Food Chemistry 135 (2012) 460–463

Contents lists available at SciVerse ScienceDirect

Food Chemistry

journal homepage: www.elsevier.com/locate/foodchem

Nutritional and antioxidant profiles of pumpkin (*Cucurbita pepo* Linn.) immature and mature fruits as influenced by NPK fertilizer

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ARTICLE INFO

Article history:
Received 21 December 2011
Received in revised form 2 April 2012
Accepted 23 April 2012
Available online 30 April 2012

Keywords:
Antioxidant
Fertilizer
Fruits
Proximate
Pumpkin

ABSTRACT

This study evaluated the influence of NPK fertilizer on protein, fibre, ash, fat, carbohydrate, antioxidant activities and antioxidant phenolic compounds in immature and mature fruits of pumpkin. The treatment consisted of six NPK levels (0, 50, 100, 150, 200 and 250 kg/ha), and was replicated six times in a randomized complete block design (RCBD). Proximate analysis and antioxidant assays were done using standard analytical methods. At control and lower NPK rates, the proximate compositions and antioxidant profile of pumpkin fruits decreased with increasing NPK fertilizer. Between the control and the highest fertilizer rate, proximate compositions decreased by 7–62% while the antioxidant profile decreased by 13–79% for both immature and mature fruits. Across all the measured parameters, mature fruit had higher proximate contents and higher antioxidant concentrations. For the high health value of pumpkin fruits to be maintained, little or no NPK fertilizer should be applied.

1. Introduction

Fruits and vegetables are naturally rich in nutrients and antioxidants. Biochemical and *in vitro* studies of antioxidants in fruit and vegetables have indicated that they can provide protection against free radical damage. Free radicals, involved in many degenerative diseases of ageing and health hazards, are destroyed by the relatively high antioxidant concentrations in fruit and vegetables (Ames, Shigenaga, & Hagen, 1993; Rice-Evans & Miller, 1995).

Pumpkin, a member of the Cucurbitaceae family, is one of the largest families in the vegetable kingdom, consisting of largest number of edible plant species (Manjunath Prasad, Ashok, Vyakaranahal, Nadaf, & Hosamani, 2008). Pumpkin fruits are extensively used as vegetable, both at immature and mature stages. The immature fruits, called courgettes, are eaten as a vegetable, boiled, fried or stuffed. Mature fruits, called pumpkin, are used peeled and cooked or prepared as pumpkin pie. In west Africa, the fruits are used in soup. Gem squashes, small globular fruits popular in southern Africa, are cooked whole or cut in half, and their flesh is scooped out and eaten (Messiaen & Fagbayide, 2004). In the Middle East, nearly mature fruits of 'Causa' are stuffed with meat and other ingredients, followed by baking. Native Americans dried strips of squash flesh in the sun for preservation. Today, summer squash is usually cooked by boiling or frying, and winter squash by baking, boiling or microwaving (Robinson & Decker-Walters, 1997). In Africa, the fruit pulp is used as a poultice to treat burns and inflammations and as a cooling compress to treat headache and neuralgia; it has also been applied to tumours (Messiaen & Fagbayide, 2004).

In Nigeria, immature pumpkin fruits, called "kundu" in the south west, are consumed steamed in combination with the pumpkin young shoots called "Gboro". The mature fruits are consumed fresh or dried and added to soups. The immature fruit, at 14 days after fruit formation, contains between 75% and 85% water and weighs 120–400 g per fruit, while the water content of mature fruit has 75–80% and weighs between 1 and 2.5 kg per fruit.

In modern agriculture, chemical fertilizers constitute the major portion of total cost of crop production. The problem of soil limited availability and low fertility is solved, partially or completely, by using inorganic fertilizer to enhance crop productivity. This practice also has effect on the chemical composition of crops grown in such soils. This study investigated the effect of NPK 15:15:15 compound fertilizer on the proximate contents, antioxidant activities and phenolic compounds of pumpkin immature and mature fruits. NPK 15:15:15 compound fertilizer was chosen because it is the most available fertilizer and most commonly used by the farmers.

2. Material and methods

2.1. Field study

Field study was conducted at the Teaching and Research Farm, Obafemi Awolowo Univerity, Ile-Ife, Nigeria, for 2 seasons in 2010.

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http://dx.doi.org/10.1016/j.foodchem.2012.04.124

Pumpkin fruits were raised from seeds in a randomized complete block design, consisting of six rates of NPK 15:15:15 fertilizer at 0, 50, 100, 150, 200, 250 kg/ha with six replications of 10×12 m plot size. Immature (14-day old) fruits ("kundu") were harvested from all the plots to form six composite samples. The samples were dried at 70 °C for 24 h, milled and kept in the refrigerator prior to analysis. At harvest, mature (40-day old) fruits of pumpkin from different plots were also oven-dried at 70 °C for 24 h. The composite samples for the six treatments were also milled and stored in the refrigerator.

2.2. Laboratory analyses

Five grammes each of the composite samples were extracted (cold extraction, i.e. extraction not involving heat), for 24 h, using 80% methanol. The crude extract was obtained by evaporation of the methanol-soluble extract to dryness in a rotary evaporator at 45 °C. The antioxidant activities or hydrogen-donating or radical-scavenging of the extract was determined using the stable radical DPPH· (2, 2-diphenyl-2-picrylhydrazyl hydrate) according to the method described by Brand-Williams, Cuvelier, & Berset (1995). DPPH· reacts with an antioxidant compound which can donate hydrogen, it is reduced. The change in colour from deep violet to light yellow was measured spectrophotometrically at 517 nm. Total phenol content was determined by the method of Singleton and Rossi (1965), using the Folin–Ciocalteau reagent in alkaline medium. Total flavonoid content was determined using the $AlCl_3$ method, as described by Lamaison & Carnet, 1990. The proanthocyanidin content was determined by a modified method of Porter, Hristch, and Chan (1986), using the $AlCl_3$/Butan – 1–0 l assay method. The total anthocyanin content of the test samples was determined using the pH differential method of Fuleki and Francis (1968), as described by Guisti (2001). Crude protein, carbohydrate, ash, crude fibre, ether extract (fat) and moisture contents were determined using the routine chemical analytical methods of the AOAC (1995).

2.3. Statistical analysis

All data were subjected to combined analysis of variance SAS (2003). Means squares, where significantly different for NPK levels were separated using the Duncan multiple range test (DMRT) at 5% level of probability. Least significant difference (LSD) and regression curves were used to compare the nutritional and antioxidant profiles of pumpkin fruits at different maturity stages.

3. Results

The protein, fat, ash, crude fibre and carbohydrate in pumpkin young and mature fruits were generally significantly influenced by fertilizer, the values decreased with increasing fertilizer. Fertilizer influence showed that proximate values of protein, fat, ash and crude fibre in pumpkin young and mature fruits were similar at 0, 50 and 100 kg NPK/ha. The nutrient values were reduced significantly when compared to the control from the application of 150 kg/ha of NPK and this continued to the highest fertilizer rate (250 kg NPK/ha). The carbohydrate result contrasted with the others. The values increased with increased fertilizer rates. Carbohydrate content in pumpkin young and mature fruits was highest with the application of 200 and 250 kg/ha and lowest in 0 and 50 kg/ha (Figs. 1–5).

In comparison, at 0–150 kg/ha fertilizer application, the protein content of the mature fruit was significantly ($p = 0.05$) higher than that of young fruit, while the fat content was only significantly higher at 250 kg/ha (Figs. 1 and 2). The ash content, which is an index of the mineral content, was significantly higher also in the mature fruit across all the fertilizer levels (Fig. 3). The crude fibre of both young and mature fruits were not significantly different at 0–100 kg/ha fertilizer application; however, at higher fertilizer level, crude fibre was significantly higher in the mature fruit (Fig. 4). The carbohydrate content of both young and mature fruits increased with increasing fertilizer application, but the mature fruit had significantly higher carbohydrate content at 0–100 and 250 kg/ha fertilizer rates (Fig. 5).

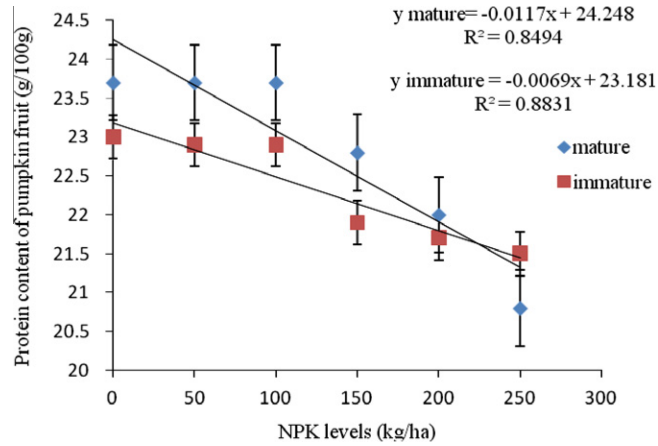

Fig. 1. Protein contents of pumpkin young and mature fruits (dry weight basis).

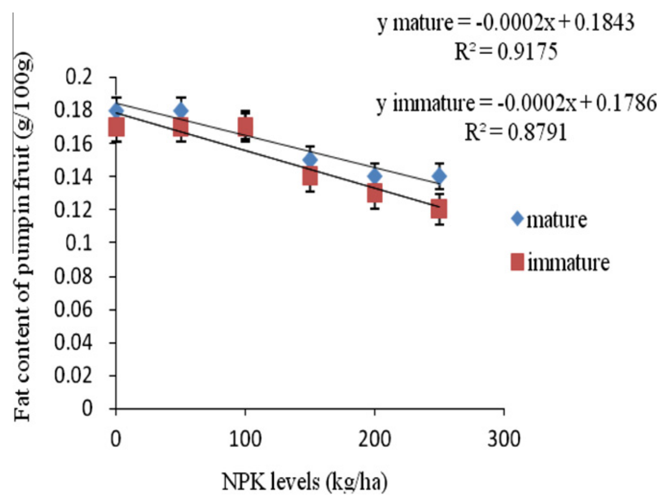

Fig. 2. Fat contents of pumpkin young and mature fruits (dry weight basis).

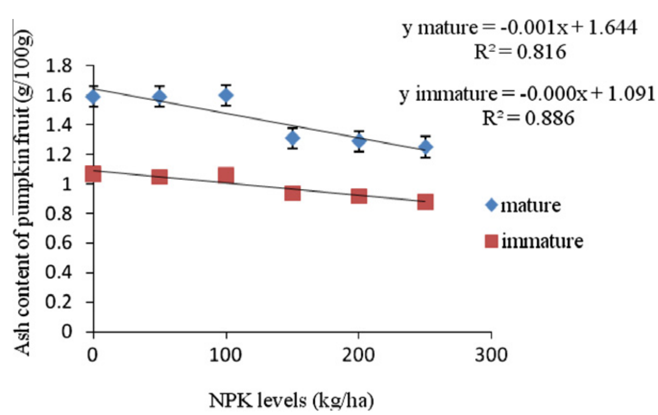

Fig. 3. Ash contents of pumpkin young and mature fruits (dry weight basis).

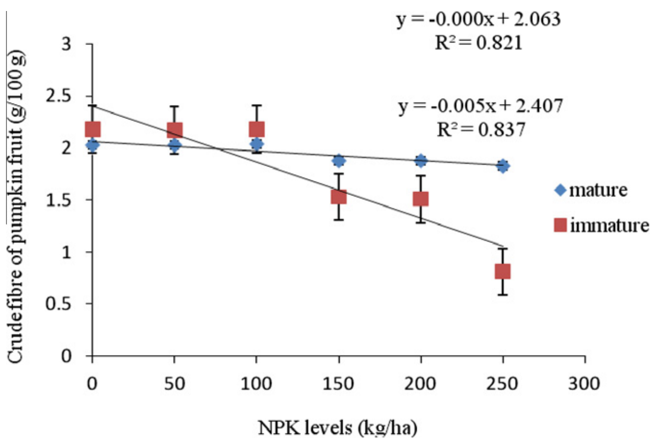

Fig. 4. Crude fibre contents of pumpkin young and mature fruits (dry weight basis).

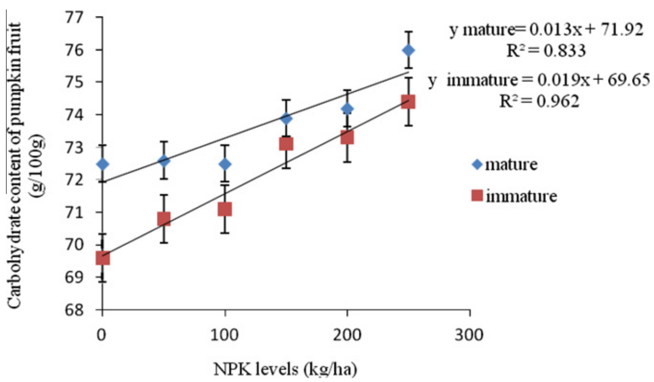

Fig. 5. Carbohydrate contents of pumpkin young and mature fruits (dry weight basis).

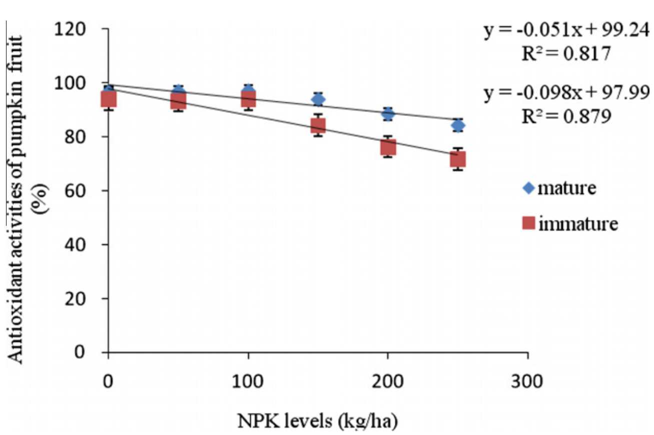

Fig. 6. Antioxidant activities of pumpkin young and mature fruits (dry weight basis).

Fig. 6 shows that antioxidant activities in pumpkin young and mature fruits were significantly influenced by fertilizer. Fertilizer influence showed consistent decrease in values of antioxidant activities as fertilizer rates increased. The control had significantly higher values than the values from the application of 150–250 kg/ha. The addition of 50 and 100 kg gave similar antioxidant activities with the zero fertilisation at 0.05 probability level, in both young and mature fruits. However, the antioxidant activities in the mature fruit were higher than those of young fruit, though not significantly different at 0–100 kg/ha fertilizer application.

Fertilizer influence showed consistent reduction in values of antioxidant phenolic compounds of both mature and young fruits as fertilizer rates increased. The control had a significantly higher values than the values from the application of 150–250 kg/ha. The addition of 50 and 100 kg gave similar phenol, flavonoid, anthocyanin and proanthocyanidin concentrations with the zero fertilisation at 0.05 probability level (Table 2). The concentration of total phenol in mature fruit is 71% higher than that in the young fruit. Flavonoid, anthocyanin and proanthocyanidin concentrations were 4.5%, 14% and 17% higher, respectively, in the mature fruit than in the young fruit (Table 1).

4. Discussion

The protein in immature and mature fruits was 23 g/100 g while the carbohydrate content was 70% at 0–100 kg NPK/ha. This portion can supply more balanced nutrients than most vegetable crops, such as amaranth, with protein content (5.9/100 g), *Celosia argentea* (5.8/100 g), cowpea leaves (21/100 g), cabbage (1.6/100 g), carrot (1.0/100 g), fluted pumpkin leaves (4.3/100 g), *Celosia trygina* (5.1/100 g) and *Momordica balsamina* (28/100 g) on a dry weight basis (FAO, 2010; Flyman & Afolayan, 2007; Oloyede, Oloyede, Obuotor, & Ibironke, 2011; Osagie & Offiong, 1997). Its low cholesterol content makes it suitable as a diet for obese and hypertensive patients. The high protein content serves as a good supplement to cereals grains or as a meat substitute (Bressani, 1985; Matlhare et al., 1999) for poor rural communities. The antioxidant activities of pumpkin fruits analysed in this study ranged from 72% to 97%, irrespective of the harvesting stages and fertilizer application levels. These values are above what Olajire, 2011 reported for some commonly consumed vegetables in Nigeria. However, the concentrations of the antioxidant phenolic compounds analysed were slightly lower.

The addition of fertilizer reduced the protein by 7%, antioxidant activities by 24% and antioxidant phenolic compounds by 19–71%. These reductions were due to the increasing N concentration in plants through fertilizer. The influence of N on protein concentration may be worse, when the concentration of essential nutrients in protein, which determines the quality of the fruits, is considered. The antioxidant components were reduced by 20–30% at fertilizer rates above 100 kg NPK/ha. The result is similar to the reduction in total phenolics and antioxidant activities observed in asparagus (Paschold, Hermann, & Artell, 1999) due to increased N fertilisation. At the University of Illinois, comparison was made of flavonoid content in tomatoes. Plants with limited N were shown to accumulate more flavonoids than those that are well supplied with inorganic fertilizers. It was concluded that synthetic fertilizers, in which N is easily accessible to the plant, may reduce the health benefits of tomatoes (Mitchell, & Chassy, 2007). This agrees with the observation in this study that over fertilisation with N reduces nutrient quality and the health benefits of pumpkin fruits.

The proximate contents, the antioxidant activities and the concentrations of the antioxidant phenolic compounds obtained in pumpkin mature fruits were higher than those in the young fruit. Biologically active compounds have been found to increase in plant leaves during the process of maturation, depending on the different biosynthetic pathways and mechanisms of metabolic control (Bergquist, 2006; Khader & Rama, 1998; Khader & Rama, 2003; Lee & Kader, 2000). However, according to Baranga (1983), the nutrient quality of plant leaves generally declines with advancing maturity, resulting in a decrease in protein levels and a concomitant increase in the amount of indigestible structural carbohydrates.

In conclusion, the mature fruit is better than the young fruit in terms of nutritional and antioxidant profiles while the fruit requires little or no NPK fertilizer for optimal nutritional values.

Table 1
Antioxidant phenolic compounds in pumpkin fruits, as affected by NPK fertilizer (dry weight basis).

Phenolic antioxidant	Total phenols (mg/100 g)		Flavonoids (mg/100 g)		Anthocyanins (mg/100 g)		Proanthocyanidins (mg/100 g)	
NPK level (kg ha^{-1})	Mature fruit	Immature fruit	Mature fruit	Immature fruit	Mature fruit	Immature fruit	Mature fruit	Immature fruit
0	33.5a	10.3a	6.01a	5.44a	0.0220a	0.016a	0.025a	0.021a
50	33.3a	10.2a	5.95a	5.47a	0.0220a	0.016a	0.026a	0.021a
100	33.3a	10.4a	5.93a	5.46a	0.0220a	0.016a	0.025a	0.021a
150	20.4b	7.71b	3.19b	3.74b	0.0098b	0.011b	0.015b	0.015b
200	14.7c	6.65c	2.78c	2.65c	0.0066c	0.009c	0.012c	0.008c
250	7.08d	4.94d	2.34d	2.51d	0.0050d	0.007d	0.005d	0.006d

Means with the same letter in each column are not significantly different at 5% level of probability using Duncan's multiple range test.

Table 2
Antioxidant phenolic compounds in pumpkin young and mature fruits across the NPK levels (dry weight basis).

Phenolic antioxidant	Total phenols (mg/100 g)	Flavonoids (mg/100 g)	Anthocyanins (mg/100 g)	Proanthocyanidins (mg/100 g)
Mature fruit	23.7	4.4	0.14	0.18
Immature fruit	8.4	4.2	0.12	0.15
LSD (0.05)	2.0	NS	0.004	0.008

NS = not significant at 5% level of probability.

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