



EVALUATION OF BEETROOT DURING ITS DEVELOPMENTAL STAGES: A CHEMICAL COMPOSITION STUDY

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ABSTRACT

Presently there is a lack of knowledge that addresses the effect of the development stages of beetroot on its nutrients, anti-nutrients, and pigments. In the present study, the different development stages of beetroot (60, 80 and 100 days) were analyzed for nutrients, anti-nutrients, and pigments by applying the standard method of AOAC. The beetroot showed a significantly higher amount of moisture, calcium, iron, choline, folate, and pigments (betaine and betanin) at 60 days of maturity, and ash, crude fiber, total carbohydrate, phosphorus, magnesium, potassium, and anti-nutrients (oxalic acid and saponin) contents were highest at 100 days of maturity in beetroot. Although vitamin C content was increased from 60 days to 80 days of maturity (8.96 to 9.03), while at 100 days it was decreased. The study shows that all maturity stages produced satisfactory results but the amounts of anti-nutrients (oxalic acid and saponins) were increased during the stages of development.

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INTRODUCTION

Epidemiological studies have shown that consumption of fruits and vegetables have protective effect against many chronic and degenerative diseases such as cancer (Biondo *et al.*, 2014).

According to the world health organization as the population's ages, thus there is increasing importance for health policy and also increasing strategies to consumption of fruits and vegetables as to prevent the large and growing global burden of chronic conditions which impose substantial personal and social costs (Conklin *et al.*, 2014). Beetroot (*Beta Vulgaris*) is a taproot portion of beet plant, belongs to the Chenopodiaceae family (Chawla *et al.*, 2016). In India the optimum time for beetroot cultivation is from June to November. In whole world the main production of beetroot is taken place in North America, Europe, and the USA (Neelwearne, 2013). In recent years beetroot has attracted much attention as a health promoting functional food (Clifford *et al.*, 2015). It is very nutritious vegetable it contains no fat, very few calories, excellent source of calcium, iron, dietary fiber, folic acid, vitamin C and choline (Kumar, 2015). Betanin is the main pigment present in beetroot have 1.5 to 2.0 fold more antioxidant capacity than some other anthocyanins (Gliszczynska *et al.*, 2006). However, betaine is highly biologically active compound present in beetroot and it acts as organic osmolyte, methyl donor for the remethylation of

homocysteine, protector against alcohol-induced liver injury, biologically important in cancer development and it is also promising agent that attenuates homocysteine rise after meals (Rivoira *et al.* 2017). In addition to the health beneficial compounds beetroot also contains significant amount of oxalic acid. It is a strong metal chelator which interferes with calcium and iron metabolism and also leads to the formation of nephroliths (Wruss *et al.*, 2015). Beetroot also contains compound saponins. After consumption which provide bitter taste (Mikolajczyk *et al.*, 2016). Some scientific studies have reported that changes in chemical composition of fruits, vegetables and leaves during its different stages of development. However, presently there are no studies available that address the effect of different development stages of beetroot on nutrients, anti-nutrients and pigments.

So the goal of the present study was to evaluate the nutrients, anti-nutrients and pigments at different development stages (60, 80 and 100 days) in order to evaluate the optimum maturity stage of beetroot for food. Presently in literature the study is available on maturity stages of beetroot leaves (Biondo *et al.*, 2014), but the study on maturity stages of beetroots are unprecedented.

MATERIALS AND METHODS

Sample Collection

Beetroot were organically produced in Ghala farm, Lucknow, Uttar Pradesh. They were harvested at 60, 80 and 100 days after sowing. However, commonly the harvesting is done after 80 days of sowing. After sowing the harvest was done at 60,

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80 and 100 days between the months of July to November. Then different maturation stages of beetroot were analyzed for nutrients, anti-nutrients and pigments with standard analysis methods at Regional Food Research and Analysis center (r-frack) at Lucknow.

Preparation of Sample

The beetroot which was picked up at three stages of maturity i.e. 60, 80 and 100 days were washed under tap water then peeled and sliced them. After that roots were wiped and sliced into small pieces.

Nutrients

Moisture, ash, crude fiber and total carbohydrates contents were determined in accordance with AOAC (Association of official analytical chemistry), 2000 method (AOAC, 2000). However, for mineral determination, the ground sample was added in 25-30 ml of diacid mixture (HNO₃: 5:1, v/v) and kept them for overnight. Then the contents were digested by applying heat. After that these crystals were used for estimation of total minerals (calcium, magnesium and iron). However, the phosphorus content was determined by applying calorimetrically method. The potassium content was estimated by adding con. Nitric acid-conc. Perchloric acid-conc. Sulphuric acid in the ratio of 3:2:1. Choline, vitamin C and folate were determined by using AOAC (2000) method.

Anti-Nutrients

The saponins contents were determined by applying a modified method of Gestetner *et al.* (1966). However, oxalic acid was estimated by standard method of analysis AOAC (1980). Oxalic acid content was calculated in mg/100g by using Equation 1.

$$1350 = 0.45(\text{mg anhyd. Oxalic acid equivalent to } 1\text{ml } 0.01 \text{ N KM11O4}) \times (30/20 \times 500/25) (\text{diluted factor}) \times 100 (\text{to convert to } 100 \text{ g Product}).$$

Pigments

In pigments the contents of betanin in beetroots were purified by using column chromatography methods. However, the analysis of purified betanin was done by applying spectrophotometer method. Lastly the spectra were deconvoluted by using Fytik analysis software (Goncalves *et al.*, 2012). The content of betaine was estimated by using HPLC method. In which the peak area of the absorbance was plotted on 0.05-4m M of concentration. After that measuring the absorbance peak area of extract at the unknown concentration of betaine was calculated according to Equation 2, as follows (Bessieres *et al.*, 1999).

Areaextract - peak area (absorbance at 200 nm) determined with the extract.

Slope (expressed as absorbance-nmol-1) is determined by linear regression

Vol_{extract} - The total volume of the extract

Vol_{aliquot} - The volume of extract injected on to the HPLC column

Abs_{extract} - within the linear range

RESULT

Nutrients

Table 1 presents the nutrients composition of beetroot at different stages of maturity (60, 80 and 100 days). Among all the developmental stages of beetroot the moisture and ash contents were significantly (p<0.05) decreasing with advancement of maturity stages. The crude fiber content was highest at 100 days (3.50±0.26 g/100g) and lowest at 60 days (0.88±0.01 g/100g). So it was found that the content of crude fiber was significantly increased with stages of maturity. The total carbohydrate content of beetroot ranged from 10.93±0.5 g/100 g (100 days); thus it can be observed that the content of carbohydrate was significantly (p<0.05) increased from 60 days (stage-I) to 100 days (stage-III) of maturity of beetroot. However, in minerals contents the content of calcium and iron were ranged from 114.55±0.06 mg/100g (60 days) to 110.00±0.00 mg/100g (100 days) and 6.07±0.02 mg/100g (60 days) to 4.01±0.01 mg/100g (100 days). So it was found that the content of calcium and iron were significantly decreased from stage-I (60 days) to stage-III (100 days). However, the content of magnesium, phosphorus and potassium were significantly increased with advancement of maturity stages from stage-I (60 days) to stage-III (100 days) of maturity. Among all the stages of maturity of beetroot the contents of folate and choline were highest at 60 days of maturity (80.37±0.55 and 6.00±0.0 mg/100g) and the lowest values were found at 100 days of maturity i.e. 64.50±0.44 mg/100 g and 3.57±0.40 mg/100 g. But in the case of vitamin C the content of vitamin C were firstly increased from 60 days to 80 days but after that at 100 days it was decreased.

Results were presented in mean ± standard deviation. The analyses of all nutrients were performed in triplicate.

Anti-nutrients

The presence of oxalic acid and saponins in beetroot at different stages of maturity (60, 80 and 100 days) were investigated in Table 2. The content of oxalic acid of beetroot at 60, 80 and 100 days of maturity was 29.16±0.14, 33.33±1.15 and 36.67±0.55 mg/100g respectively.

Table 1 Composition of Nutrients in beetroot at different stages of maturity

| Development Stages (days) | Moisture (g/100g) | Ash (g/100g) | Crude fiber (g/100g) | Total Carbohydrates (g/100g) | Ca (g/100g) | Mg (g/100g) | Fe (g/100g) | P (g/100g) | K (g/100g) | Choline (mg/100g) | Vit C (mg/100g) | Folate (mg/100g) |
|---------------------------|-------------------|--------------|----------------------|------------------------------|-------------|-------------|-------------|------------|--------------|-------------------|-----------------|------------------|
| 60 days | 87±0.02 | 0.77±0.01 | 0.88±0.01 | 10.93±0.05 | 114.53±0.06 | 219.00±1.73 | 6.07±0.02 | 38.83±0.15 | 205.33±0.58 | 6.00±0.00 | 8.96±0.02 | 80.37±0.55 |
| 80 days | 85.07±0.12 | 0.99±0.01 | 1.09±0.01 | 13.27±0.21 | 112.00±0.01 | 231.00±1.00 | 6.01±0.02 | 41.06±0.02 | 211.00±1.00 | 5.96±0.06 | 9.03±0.02 | 70.37±0.32 |
| 100 days | 65.50±0.10 | 1.17±0.15 | 3.50±0.26 | 16.23±0.15 | 110.00±0.0 | 266.67±5.77 | 4.01±0.01 | 44.23±0.21 | 246.67±15.28 | 3.57±0.40 | 8.03±0.02 | 64.50±0.44 |
| P-Value | <0.001 | <0.001 | <0.001 | <0.001 | <0.001 | <0.001 | <0.001 | <0.001 | <0.001 | <0.001 | <0.001 | <0.001 |

So according to the present study the amount of oxalic acid was significantly ($p < 0.05$) increased with advancement of maturity stages of beetroot. Similarly the content of saponins were also increased significantly with increasing the development stages of beetroot from 60 to 100 days of maturity i.e. it was ranged from 8.40 ± 0.53 mg/100g (60 days) to 15.67 ± 0.58 mg/100g (100 days).

Table 2 Composition of Anti- nutrients in beetroot at different stages of maturity

| Development Stages (days) | Oxalic Acid (mg/100g) | Saponins (mg/100g) |
|---------------------------|-----------------------|--------------------|
| 60 days | 29.16 ± 0.14 | 8.40 ± 0.53 |
| 80 days | 33.33 ± 1.15 | 11.67 ± 0.58 |
| 100 days | 36.67 ± 0.58 | 15.67 ± 0.58 |
| P-Value | <0.001 | <0.001 |

Results were presented in mean \pm standard deviation. The analysis of all Anti-nutrients were performed in triplicate

Pigments

Pigments betaine and betanin have been presented in Table 3. The contents of betaine ranged from 12.67 ± 0.53 mg/100g (60 days) to 110.00 ± 0.00 mg/100g (100 days). Among all the stages of maturity the content of pigments (betaine and betanin) were significantly decreased from increased the advancement of maturity stages.

Table 3 Composition of Pigments in beetroot at different stages of maturity

| Development Stages (days) | Betaine (mg/100g) | Betanin (mg/100g) |
|---------------------------|-------------------|-------------------|
| 60 days | 127.67 ± 0.58 | 181.33 ± 1.15 |
| 80 days | 124.67 ± 0.58 | 150.67 ± 0.58 |
| 100 days | 110.00 ± 0.00 | 110.33 ± 0.58 |
| P-Value | <0.001 | <0.001 |

Results were presented in mean \pm standard deviation. The analyses of all pigments were performed in triplicate.

DISCUSSION

In present study the content of moisture was decreased with increasing the maturity stages. Similar finding has been reported by Gil *et al.* (2013) in swiss chard that the moisture content was decreased with advancement of maturity stages. They reported that the moisture content was decreased due to changes in tissue water holding capacity of swiss chard during development stages (Gil *et al.*, 2012). In previous study on beetroot leaves it was found that ash content was decreased with advancement of maturity stages but in present study the content of ash was highest at 60 days and lowest at 100 days of maturity stage. The present finding of crude fiber are agreement with that of Kumari (2016) who studied the effect of maturity stages on crude fiber content of Okra vegetable. She reported that the crude fiber was significantly increased with advancement of maturity stages in Okra vegetable (Kumari, 2016). The present study agreed with Seleim *et al.* (2015) who reported that the total carbohydrate contents of Zucchini vegetable were increased during its maturity stages (Seleim *et al.*, 2015). Wobeto *et al.* (2006) reported the similar decreasing trends of calcium and iron, but the contents of magnesium and phosphorus were increased with advancement of maturity stages in cassava leaf (Wobeto *et al.*, 2006). However, similar findings has been found in beetroot leaves that the potassium content was lowest at 60 days and highest at 100 days of maturity (Biondo *et al.*, 2013). According to

Gopalan. C *et al.* (2011) almost similar amount of choline was found in carrot it was 168 mg/100g (Gopalan *et al.*, 2012). In present study the content of vitamin C was firstly increased significantly with advancement of maturity stages after that it was significantly decreased. The similar finding has been also reported in Okra vegetable (Kumari, 2016). Jesus *et al.* (2009) reported that the content of folic acid was decreased in tomatoes with advancement of maturity stages (Jesus *et al.*, 2009). The present finding is agreement with Fridig and Goldman (2011) who found that oxalic acid content was increased with advancement of maturity stages in table beet and its leaf also (Freiding *et al.*, 2011). Musa *et al.* (2011) reported that the reason for increasing the amount of anti-nutrients with advancement of maturity stages in plant tissues are that it may be that the secondary plant substances or secondary metabolites are accumulated in plant tissues and organs during its aging (Musa *et al.*, 2011). In present study pigment content were decreased with advancement of maturity stages. Almost similar finding have been reported in okra vegetables. According to Kumari (2016) the anthocyanin content was decreased with advancement of maturity (Kumari, 2016). The present study shows that beetroots are the excellent sources of many valuable nutrients and bioactive physiochemical and antioxidants such as betanin and betaine, in addition to having significant amount of anti-nutrients such as oxalic acid and saponins. The chemical composition in the beetroots were changed during its development stages and the greatest amount of moisture, calcium, iron, choline, folate and pigment (betaine and betanin) were in the 60 days of maturity of beetroot. Ash, crude fiber, total carbohydrates, phosphorus, magnesium, potassium and anti-nutrients (oxalic acid and saponins) contents were highest in 100 days of maturity in beetroot. However, in beetroot the contents of vitamin C were highest at 80 days of maturity. Thus beetroot can be consumed at different stages of development for gaining highest amount of different nutrients and bioactive phytochemicals and antioxidants. But at 100 days of maturity the content of anti-nutrients (oxalic acid and saponins) were highest. So different processing's and cooking methods can be used for further reduction of anti-nutrients and enhancing the availability of dietary essential minerals of beetroot and to prepare products which are nutritionally superior.

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