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Profiles of carotenoids during post-climacteric ripening of some important cultivars of banana and development of a dry product from a high carotenoid yielding variety

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ABSTRACT

Since vitamin A deficiency is prevalent in many developing countries, we sought to focus research on local, affordable and well-accepted sources of provitamin A carotenoids. As dessert bananas are consumed fresh round the year and processed as products, this study investigated whether post-climacteric biochemical changes are linked to carotenoid degradation in four Indian varieties, one commercial (Cavendish, AAA), one Red banana (genome AAA) and two locally-important ones (genome AAB). Despite large differences in their ripening characteristics, textural loss was lesser in AABs than AAAs. High levels of carotenoids (21.0 μg/g FW), β-carotene (9.14 μg/g FW) and α-carotene (9.32 μg/g FW) were found in Red banana accounting for retinol activity equivalent of 114 μg/100 g FW. The carotenoid levels were lower in two local varieties and lowest in Cavendish, with no post-climacteric loss. Dry grits, prepared using Red banana pulp, milk powder and sugar, retained about 70% carotenoids and appeared useful in confectionaries.

1. Introduction

For humans vitamin A is required for normal vision and many other physiological functions such as gene expression, reproduction, embryonic development, enhancement of mineral bioavailability, disease prevention and to modulate immune functions (Chew & Park, 2004; Nishiyama, Sugimoto, Ikeda, & Kume, 2010). An estimated 250,000 to 500,000 vitamin A-deficient children globally are prone to blindness, with half of them losing life within 12 months every year (Micronutrient Initiative, 2009). India accounts for 25% of the 15 million blind facing grave health problems due to vitamin A deficiency and age-related macular degeneration (Micronutrient Initiative, 2009). Developing countries are also challenged with vitamin A deficiency-related mortality and morbidity of neonates, where improved immunity was observed in neonates upon dietary supplementation of vitamin A and β-carotene (Nishiyama et al., 2010).

Leafy vegetables and yellow fruits/vegetables are generally recommended as rich sources of β-carotene. However, leafy vegetables are not liked by many for routine consumption, and yellow fruits are either seasonal or inaccessible and unaffordable for people in developing countries, for various reasons. Carotenoids-rich fruits like papaya are traditionally denigrated, and hence not widely accepted. Banana is one of the most important food crops with a wide range of varieties - cooking and dessert types, providing a staple and nutritious inexpensive food, which is recently gaining importance to address vitamin A deficiency in developing countries (Davey, Van den Bergh, Markham, Swennen, & Keulemans, 2009; Englberger, Darnton-Hill, Coyne, Fitzgerald, & Marks, 2003; Englberger et al., 2006, 2010). India is a major producer of banana accounting for 21% of the world production, and one of the major centers of origin for banana cultivars (Venkatachalam, Sreedhar, & Bhagyalakshmi, 2008). Many local bananas of India have never been characterized for their nutritional compositions. Certain local Indian cultivars of banana, despite their high prices, are more relished by local people than commercial varieties such as Cavendish. Banana is also a good source of carotenoids, particularly *trans*-β-carotene, the most important one among provitamin A carotenoids (pVACs) (Englberger et al., 2003). One of the local Australasian

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Abbreviations

pVACs	provitamin A carotenoids
FW	fresh weight
DW	dry weight
L*	lightness
a*	red/green value
b*	blue/yellow value
C*	chroma
h*	hue angle
ΔE	color deviation value
DMRT	Duncan's Multiple Range Test
RAE	retinol activity equivalent

cultivars, *Utin Iap*, was found containing about 8500 μg of β-carotene/100 g FW (Englberger et al., 2006), accounting for 400 times higher β-carotene than that present in commercially reputed *cv.* Cavendish. A shift towards high carotenoid banana cultivar would lead to a natural, cheaply available source of vitamin A in a highly bioaccessible form, unlike green leafy vegetables that are not so much liked by many and are rich in phytate and fiber that are known to interfere with the bioavailability of vitamins and micronutrients (Amagloh et al., 2012). Since bananas are highly perishable in tropical climate, the present study analyzed total carotenoids and pVACs to find out whether these compounds are also lost during the post-climacteric textural degradation. Another reason being that, in India, bananas are sold mainly in open markets where semi-perished bananas are cheap, often bought by poor people who prefer highly sweet over-ripe ones. An earlier study reported carotenoids in different cultivars of banana, where highest content of carotenoids (4 μg/g DW) and β-carotene (1.17 μg/g FW) were found in the pulp of Red banana (Arora et al., 2008). Therefore, for addressing the vitamin A deficiency problem, the present study analyzed post-climacteric textural degradation and other important biochemical changes in three local cultivars, Elakkibale, Nanjanagudu rasabale, Chandran- a Red banana, and a commercial variety namely Cavendish. Nature has provided banana fruits hygienically packaged with eco-friendly peels, with the delicious seedless pulp, which is amenable for easy processing into a variety of products (Aurore, Parfait, & Fahrasmane, 2009). Carotenoid-rich banana can be processed into a product/ingredient having wide applications in confectionaries targeted for children. Since high content of total carotenoids (including β-carotene and α-carotene) was found in Red banana, the pulp of this variety was used to prepare grits adding whole and skimmed milk powder and the stability of carotenoids was analyzed.

2. Materials and methods

2.1. Samples

Fruits of two varieties each of *Musa paradisiaca* (*cv.* Nanjanagudu rasabale, Elakkibale; genome AAB) and *Musa acuminata* (Cavendish, Red banana var. *Chandran*; genome AAA) were purchased from a local market, and treated with ethylene gas in smoke chambers. Individual hands were kept in paper pouches and respiratory climacteric stage was identified for each cultivar by checking CO_2 emission peak as explained previously (Manjunatha, Lokesh, & Bhagyalakshmi, 2012), which was on 3rd day for Cavendish and Red banana and on 4th day for Nanjanagudu rasabale and Elakkibale.

2.2. Chemicals

HPLC standards β-carotene and α-carotene were from Sigma (Sigma–Aldrich, St. Louis, MO, USA). The α-carotene standard was kindly shared by Dr. V. Baskaran (Biochemistry & Nutrition Department, CFTRI, Mysore, India). Reagents used for extraction and separation of carotenoids were of HPLC grade and all other chemicals were of analytical grade procured from Merck (Darmstadt, Germany). For HPLC and MS analyses ultrapure water (Merck Millipore, India) was used and double distilled water was used for other biochemical analyses.

2.3. Analyses of physical parameters

Fruit surface color was measured using the HunterLab color measuring system Minolta CR–200 (Hunter Associates Laboratory Inc., VA, USA) at visible wavelength lights, following the procedure of McGuire (1992). Color coordinate L indicates lightness, whereas a* (− to +) and b* (− to +) indicate the change in the hue from green to red and blue to yellow respectively. L*, C* and h* values were calculated from the L*, a* and b* values. The C* value (Chroma) and the h* value (Hue angle) were calculated as $(a^{*2} + b^{*2})^{0.5}$ and $\tan^{-1}$ (b*/a*) respectively. Firmness was measured using the universal texture measuring system (UTM Lloyds, LR–5K, Lloyd Instruments Ltd., Fareham, U.K.). A cylindrical penetration probe with a diameter of 2 mm was made to penetrate the fruit at constant speed to a depth of 15 mm with load cell of 5 kN. The maximum force applied to penetrate the fruit through the peel represented the hardness, which was expressed as Newtons (N) as described by Breene (1975).

2.4. Moisture measurement

The moisture content was estimated by measuring the fresh weight and dry weight of pulp and peel separately. While fresh weights were recorded immediately after peeling, for dry weights, samples were dried at 80 °C until a constant weight for each sample was achieved. The difference between fresh and dry weights was recorded as the moisture content for each set of samples.

2.5. Analyses of biochemical parameters

Pulp (1 g) from each fruit sample was macerated in 10 ml of 0.2 mol/L phosphate buffer (pH 7.0), filtered, centrifuged and the supernatant was used to estimate total soluble protein by the method of Lowry, Rosebrough, Farr, and Randall (1951), comparing the values with the standard curve obtained with bovine serum albumin. For the estimation of total carbohydrates, a known quantity of banana pulp as well as peel from each ripening stage was separately macerated in absolute ethanol, soaked overnight in excess solvent, solubles and insolubles were separated by filtration, repeating the extraction thrice and pooling respective fractions. Total carbohydrate in soluble fraction was estimated by phenol sulphuric acid method (Dubois, Gilles, Hamilton, Rebers, & Smith, 1956) using standard curve obtained by similar estimation of glucose standard. Total reducing sugars were estimated by dinitrosalicylic acid method (Miller, 1959).

All carotenoid analyses were done in subdued light. Total carotenoids in all the four banana cultivars were extracted from fruit pulp on climacteric (day 1) and on fourth day evening, because the subsequent ripening stages in Red banana and Cavendish showed much softening, appearing unfit for analyses. Extraction and HPLC analysis of carotenoids from banana pulp was done according to Rodriguez-Amaya and Kimura (2004). Fruit pulp (5 g) was ground using mortar and pestle using ice-cold acetone. The grinding was