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Physicochemical changes of 'Phulae' pineapple fruit treated with short-term anoxia during ambient storage

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ABSTRACT

The effects of short-term anoxia exposure for 16 h on physicochemical changes of 'Phulae' pineapple fruit stored at ambient temperature (25 ± 2 °C) were investigated. The respiratory rate of the fruit was induced by the anoxia treatment. However, it retarded the increase in moisture loss and maintained both flesh and pulp colour by inhibiting polyphenol oxidase (PPO) activity of the both tissues. The anoxia exposure delayed the increase in total sugar content and enhanced total ascorbic acid content during storage. The half-cut pineapple fruit showed that the anoxia exposure completely inhibited internal transparency of the flesh tissue adjacent to core during the storage. In conclusion, the short-term anoxia exposure for 16 h maintained postharvest quality, retarded physiological disorder and enhanced nutritional values of the pineapple fruit stored at ambient temperature (25 ± 2 °C).

1. Introduction

Thailand is the tropical country which is contributing to a wide range of tropical fruit. One of the most major commercial fruit of Thailand is pineapple which has been grown in most part of Thailand. 'Phulae' pineapple (*Ananas comosus*), a Queen pineapple variety, is the important Geographical Indications (GI) of Chiang Rai Province, a northern part of Thailand. As its sweet and aromatic taste and crispy texture of both flesh and core, 'Phulae' pineapple fruit has become an important commercial fruit and its demand has been increased in both domestic market and export. Unfortunately, the loss of pineapple fruit after harvesting is still the main problem. The estimation of postharvest loss of perishable commodities including pineapple fruit would up to 50% which caused by a combination of several factors (Paull & Chen, 2003). Generally, refrigerated storage is recommended for storage perishable commodities; however, chilling injury is the most problem of tropical fruits during storage. 'Queen' pineapple fruit is recognized as being more chilling-sensitive than 'Smooth Cayenne' fruit which chilling injury symptom of 'Queen' pineapple fruit was detected within 5 days after storage at 8 °C (Youryon, Wongs-Aree, McGlasson, Glahan, & Kanlayanarat, 2013). Moreover, Hong et al. (2013) reported that the incidence of internal browning and PPO activity of pineapple fruit held at 25 °C was more intensive than the fruit held at lower temperature. In Thailand, pineapple fruit are generally stored at ambient temperature in order to prevent internal browning caused by refrigerated storage.

The development of effective methods to maintain the qualities has been widely reported including heat treatment (Wijeratnam, Hewajulige, & Abeyratne, 2005) and calcium treatment (Silva, Fernandes, & Mauro, 2014). Pre-treatment fruit and vegetables to short-term anoxia conditioning after harvest has many more effective to reduce respiration rate, to inhibit ethylene production and its action, to delay ripening and to reduce the incidence of some physiological and biochemical changes (Fallik, Alkalai-Tuvia, Shalom, Larkov, & Ravid, 2005; Kelly & Saltveit, 1988; Techavuthiporn & Boonyaritthongchai, 2016). This method involves removing O_2 using pure N_2 in short period prior to storage. Below a threshold of O_2, cells undergo anaerobic respiration and produce some anaerobic substances (Peppelenbos & Oosterhaven, 1998). The extent of anaerobic activity depends on the amount of time cells are exposed to air.

However, there is no report yet on the application of short anoxia treatment for controlling physiological quality changes of 'Phulae' pineapple. The aim of this research was to determine the possible application of short-term anoxia as a pre-treatment after harvest for 'Phulae' pineapple fruit by evaluating its effects on the changes in visual and physicochemical quality.

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2. Materials and methods

2.1. Fruit preparation

Pineapple fruit (*Ananas comosus*cv. Phulae) was harvested at the commercial maturity stage (5 months after forcing) and were then immediately delivered from a commercial orchard in Chiang Rai Province, Thailand, to the Postharvest laboratory of King Mongkut's University of Thonburi, Bangkok within 12 h. The peduncle of the fruit was cut with a sharp knife to leave 2 cm peduncle on the fruit. The crown was trimmed to a length of 3–4 cm. No damaged samples with a same colour (stage 2 of ripening; less than 1/4 gold of the fruit peel) and an average weight of 350 ± 10 g per fruit were selected. The fruit were washed by rinsing with tapped water twice and then air dried with ambient air at 25 °C.

2.2. Short anoxic stress treatment

Preliminary investigations showed that, at a time range of 0–24 h, exposure to pure N_2 for 16 h was most effective for delaying the colour changes of pineapple fruit peel held at 25 °C (data not shown). In this study, therefore, 'Phulae' pineapple fruit were divided into 2 set, 36 fruit per set. The first set was directly placed in air-tight plastic chamber (40 L), which was then flushed with humidified pure nitrogen (90–95% Relative Humidity; RH) for 16 h in darkness at room temperature (25 ± 2 °C). During treatment, It was necessary to obtain an anoxia condition (oxygen concentration in the chamber less than 0.5 kPa measured using a handheld Gas Analyser, model: Oxybaby M + X, Germany). In addition, the second set (untreated or control) was flushed with humidified air at the same temperature for 16 h. Consequently, both anoxia and air treatments were established. Six anoxia treated or untreated fruit were placed in a polypropylene baskets ($30 \times 40 \times 12$ cm^3) and then covered with perforated plastic sheet with 60 μm low density polyethylene (10 holes with 1.5 cm in diameter) to prevent the fruit moisture losses. All samples were stored at 25 ± 2 °C (85–95% RH) for 8 days. All analyses were performed at 0, 2, 4, 6 and 8 days of storage. Each treatment was applied to four replicates in randomly (2 fruit per replicate).

2.3. Fruit weight loss

The loss of fruit weight during storage was measured periodically by weighing each fruit on a digital balance (model PA2102, OHAUS Corporation, USA). The results are reported as the percentage of fruit weight loss based on the initial weight on day 0 of the same fruit.

2.4. Respiration rate

The respiration rate of the fruit was measured using a closed system method at 25 ± 2 °C according to measurement of Techavuthiporn, Nakano, and Maezawa (2008). At each sampling time, fruit was weighed and transferred to a gas-tight plastic chamber (2.5 L). The evolution of CO_2 concentration in the chamber was measured during time intervals. One mL of gas sample was withdrawn with a gas-tight syringe and then injected into gas chromatograph (Model GC-8A, Shimadzu, Japan) for measuring CO_2. The injected gas sample was separated by a WG-100 column and analyzed with a thermal conductivity detector (TCD). Helium was used as the carrier gas. The respiration rate of the fruit was calculated from an evolution CO_2 concentration with time divided by sample weight to obtain respiration in mg kg^{-1} h^{-1}.After the respiratory measurement, the chambers were opened and samples were returned to the storage basket and stored until further measurement.

2.5. Colour determination

Colour was determined using a colorimeter (Model CR-400 Chroma Meter; Konica Minolta Sensing Inc., Osaka, Japan), based on the L^*, a^*, and b^* values of the CIE (International Commission on Illumination) scale. From these values, hue angle ($h°$) value was calculated as follows.$h° = \tan^{-1}(b^*/a^*)$.

Three portions of measurement were done on the fruit including peel, flesh and core. For either control or treated N_2 fruit, four fruit were randomly measured and on each portion, four readings were made.

2.6. Extraction and assay of polyphenol oxidase activity

The activity of polyphenol oxidase (PPO) in both flesh and core pineapple fruit tissues was assayed using the method of Luh and Phithakpol (1972). Acetone powder of the fruit samples was prepared by adding 95% acetone (held at −50 °C for 24 h) into 50 g of the fruit sample. The sample was immediately homogenized for 3 min and then vacuum-filtered using Whatman No. 1 filter paper. During filtration, the cold acetone was rinsed through the sample until colourless. The filtrate was again homogenized with 50 mL of the cold acetone and then filtered through Whatman No. 2. The acetone powder was dried at ambient temperature. A 200 mg of acetone powder were extracted with citric acid phosphate buffer pH 6.2 and then stirred at 4 °C for 30 min. The sample was filtered through Whatman No. 42 filter paper. The filtrated was collected. Ten mL of filtrate was mixed with 5 mL of 0.1 M catechol for 30 s. Absorbance at 420 nm was measured after 3 min of the reaction using a spectrophotometer (UV-1600; Shimadzu Co., Japan). The PPO activity unit was expressed as unit g^{-1} fresh weight.

2.7. Sugar and ascorbic acid contents

Sugar content was measured according to a slight modification of the method of Dubois, Gilles, Hamilton, Rebers, and Smith (1956). One gram of sample was homogenized with 10 mL of 80% ethanol and incubated in a water bath at 60 °C for 1 h. One mL of the extract was mixed with 1 mL of 5% phenol and 5 mL of concentrated sulfuric acid. The absorbance of mixed solution was measured at 490 nm with a spectrophotometer. The concentration of total sugars content was determined using a standard curve of glucose and expressed as mg kg^{-1} of sugar on a fresh weight basis.

AA content was analysed by the method of Roe, Mills, Oesterling, and Damron (1948), the 2,4-dinitrophenol hydrazine method, which includes an extraction with 5% metaphosphoric acid solution (Merck KGaA, Germany). The absorbance of the sample solution was measured with a spectrophotometer at 540 nm. The calibration curve was obtained using standard AA solutions (Carlo ErbaReagentiSpA., Italy) and values for total ascorbic acid content were expressed as mg kg^{-1} on a fresh weight basis.

2.8. Statistical analysis

The experimental data were the average of four replications from four different fruit. The experiments were performed using a completely randomized design. All results herein are presented as the mean ± S.E. Statistical significance was assessed using an ANOVA at the 99% confidence level. Statistical analyses were performed by SAS Institute Inc. (1999).