

## Estimation of $\beta$ -Carotene in Local Banana Cultivars from Kerala

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Bananas, a major staple food are the forth important food in the world. Bananas are nourishing fruits contains many minerals and vitamins. It is a rich source of energy low in protein and fat content and provides a balanced diet. At present a number of edible banana cultivars are spread throughout the world. India has more than 300 cultivars. Edible cultivars are derived from interspecific hybridization between two wild species *Musa accuminata* and *Musa balbisiana*. In Kerala cultivars like Palayamcodan (AAB), Red banana (AAA), Robusta (AAA), Nendran (AAB), Rasakadali (AB) and Chingan (AA) are available. In present study, biochemical analysis of 13 cultivars which belongs to AA, AB, AAA, AAB genomes, respectively was conducted. The analysis involved estimation of carbohydrates, starch, reducing sugars, proteins as well as  $\beta$  carotene. The cultivars which are studied are Kadali, Matty (AA), Annanvazha, Rasakadali (AB), Red banana, Robusta (AAA), Palayamcodan, Nendran, Poovan, Padatti (AAB). Among the cultivars Nendran has high carbohydrates, starch, reducing sugars as well as  $\beta$  carotene content and padatti has maximum proteins.  $\beta$  carotene content was quantified at differential stages of fruit development using HPLC.

**Key Words:** Banana, Musaceae, Scitaminae, *Musa accuminata*, *Musa balbisiana*,  $\beta$ -carotene, HPLC

### Introduction

Vitamin A deficiency is considered as a global health problem (UNICEF, 1990). Food sources of Vitamin A include animal foods rich in Vitamin A and plant foods containing provitamin A carotenoids such as  $\beta$  carotene (Mc Laren, 2001). Carotenoids are widely distributed in the plant kingdom. These components contribute to the red, orange and yellow colours to fruits (Burns, 2003; Hughes, 2001). A number of carotenoids including  $\beta$  carotene,  $\beta$  carotene and  $\beta$  cryptoxanthin have provitamin A activity and they are converted to retinal by mammals (Burns, 2003) have twice provitamin A activity. There are several factors that restricts the potential of known food sources of Vitamin A. Animal foods as source of vitamin A are often too expensive for low income people, unacceptable for religious reasons or less available (Gibson, 2000). In many regions of world dark green leafy vegetable may not be given to young children, because of concern that it may not be digestible and Papaya is avoided in Southern India by pregnant woman because of a belief of miscarriage (Kuhnlein, 1997). Some people in India consider it as a food causes dysmenorrhea in women and impotency in men. Other vegetables like mangoes, pumpkins, squash, orange fleshed sweet potatoes have the problem of indigestion, taste and season dependent availability (Englberger, 2003).

Banana is a major food in the world, after rice, wheat and maize. These fruits are wholesome and fairly

well balanced source of nutrients containing various mineral salts, vitamins and high amount of carbohydrate with a little fat and protein (Simmonds, 1966; Ketiku, 1973; Ahenkaro *et al.*, 1997). These fruits are eaten raw as desert fruits or cooked as vegetable and available throughout the year. These are rich in antioxidant vitamins (Vitamin C, A and E), calcium, magnesium and potassium (Marisa, 2006). Bananas are not listed as a fruit rich in  $\beta$  Carotene since the Cavendish varieties available in US and Europe has little  $\beta$  Carotene content. It is observed that some banana cultivars from Micronesia and South East Asia have significant amount of  $\beta$  carotene, a precursor of Vitamin A (Abdon *et al.*, 1980; Puwastein *et al.*, 1999; Siong, 1985). Some cultivars of Micronesia like Karat and Uhten Yap have significant carotenoid contents (Stover, 1987) various cultivars from Africa also have high content of carotene (Daniells, 2001). However the nutritional analysis of many cultivars available in Kerala is not evaluated and present study reports the biochemical analysis of ten banana fruits locally available in Kerala.

### Materials and Methods

Ten cultivars of banana fruits were purchased from the banana nursery, Peringmala, Kerala, South India and used in the present study. This includes Kadali, Matty (AA), Annanvazha, Rasakadali (AB), Red banana, Robusta (AAA), Palayamcodan, Nendran, Poovan, Padatti (AAB). The analyses of organic compounds, minerals as well as  $\beta$ -carotene were estimated from

ripened fruits. The  $\beta$ -carotene content at un-ripened, ripened and over-ripened fruits were estimated using HPLC only in the cv. Nendran.

#### **Estimation of Carbohydrates**

Carbohydrate estimation of the banana fruit was done as per the protocol defined by Hedge and Hofreiter (1962). Glucose was used as standard. UV spectrometer (Shimadzu 1700) was used for the estimation of carbohydrate present in the sample.

#### **Estimation of Starch**

The estimation of total starch content was done using the modified protocol by Thayumanavan and Sadasivam (1984). Glucose was used as standard. UV spectrometer (Shimadzu 1700) was used for the estimation of starch present in the sample

#### **Estimation of Reducing Sugar**

The reducing sugar was estimated by using Di nitro Salicylic acid with reference to the procedure Miller (1971). Amount of reducing sugar was calculated with the help of a standard graph using glucose. UV spectrometer (Shimadzu 1700) was used for the estimation of carbohydrate present in the sample

#### **Estimation of Total Proteins**

The estimation of total protein was done with the help of procedure by Lowry *et al.* (1951). The amount of protein present was calculated with the help of standard graph using BSA. UV spectrometer (Shimadzu 1700) was used for the estimation of protein present in the sample

#### **Determination of Minerals**

The mineral content of different banana samples was determined by procedure mentioned by Health Protection Branch Laboratories Bureau of Nutritional Science. Determinations of elements were done by the help of Flame Atomic absorption Spectrophotometer.

#### **Spectrophotometric estimation of $\beta$ -carotene**

The extract was prepared according to the protocol. The carotene were extracted with ice cold acetone and it was filtered under vacuum. The extracts were then transferred to 100 ml petroleum ether and acetone was removed by washing with water. The colour developed was analyzed at 455 nm using spectrophotometer. Then the amount of  $\beta$ -carotene was estimated by the help of a standard graph. UV spectrometer was used for the estimation of  $\beta$ -carotene present in the sample.

#### **HPLC Analysis of $\beta$ -Carotene**

The HPLC analysis was done with the help of procedure mentioned by Weissenberg *et al.* (1997).

#### **Pigment Extraction**

$\beta$ -carotene was extracted from fruit paste using petroleum ether (bp 40-60°C) repeatedly in subdued light until colorless extracts were obtained. 100 ml of aliquots were evaporated in a stream of Nitrogen.

#### **HPLC Determination**

The extracts obtained were dissolved in mobile phase (1 ml), filtered through a 0.45 mm membrane disc and injected in to chromatograph. The column was regenerated by washing with 2-Propanol after analysis and then equilibrated with mobile phase.

#### **Standard Solutions**

Stock solution of  $\beta$ -carotene (Hi-Media, Bangalore) was prepared in petroleum ether. Aliquots (10-200  $\mu$ l) were evaporated in a nitrogen stream and the residues were dissolved in mobile phase (1 ml) and subjected to HPLC.

#### **Results and Discussion**

Estimation of organic compounds such as carbohydrates, starch, reducing sugar and protein showed that depending upon the cultivar, the nutritional components varied in the ripened fruits. Among different cultivars Padatty (AAB) have highest protein and Matty (AA) has the minimum protein. Nendran (AAB) has the higher content of total carbohydrate, starch as well as reducing sugar. From the analysis of different minerals, most bananas have a significant amount of minerals. Kadali (AA) have a higher content in Manganese and Magnesium but copper content is nil. Copper content is higher in Padatty (AAB), Iron in Rasakadali (AB), Potassium in Matty (AA), Calcium in Red banana (AAA) and Zinc in Robusta (AAA).  $\beta$ -carotene estimation showed that Nendran (AAB) has maximum content and Kadali (AA) has minimum content. The HPLC analysis of  $\beta$ -carotene in different stages of fruit ripening in Nendran showed that unripened fruit has higher  $\beta$ -carotene than ripened and over ripened fruits.

Studies in banana reported that protein content increases only the first 60-70 days but after that it remains constant (Peumans *et al.*, 2000). It is also observed that reducing sugar content is higher than that of starch in most cases. During ripening starch content decreased and the reducing sugar content increased (Henderson,

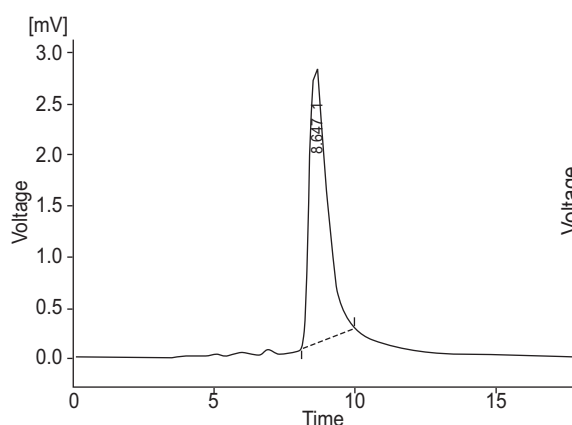


Fig. 1: HPLC chromatogram of  $\beta$  carotene standard

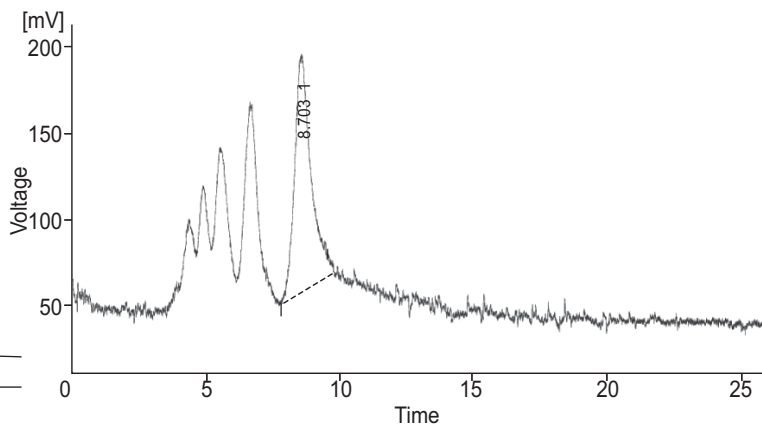


Fig. 2: HPLC chromatogram of pre-ripened Nendran

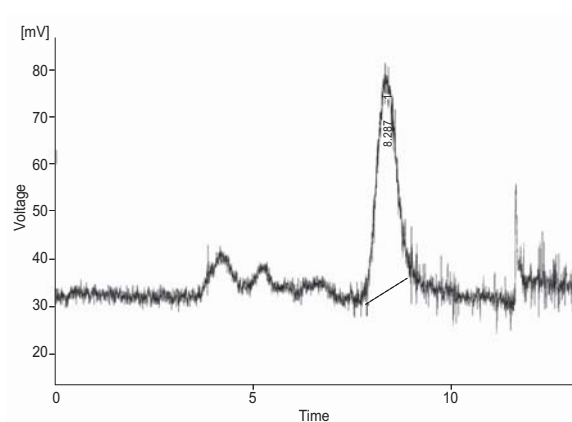


Fig. 3: HPLC chromatogram of ripened Nendran

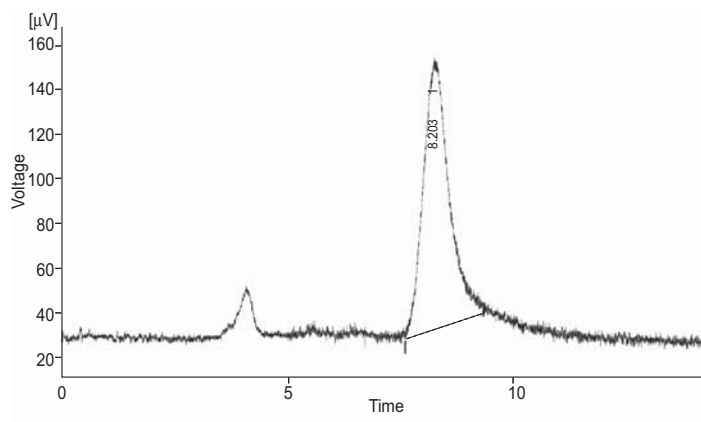


Fig. 4: HPLC chromatogram of over ripened Nendran

1956). Even though Zinc is higher in Robusta, Nendran (AAB) also has significant amount Zinc. This may indicate the high carotene content in Nendran because Zinc is an important component in carotenoids. The HPLC analysis of  $\beta$ -carotene was different from Spectrophotometric analysis. This variation may be due to the difference in procedure during extraction. Spectral analysis used acetone and HPLC analysis used Petroleum ether. In Nendran unripened fruits showed high  $\beta$ -carotene Giami and Alu (1994) also reported that total carotenoids in plantain almost halve during ripening

From this preliminary analysis the nutritional status of locally available banana fruits indicated variation among the cultivars. Also the cultivar Nendran showed potential for improvement in  $\beta$ -carotene content.

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