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Evaluation of the freshness of fresh-cut green bell pepper (*Capsicum annuum* var. *grossum*) using electronic nose

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ABSTRACT

This study investigated a freshness assessment method utilizing electronic nose (e-nose) for fresh-cut green bell pepper (*Capsicum annuum* var. *grossum*) stored at 7 ± 1 °C. Physicochemical results showed that the fresh-cut green bell peppers were fresh until 5 days and became spoiled at 7 days, particularly as evidenced by a surge in aerobic plate count and malondialdehyde content on subsequent days. The e-nose data combined hierarchical cluster analysis (HCA) can preliminarily distinguish between fresh (days 0, 1, 3 and 5) and spoiled (days 7 and 9) samples. Principal component analysis (PCA) result showed that days 0 and 1 samples were mixed together in PCA plot, and the other different groups can be obtained according to the different sampling days. Partial least squares (PLS) statistical model (R^2 = 0.9783, RMSE = 0.3317) was used to correlate the e-nose data with the aerobic plate counts. The results suggested the promising possibility of e-nose system for monitoring freshness of fresh-cut green bell pepper.

1. Introduction

Green bell pepper (*Capsicum annuum* var. *grossum*) is rich in vitamins which is popular crops throughout the world for fresh market consumption (Singh, Giri, & Kotwaliwale, 2014). A good storage temperature for some peppers is 7–13 °C, because some cultivars are susceptible to chilling injury when stored below 7 °C (González-Aguilar, Ayala-Zavala, Ruiz-Cruz, Acedo-Félix, & Dıaz-Cinco, 2004). Thanks to demands of convenience and healthiness, sales of minimally processed vegetables with a fresh-like quality are rapidly increasing. However, fresh-cut vegetables are highly perishable and their quality deterioration usually occurs in a short time. The storage life of fresh-cut pepper is limited by various factors like different storage conditions, fresh-cut processes and species which influence the freshness loss and spoilage pattern of fresh-cut vegetables.

Traditional methods used to determine shelf life of fresh-cut vegetable are based on chemical, microbiological, physical and sensory indices, such as phenolic compounds, molds and bacterial counts, texture, color, and sensory evaluation (Rodoni, Zaro, Hasperué, Concellón, & Vicente, 2015). Most traditional methods are time-consuming and require skilled personnel. Moreover, gas chromatography-mass spectrometry (GC-MS) has been applied to analyze the volatile compounds of foods. However, it is neither feasible to realize rapid diagnosis of volatile profiles.

Electronic nose (e-nose) is composed of a variety of gas sensors that interact with odor molecules to generate electronic signals called sensor responses. Then, the responses are collected by a computer system and handled using multivariate data analysis methods (Kiani, Minaei, & Ghasemi-Varnamkhasti, 2016a). Data analysis methods of sensors' response involve partial least squares (PLS), cluster analysis (CA), principal component analysis (PCA), linear discriminate analysis (LDA), functional discriminate analysis (FDA), and so on which are considered as linear approaches, while fuzzy logic, artificial neural network (ANN) and probabilistic neural network (PNN) are based on nonlinear methods (Loutfi, Coradeschi, Mani, Shankar, & Rayappan, 2015).

The use of e-nose is a promising method for the simple measurement of maturity and other quality indicators of fruits and vegetables. Application of e-nose to evaluate fruit ripening stage during storage has been studied over the past years (Hernández Gómez, Wang, Hu, & García Pereira, 2007), because e-nose

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sensors are able to detect volatile aroma related to fruit ripening. Metal oxide semiconductor (MOS) sensors, particularly SnO_2 sensors, are able to detect ethylene which is heavily involved in modulating the volatile emissions of fruits. Thus the MOS e-nose can successfully distinguish two different stages of apricots maturity after simulated shelf life storage (Defilippi et al., 2009). Lebrun, Plotto, Goodner, Ducamp, and Baldwin (2008) used e-nose for identifying volatiles of mango fruit in order to differentiate its harvest maturity. Gómez, Wang, Hu, and Pereira (2008) found that e-nose could assess tomato ripening stage during storage by PCA and LDA, but showed poor prediction performance on fruit quality factors like soluble solids content, pH and puncture force. Moreover, rapid diagnosis of microbial contamination of food products by e-nose has been proposed. In a study of Gobbi et al. (2015), e-nose with four MOS sensors was available for rapid diagnosis of Enterobacteriaceae in vegetable soups. Results of Giovenzana, Beghi, Buratti, Civelli, and Guidetti (2014)'s work demonstrated that a commercial portable e-nose composed of 10 MOS sensors was able to monitor freshness decay of fresh-cut *Valerianella locusta* L. based on evolution of the aroma profile during storage.

Despite these encouraging examples, e-nose application for freshness assessment of fresh-cut vegetables is still scarce. Therefore, it will be beneficial to develop reliable methods to assess freshness of fresh-cut vegetables and to establish evaluation criteria of their quality. The objective of this study is to establish an easy-to-use method based on e-nose for freshness assessment of fresh-cut green bell pepper.

2. Materials and methods

2.1. Vegetable materials

Physiologically mature green bell peppers (*Capsicum annuum* var. *grossum*) were purchased in located market, Wuxi, China. Peppers without damage and defects were washed, cut in pieces of 2.2 ± 0.3 cm × 8.0 ± 0.5 cm, dipped in 0.1 mL/L sodium hypochlorite solution for 5 min, washed again with water, and weighted within 150 ± 10 g. Then they were packaged in circular polyethylene terephthalate (PET) trays and capsed (15.3 cm × 5.3 cm). Forty-eight trays containing samples were stored at 7 ± 1 °C.

2.2. Determinations

Samples were analyzed for traditional physicochemical indices and e-nose responses to green bell pepper samples at different storage periods (0, 1, 3, 5, 7 and 9 days). Eight trays with samples were prepared for each point of determinations. Aerobic plate count and e-nose test were conducted in at least eight repeats from these eight trays and other traditional physicochemical experiments were conducted in at least three repeats randomly from three different trays.

2.2.1. Weight loss

Physiological loss in weight is leading to shriveling appearance for fruits and vegetables. Weight loss was measured by a laboratory level weighting scale (Precision Balance XPE303S, Mettler-Toledo International Inc., Switzerland) at different storage period. The weight loss can be expressed on a wet weight basis (g/100 g) by the difference in initial and final weights of the sample (Singh et al., 2014).

2.2.2. Chlorophyll content

Chlorophyll content is an important indicator for green vegetables, which is related to their color attribute. Determination of chlorophyll content was adapted from Sgroppo and Pereyra (2009). Four g of samples was homogenized with 100 mL of 0.8 L/L acetone (mixing 4 L acetone and 1 L distilled water) and filtered through filter paper. The absorbance of filtrate was determined using spectrophotometer (UV2600, TECHCOMP, China) at 645 nm and 663 nm. Results are calculated using Eq. (1).

$$\text{Chlorophyll content (mg/g)} = (20.21A_{645}+8.02A_{663}) \times V \times N/(W \times 1000) \quad (1)$$

Where A_{645} and A_{663} are the absorbance values at 645 nm and 663 nm, respectively. V is the volume of the extract solution (mL), N is the dilution factor, W is the mass of the sample (g).

2.2.3. Aerobic plate count

Microbiological analysis is also one of important indexes for judging the quality of fresh-cut vegetables. Microbiological examination was determined by aerobic plate count according to National Standard of the People's Republic of China (GB 4789.2–2010). Twenty-five g of samples were put in sterile stomacher bags and homogenized for 2 min in 225 mL of 0.85 g/100 mL aseptic physiological saline. A series of decimal dilutions were prepared and spread over agar plates. The experiment was carried out in a clean bench. Colonies on the plates were counted after incubation for 48 ± 2 h at 36 ± 1 °C. The data were recorded as colony-forming units (CFU) and expressed as $\log_{10}$ CFU/g.

2.2.4. Malondialdehyde (MDA) content

MDA content is considered as an indicator to assess the degree of plant oxidative stress. Five g of samples were homogenized with 50 mL of 10 g/100 mL trichloroacetic acid and centrifuged (2-16 PK, Sigma Laboratory Centrifuges, Germany) for 10 min at 1700 g. Four mL of the supernatant was mixed with equal volumes of 0.6 g/100 mL 2-thiobarbituric acid (dissolved in 10 g/100 mL trichloroacetic acid). The solution was put into boiling water bath for 15 min and then centrifuged for 10 min at 1700 g after rapidly cooling down. The absorbance was measured at 450 nm, 532 nm and 600 nm (Hodges, DeLong, Forney, & Prange, 1999). MDA content was calculated using Eq. (2).

$$\text{MDA content (nmol/g)} = (6.45 \times (A_{532}-A_{600})- 0.56 \times A_{450}) \times V_1 \times V_3/(V_2 \times W) \quad (2)$$

Where A_{450}, A_{532} and A_{600} are the absorbance values at 450 nm, 532 nm and 600 nm, respectively. V_1 is the total volume of the solution obtained after reaction (mL), V_2 is the volume of the extract solution using for reaction (mL), V_3 is the total volume of the extract solution (mL), W is the mass of the sample (g).

2.2.5. Membrane permeability

Membrane permeability, an indicator of quantification of plant cellular disruption, is also correlated to the quality of vegetables and fruits during storage (Gonzalez, Anthon, & Barrett, 2010). The samples were cut into 5 mm × 5 mm pieces and 10 g of samples were rinsed with deionized water. After dehydrating the surface water with filter paper, the sample were put into breaker containing 50 mL of deionized water and kept in 30 °C water bath for 1 h. Then the electric conductivity value (A_0) of deionized water was measured using a conductivity meter (DS-11AT, Shanghai Precision Science Instrument Co., Ltd, China). The electric conductivity value (A_1) was determined again after heating and boiling the water with samples for 15 min. Tests were carried out in three repeats. Membrane permeability was calculated using Eq. (3) (Meng, Zhang, & Adhikari, 2012).

$$\text{Membrane permeability (\%)} = (A_0/A_1) \times 100\% \quad (3)$$