatoes and to ascertain the effects of tomato peel and variety on the optical vision prediction of fruit softness
(d) Select feature wavelengths to determine optimal threshold values for microbial spoilage detection
(e) Visualize the distribution of microbial spoilage on unpeeled and peeled whole tomatoes.

The findings of this preliminary study will characterize the potential of the technology in terms of predicting tomato softness and identification of tomato microbial spoilage for further development by the industry.
3. Plans and Procedures

Two spectral vision systems in terms of HSI and MSI systems will be developed. The lab-based line-scan HSI system equipped with two light sources can operate in either reflectance or fluorescence mode. The core components of the HSI system are a computer, a spectrograph with a wavelength range of 384–810 nm (ImSpector V8_4_102, Spectral Imaging Ltd., Oulu, Finland), a charge-coupled device (CCD) camera (Photometrics CoolSNAPcf, Roper Scientific® Photometrics, Tucson, AZ, USA), two tungsten–halogen bulbs for reflectance measurement, and two kinds of UV excitation sources (365 nm and 385 nm) for fluorescence measurement. The area-scan MSI system mainly consists of a computer, a quantum scientific imaging (QSI) 660 cooled CCD camera equipped with a filter wheel, wavelength filters, and excitation sources (such as UV, Green, Blue, and Red lights for excitation). Wavelengths (409, 466, 520, 542, 575, 579, 675, and 775 nm) are the filters available in the lab which may relate to specific molecules.

A total of 504 tomatoes produced from the farm will be used in this research. Healthy red tomatoes (252 for three cultivars) and microbial spoilage samples (252 for three cultivars) will be respectively collected at a grading station seven times (72 samples at one time). These 72 samples will be tested over a period of 1 week to obtain a greater variation in tomato softness and microbial spoilage. Alternaria and Stachybotrys (black mold) are two common types of mold in tomatoes in CA. Such tomatoes will be obtained from the mold bucket at the grading station. Samples will be first cleaned using a water tank dip then sprayed with water. The washed tomatoes will be first transported to a ventilated place to remove the moisture on the skin, then moved into storage at about 65 °F until completion of the experiment. In each test, tomato samples will be divided into two equal parts. Samples in one group will be peeled, with tomatoes of another group not being peeled. Tomato images of both groups will be captured by the above two spectral imaging systems.

Individual tomatoes will be oriented so the stem–calyx axis is kept horizontal and also perpendicular to the imaging system. The defective samples will be arranged to expose the microbial spoilage for imaging. Hyperspectral reflectance and fluorescence images will be obtained from the fruit in sequence by performing line scans at the surface of the tomatoes. Additionally, multispectral fluorescence data from the same test samples based on different excitation sources and filters will be acquired. Analyses of the spectral images will be performed using Matlab and ImageJ software. The high-resolution spectral reflectance and fluorescence from regions of interest (ROIs), such as good and defective surface areas, will be extracted in software. Different computer algorithms will be developed for selection of the most significant waveband combinations for defect detection and classification. Following the waveband selection, the feature wavebands will be used to determine the threshold values for microbial spoilage detection. The threshold value at which the classification accuracy is highest will be determined.

After spectral reflectance and fluorescence images are acquired from both peeled and unpeeled samples, the softness or hardness of whole tomatoes will be assessed by using a TA.XT2 Texture Analyzer (Texture Technologies Corp., Scarsdale, NY, USA) in a compression test using a cylindrical plate (100 mm high, 52 mm internal diameter) [32]. The probe will travel at a constant speed of 2 mm/s while compressing and extruding the sample down to 10 mm above the bottom of the cell [33,34]. Softness values will be taken as the area under the curve after the maximum peak was reached. Spectral data will be analyzed. Data processing includes the preprocessing of spectral data and the development and validation of calibration models. The information from spectral images will be linked, based on multivariate analyses, to the measured reference values, and optimal prediction models will be generated to quantify tomato softness. Statistical analysis of variance (ANOVA) and multivariate analysis will be performed using SPSS and Matlab software.

Based on the new method found in the study, it is expected that an online computer vision system mounted with an advanced back-illuminated complementary metal–oxide semiconductor (CMOS) camera (Prime 95B 25MM, Photometrics) with a price of $30,000 will be eventually established to enhance the detection speed of spoilage and softness of
tomato products during real-time applications. As the latest camera in use, the new Prime 95B 22MM camera can acquire images with low illumination levels, and high resolution (number of pixels) and speeds (frame rate), such as images captured in 1 to 3 ms, which is more suitable for industrial machine vision detection speeds. For example, it could measure tomatoes at above 1 mph when they move down the processing line.

4. Conclusions

This study could be of benefit to the grand challenges facing planetary health—local and global environments and human health. The developed spectral imaging system will characterize the potential for automatic quality detection of whole-tomato products. The results will be summarized to produce a research paper including reference tables that can be used by the industry as a guide to better understand the advanced techniques used for softness and microbial spoilage detection of whole tomato.

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References

1. Barrett, D.M.; Garcia, E.; Wayne, J.E. Textural Modification of Processing Tomatoes. *Crit. Rev. Food Sci. Nutr.* 1998, 38, 173–258. [CrossRef]
2. Frez-Muñoz, L.; Steenbekkers, B.; Fogliano, V. The Choice of Canned Whole Peeled Tomatoes is Driven by Different Key Quality Attributes Perceived by Consumers Having Different Familiarity with the Product. *J. Food Sci.* 2016, 81, S2988–S2996. [CrossRef]
3. Tucker, G.A.; Robertson, N.G.; Grierson, D. Purification and changes in activities of tomato pectinesterase isoenzymes. *J. Sci. Food Agric.* 1982, 33, 396–400. [CrossRef]
4. Anthon, G.E.; Blot, L.; Barrett, D.M. Improved Firmness in Calcified Diced Tomatoes by Temperature Activation of Pectin Methylesterase. *J. Food Sci.* 2005, 70, C342–C347. [CrossRef]
5. Su, W.-H.; Bakalis, S.; Sun, D.-W. Fingerprinting study of tuber ultimate compressive strength at different microwave drying times using mid-infrared imaging spectroscopy. *Dry. Technol.* 2019, 37, 1113–1130. [CrossRef]
6. Su, W.-H.; Bakalis, S.; Sun, D.-W. Fourier transform mid-infrared-attenuated total reflectance (FTMIR-ATR) microspectroscopy for determining textural property of microwave baked tuber. *J. Food Eng.* 2018, 218, 1–13. [CrossRef]
7. Doan, H.K.; Perez, K.; Davis, R.M.; Slaughter, D.C. Survey of Molds in California Processing Tomatoes. *J. Food Sci.* 2016, 81, M2785–M2792. [CrossRef]
8. Thornton, C.R.; Slaughter, D.C.; Davis, R.M. Detection of the sour-rot pathogen Geotrichum candidum in tomato fruit and juice by using a highly specific monoclonal antibody-based ELISA. *Int. J. Food Microbiol.* 2010, 143, 166–172. [CrossRef]
9. Su, W.-H.; Arvanitoyannis, I.S.; Sun, D.-W. Trends in Food Authentication. In *Modern Techniques for Food Authentication*; Elsevier: Amsterdam, The Netherlands, 2018; pp. 731–758.
10. Su, W.-H. Advanced Machine Learning in Point Spectroscopy, RGB- and Hyperspectral-Imaging for Automatic Discriminations of Crops and Weeds: A Review. *Smart Cities 2020*, 3, 767–792. [CrossRef]
11. Su, W.-H. Crop plant signaling for automated crop/weed identification: A systematic review and new concept. *Artif. Intell. Agric.* 2020, 4, 262–271.
12. Su, W.-H.; Zhang, J.; Yang, C.; Page, R.; Szinyei, T.; Hirsch, C.D.; Steffenson, B.J. Automatic Evaluation of Wheat Resistance to Fusarium Head Blight Using Dual Mask-RCNN Deep Learning Frameworks in Computer Vision. *Remote Sens.* 2021, 13, 26. [CrossRef]
13. Barrett, D. Future Innovations in Tomato Processing. In Proceedings of the XIII International Symposium on Processing Tomato, Sirmione, Italy, 8–11 June 2014; Volume 1081, pp. 49–55.
14. Slaughter, D.; Barrett, D.; Boersig, M. Nondestructive Determination of Soluble Solids in Tomatoes using Near Infrared Spectroscopy. *J. Food Sci.* 1996, 61, 695–697. [CrossRef]
15. Wilkerson, E.D.; Anthon, G.E.; Barrett, D.M.; Sayajon, G.F.G.; Santos, A.M.; Rodriguez-Saona, L.E. Rapid Assessment of Quality Parameters in Processing Tomatoes Using Hand-Held and Benchtop Infrared Spectrometers and Multivariate Analysis. *J. Agric. Food Chem.* 2013, 61, 2088–2095. [CrossRef]
16. Su, W.-H.; He, H.-J.; Sun, D.-W. Non-Destructive and rapid evaluation of staple foods quality by using spectroscopic techniques: A review. *Crit. Rev. Food Sci. Nutr.* 2017, 57, 1039–1051. [CrossRef]
17. Su, W.-H.; Sun, D.-W. Mid-infrared (MIR) Spectroscopy for Quality Analysis of Liquid Foods. *Food Eng. Rev.* 2019, 11, 142–158. [CrossRef]
18. Su, W.-H.; Bakalis, S.; Sun, D.-W. Potato hierarchical clustering and doneness degree determination by near-infrared (NIR) and attenuated total reflectance mid-infrared (ATR-MIR) spectroscopy. *J. Food Meas. Charact.* 2019, 13, 1218–1231. [CrossRef]

19. Su, W.-H.; Sun, D.-W. Fourier Transform Infrared and Raman and Hyperspectral Imaging Techniques for Quality Determinations of Powdery Fruits: A Review. *Compr. Rev. Food Sci. Food Saf.* 2018, 17, 104–122. [CrossRef]

20. Su, W.-H.; Sun, D.-W. Multivariate analysis of hyper/multi-spectra for determining volatile compounds and visualizing cooking degree during low-temperature baking of tubers. *Comput. Electron. Agric.* 2016, 127, 561–571. [CrossRef]

21. Su, W.-H.; Bakalis, S.; Sun, D.-W. Chemometric determination of time series moisture in both potato and sweet potato tubers during hot air and microwave drying using near/mid-infrared (NIR/MIR) hyperspectral techniques. *Dry. Technol.* 2019, 38, 806–823. [CrossRef]

22. Su, W.-H.; Sun, D.-W. Advanced Analysis of Roots and Tubers by Hyperspectral Techniques. In *Advances in Food and Nutrition Research*; Elsevier: Amsterdam, The Netherlands, 2019; Volume 87, pp. 255–303.

23. Su, W.-H.; Bakalis, S.; Sun, D.-W. Chemometrics in tandem with near infrared (NIR) hyperspectral imaging and Fourier transform mid infrared (FT-MIR) microspectroscopy for variety identification and cooking loss determination of sweet potato. *Biosyst. Eng.* 2019, 180, 70–86. [CrossRef]

24. Cheng, J.-H.; Sun, D.-W. Rapid and non-invasive detection of fish microbial spoilage by visible and near infrared hyperspectral imaging and multivariate analysis. *LWT* 2015, 62, 1060–1068. [CrossRef]

25. Cho, B.-K.; Kim, M.S.; Baek, I.-S.; Kim, D.-Y.; Lee, W.-H.; Kim, J.; Bae, H.; Kim, Y.-S. Detection of cuticle defects on cherry tomatoes using hyperspectral fluorescence imagery. *Postharvest Biol. Technol.* 2013, 76, 40–49. [CrossRef]

26. Egging, V.; Nguyen, J.; Kurouski, D. Detection and Identification of Fungal Infections in Intact Wheat and Sorghum Grain Using a Hand-Held Raman Spectrometer. *Anal. Chem.* 2018, 90, 8616–8621. [CrossRef] [PubMed]

27. Lemos, M.A.; Sárniková, K.; Bot, F.; Anese, M.; Hungerford, G. Use of Time-Resolved Fluorescence to Monitor Bioactive Compounds in Plant Based Foodstuffs. *Biosensors* 2015, 5, 367–397. [CrossRef]

28. Sherlock, B.E.; Harvestine, J.N.; Mitra, D.; Haudenschild, A.; Hu, J.; Athanasiou, K.A.; Leach, J.K.; Marcu, L. Nondestructive assessment of collagen hydrogel cross-linking using time-resolved autofluorescence imaging. *J. Biomed. Opt.* 2018, 23, 036004. [CrossRef]

29. Noh, H.K.; Lu, R. Hyperspectral laser-induced fluorescence imaging for assessing apple fruit quality. *Postharvest Biol. Technol.* 2007, 43, 193–201. [CrossRef]

30. Su, W.H.; Sun, D.W. Multispectral Imaging for Plant Food Quality Analysis and Visualization. *Compr. Rev. Food Sci. Food Saf.* 2018, 17, 220–239. [CrossRef] [PubMed]

31. Peng, Y.; Lu, R. Prediction of apple fruit firmness and soluble solids content using characteristics of multispectral scattering images. *J. Food Eng.* 2007, 82, 142–152. [CrossRef]

32. Lee, S.-Y.; Luna-Guzman, I.; Chang, S.; Barrett, D.; Guinard, J.-X. Relating descriptive analysis and instrumental texture data of processed diced tomatoes. *Food Qual. Prefer.* 1999, 10, 447–455. [CrossRef]

33. Bourne, M.; Moyer, J. Extrusion principle in texture measurement of fresh peas. *Food Technol.* 1968, 22, 81.

34. Lu, R.; Guyer, D.E.; Beaudry, R.M. Determination of firmness and sugar content of apples using near-infrared diffuse reflectance 1. *J. Texture Stud.* 2000, 31, 615–630. [CrossRef]