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Development of simple algorithms for the detection of fecal contaminants on apples from visible/near infrared hyperspectral reflectance imaging ☆

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Abstract

Hyperspectral reflectance images of two cultivars of apples were acquired after fecal treatments at three different concentrations to explore the potential for the detection of fecal contaminants on apple surfaces. Region of interest (ROI) spectral features of fecal contaminated areas showed a reduction in reflectance intensity compared to those of uncontaminated skins. Large spectral differences between uncontaminated and fecal contaminated skins of two types of apples occurred in the 675–950 nm visible/NIR region, which provided the basis for developing universal algorithms in the detection of fecal spots. Comparison of a number of processed images revealed that a dual-band ratio ($Q_{725/811}$) algorithm could be used to identify fecal contaminated skins effectively. The result was most important as the two bands are away from the absorptions of natural pigments (such as chlorophylls and carotenoids), and hence can reduce the influence from color variations due to different apple cultivars.

Keywords: Hyperspectral imaging spectroscopy; Image processing; Algorithm; Principal component analysis; Apple; Fecal contamination; Food safety

1. Introduction

Contamination of apple products with bacterial food-borne pathogens can potentially occur as a result of exposure of apples to fecal materials during the growing and harvesting phases. Animal feces are the most likely source of pathogenic *E. coli* O157: H7 contamination. In addition, the potential of contamination increases with physical damages on apples, such as lesions and bruises, which provide a site for bacterial growth. Cleaning processes can reduce, but are unlikely to eliminate, pathogens from the surfaces of produce even if antimicrobial chemicals are contained in the wash water (FDA, 1998). Bacterial pathogens can be transmitted to humans by consumption of contaminated apples or raw (unpasteurized) apple juice/cider. There have been several reported food-borne illness outbreaks attributed to unpasteurized apple juice and cider (CDC, 1996, 1997). These outbreaks have raised the concerns of public health officials and apple cider/juice producers.

To ensure healthy and safe apple products to the consumers, the US Food and Drug Administration (FDA) has issued an HACCP system to minimize the likelihood of bacterial pathogens in fruit juices, and also identified an urgent need to develop methods for the detection of fecal matters on apples (FDA, 2001). Preventing apples with visible fecal contamination from entering the washer tank is critical for preventing cross-contamination of other apples. Thus, removal of fecal contaminated apples, before

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entering the washing pool, has been suggested by FDA guidance on good agricultural practices (GAPs) and good manufacturing practices (GMPs) for fruits and vegetables (FDA, 1998).

Currently, inspection of fecal contamination is through visual observation over an inspection table. Inspectors use the guidelines of Current Good Manufacturing Practices (CGMPs) to prevent apples with visible fecal contaminants from entering the next step. Current visual inspection is labor intensive and prone to human error and inspector-to-inspector variation. Therefore, researchers at the US Department of Agriculture (USDA) Agricultural Research Services have been developing hyperspectral reflectance imaging system for the detection of fecal contaminated apples (Kim et al., 2002; Mehl, Chao, Kim, & Chen, 2002; Mehl, Chen, Kim, & Chan, 2004). The preliminary results have demonstrated that spectral imaging technique can be used effectively for detecting fecal spots on apples in the visible and near-infrared (NIR) region.

To implement hyperspectral reflectance imaging in potential on-line inspection, less spectral bands (usually two or three) were selected for the design of rapid sensing instruments; namely multispectral imaging systems. From hyperspectral reflectance imaging spectra, the essential bands were obtained through a number of data analysis, such as performing principal component analysis (Kim, Chen, & Mehl, 2001), and observing the separations visually on images at specific wavelengths (Mehl et al., 2004). The representative bands should not only reflect the chemical/physical information, but also maintain the successive discrimination and classification efficiency.

The main objectives of this study were to obtain the characteristic bands for fecal contaminated apples and to develop simple algorithms for the detection of fecal spots. The ultimate purpose was to lead to the development of faster and more efficient multispectral techniques for real-time inspection of fecal contaminants for apple related product safety.

2. Materials and methods

2.1. Apples and fecal contamination treatments

A number of two cultivars of apples (Golden Delicious and Red Delicious) were collected from containers that were used to store the harvested apples in Pennsylvania (Rice Fruit Co., Gardners, PA). The apples were transported to the Laboratory in Beltsville, MD, and kept in a cold storage room (2–4 °C). To address the variations among the apples, 96 apples from each cultivar were used randomly.

Cattle feces were selected to represent the fecal contaminant, as it is one of the most common sources of fecal contamination (Cody et al., 1999). Fresh cow feces were obtained from a pasture at USDA Beltsville dairy facility in Maryland. They were diluted with drinkable water (H_2O) to produce three different concentrations of 50%, 5%, and 0.5% (weight/weight, w/w). Then, three fecal spots were formed by applying three solutions to one side of each apple with the use of a pipette. After the evaporation of water, two fecal spots in circles, with the size of around 1 cm in diameter, were observed clearly. The apparent two spots represented the deposition from two concentrated solutions, 50% and 5%, respectively. For the collection of images, a set of 12 fecal treated apples was placed on a tray painted with non-fluorescent, flat black paint to minimize background scattering.

2.2. Hyperspectral imaging acquisition and image analysis

A hyperspectral reflectance imaging system developed by the USDA Instrumentation and Sensing Laboratory was used to scan the apples (Kim et al., 2001). It was operated in a line-by-line scan spectrograph with a spectral resolution of approximately 7-nm full width at half maximum (FWHM). A thermo-electrically cooled electron multiplying charge-coupled device (EMCCD) camera with 288 (vertical) × 560 (horizon) pixels (Andor, Inc., South Windsor, Conn.) was used, and the effective spectral and spatial dimensions were limited to 112 pixels (channels) with 2× binning and 460 pixels without binning, respectively. It produced an image cube of 460 × 1200 spatial and 112 spectral bands for each group of 12 apples. The spectral wavelength range was in visible/NIR region of 447–951 nm with a 4.5 nm interval. Two 150 W halogen lamps were applied to provide the illumination for image collection. A Spectralon™ white reference panel with nearly 99% reflectance (Labsphere, North Sutton, NH, USA) was employed as a reference. The camera dark image and the white reflectance image were recorded prior to the acquisition of the hyperspectral images. During the scanning process, room lights were turned off to prevent interference from ambient light.

In-house laboratory-developed software (Kim et al., 2001) and the commercial ENVI 3.2 software package (Research Systems, Inc., Boulder, CO) were used for the analysis of hyperspectral images. Prior to algorithm and principal component image analysis, images at 665 nm were processed with binary function at a threshold of 0.4% of reflectance intensity to create a mask for the apple surface in each image. The threshold value was determined by visual observation so as to exclude the image background.

During principal component analysis (PCA) of images, a correlation matrix of the image is calculated (Lay, 2002). Then this correlation matrix, a diagonal matrix, is used to compute the eigenvalues. The eigenvalues are equivalent to the variance of each principal component (PC) image. These PC images are ordered in the decreasing degree of variance sizes, where first PC accounts for the largest variance. Hence, PCA transforms the original data set into a set of new un-correlated linear combinations of the original data with much less variables.