Australian Journal of Basic and Applied Sciences, 3(4): 3609-3616, 2009
ISSN 1991-8178

Antioxidant Activities, Total Anthocyanins, Phenolics and Flavonoids Contents of Some Sweetpotato Genotypes under Stress of Different Concentrations of Sucrose and Sorbitol.

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Abstract: Antioxidant activity, total anthocyanins, phenols and flavonoids Contents were monitored in a callus induced from stem segment culture of sweetpotato (*Ipomoea batatas L.*) Abees, the Egyptian orange-fleshed cultivar in addition to four genotypes (199062.1-1999026.1-199015.14-199004.2) obtained from the International Potato Centre (CIP). *In vitro* callus culture were grown over period of four weeks in dark on Murashige and Skoog (MS) medium supplemented with different concentrations of osmotic potential induced by (0.03, 0.1, 0.15 molar) sucrose or sorbitol, MS medium without any sugar used as a control. It was found that the dominant DPPH radical-scavengers of Abees cultivar was due to the presence of anthocyanins and phenolic compounds rather than flavonoides, while at 199062.1 was as a consequence of flavonoids and at 199004.2 was a result of the phenolic compounds. Anthocyanins and flavonoids accumulation stimulated by increasing the sugar concentration up to 0.1 M and 0.15 M respectively using sucrose; however phenol compounds increased in 0.1 M sorbitol. Radical-scavenging activity contributors varied according to the cultivar and the osmotic potential.

Key words:sweetpotato; antioxidant activities; anthocyanin; phenolics; flavonoides; radical scavenging.

INTRODUCTION

Antioxidants are substances or nutrients presented in our foods and can increase cellular defense and help prevent oxidation damage to cellular component of our bodies in the hope of maintaining health and preventing diseases such as cancer and coronary heart disease in addition to many other uses in medicine and industrial field, for instance cosmetic manufacturing and food preservation and colorant.

On the other hand, the synthetic antioxidants have restricted use in food, as they are suspected to be carcinogenic (Namiki, 1990). Therefore the importance of searching for and exploiting natural antioxidants has increased greatly in recent years (Patil *et al.*, 2009). While, antioxidant are a class of plant secondary metabolites, the plant kingdom offers vast array of valuable natural polyphenolic compounds, several isolated plant constituents as well as crude extracts of vegetables and fruits have been recognized to possess beneficial effects against free radicals in biological systems as antioxidants (Yi Fang and Xianzona, 2002). This beneficial effect of fruits and vegetables can be attributed to the antioxidant capacity of the polyphenolic compounds which present in them (Hertog *et al.*, 1993), Polyphenolic compounds are divided to two major subgroups: flavonoids and phenolic acids. Flavonoids are concerned the major important polyphenolic component presented in food. It includes anthocyanins, proanthocyanidins, flavonols and catechins. They attracted attention in the past few years since it has been established that they are found universally and have potentially protective roles for human health. Anthocyanins are example of flavonoid polyphenols that is intensively investigated for its *in vitro* production. Anthocyanins possess a high thermostability (Odake *et al.,* 1992&1994) and contribute towards antioxidative activity (Furuta *et al.*, 1998). According to Kamei *et al.* (1993), anthocyanins inhibit the growth of human cancer cells; therefore, the addition of natural anthocyanins as food colorants would not only enhance the decorative value of the food but also improve the positive properties.

Advances in the area of tissue culture for the production of secondary metabolites have made it possible to increase the yield of a wide variety of substances with pharmaceutical values such as alkaloids, terpenoids,

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steroids, phenolics and flavonoids (Rout *et al.*, 2000). *In vitro* technology offered control conditions for culture medium compositions as well as incubation conditions to globalize the production of desired variants in shorter and flexible production cycles.

Sugars were reported to have an essential role in general metabolism that regulates many important processes in all stages of the plant life cycle (Smeekens, 2000 and Rolland *et al*. 2002). The stimulatory effects of sugars on anthocyanin biosynthesis in different organs of several plant species have been demonstrated. Anthocyanin biosynthesis was improved in radish (*Raphanus Sativus*) hypocotyls (Hara *et al.*, 2003), and arabidopsis cotyledons or leaves in reflect to sugar concentrations of culturing medium (Sheng *et al.*, 2005). Sweetpotato (*Ipomoea batatas L.*) a root crop cultivated for centuries in various geographic regions and today the world's sixth largest food crop. This crop has been known to have some good components for maintaining and improving human health (Mano *et al.*, 2007), it contains vitamins and minerals at favorable ratios (Woolfe, 1992 and El-Bastawesy *et al.*, 2007) it accumulates high level of antioxidative substances, (Hayase and Kato, 1984; Furuta *et a.*, 1998; Martin *et al*, 2003; Yoshimoto *et al*, 2001; Dong *et al*, 2004), such as anthocyanins and phenolic acids. Sweetpotato can utilize as a high-quality natural food colorant and at the same time being a superior health protector against cancer, aging and degenerative diseases. However its potentiality as an applicant for industry is neglected. This paper presents our results on the evaluation of antioxidant activities, total flavonoids, total phenolics and anthocyanins contents of callus induced from five different genotypes of sweetpotato under different sugar types and concentrations.

MATERIALS AND METHODS

Plant Material:

Four different sweetpotato genotypes (199062.1 – 1999026.1 – 199015.14 – 199004.2) delivered from International Potato Centre (CIP), in addition to the Orange-fleshed Egyptian local variety Abees were routinely in vitro subcultured every 6 weeks onto a fresh MS basal medium (Murashige and Skoog 1962) supplemented with 3% sucrose and 0.8% agar and maintained at 28 °C under cool white fluorescent light 300 foot-candle for 16 h/8h light/dark cycle.

Callus Induction:

Dissected stem segments not including buds from the *in vitro* manipulated stock were used as an explant. Stem segments (C. 5mm) were cultured onto petri dishes (9cm) containing 10 ml of MS-agar solidified basal medium enriched with 0.2 mg/l picloram in addition to (0.03 or 0.1 or 0.15 molar) sucrose or sorbitol. A basal MS-agar medium without any osmotic source was used as a control. Cultures were then incubated in dark in controlled growth chamber at temperature of 28 °C for 4 weeks .The experiment was carried out two times. Fresh callus were instantly frozen under liquid nitrogen and stored at -20 °C for further analysis. While the dried Callus obtained after the incubation in an oven at 60 °C for 24 h.

Antioxidant Activity (DPPH Assay):

The free radical scavaging activity using the 1,1-diphenyl-2-picryl-hydrazil (DPPH) reagent was determined according to (Brand-Williams *et al.*,1995) The powdered dry callus (1g) was extracted with 50% methanol:water to 0.75 ml of the extract sample 1.5 ml of freshly prepared methanolic DPPH solution (20 µg ml^{-1}) was added and stirred. The decolorizing process was recorded after 5 min. of reaction at 517 nm and compared with a blank control.

Antioxidant activity = [(control absorbance-sample absorbance) / control absorbance] X 100

Determination of Total Anthocyanins:

Anthocyanins were extracted overnight from fresh callus with ethanol and 1% HCl (85:15) at 4°C. The optical density of the extract solution was measured at 535 nm. The total anthocyanins concentrations were calculated after (Francis, 1982) using the extinction coefficient

$\left(E_{1\text{cm}}^{1\%} = 98.2 \text{ at } 535 \text{ nm}\right)$

Determination of Total Phenolics:

The total phenolics content of ethanol extract of dried sweetpotato callus was determined according to the method described by (Makkar *et al.*, 1997). Aliquats of the extract were taken in a 15 ml glass tube and made up to the volume of 1ml with distilled water. Then 0.5 ml of Folin-Ciocalten reagent (1:1 with water) and 2.5

ml of sodium carbonate solution (20%) were added sequentially in each tube. after vortexing tubes were incubated at room temperature in the dark for 40 min. Absorbance was measured at 725 nm against the reagent blank. The amount of total phenolics was calculated as gallic and equivalent from a calibration curve.

Determination of Total Flavonoids:

The $AlCl_3$ method (Lamison and Carnet 1990) was used for the determination of the total flavonoid content of the sample extracts. Aliquats of 1.5 ml of extracts were added to equal volumes of a solution of 2% $AlCl_3.6H_2O$ (2 g in 100 ml methanol). The mixture was vigorously shaken and absorbance at 367 nm was read after 10 min. of incubation. Flavonoid contents were expressed as mg catchin equivalent/g dry weight.

Statistical Analysis:

Data obtained was recorded after four weeks, triplicate analysis for each measurement were conducted for each sample and were exposed to the proper statistical analysis of variance as outlined by (Gomez and Gomez 1984) based on MSTATC program. Means obtained were differentiated using Duncan's new multiple range test as described by (Duncan 1955).

RESULTS AND DISCUSSION

Callus Induction:

Callus culture has been successfully used for the production of plant secondary metabolites over their production from the intact plants since their *in vitro* production is apart of climatic and seasonal factors. It allows flexible production cycles of secondary metabolites. Additionally, the *in vitro* production overcomes the secondary metabolites asymmetrical distribution in the intact plants (Verpoorte, 2002; Vanisree and Tsay 2004; Callebaut, *et al*, 1993). In the extensively studied *I.batatas*, in the tissue cultures induced from storage roots the accumulation of anthocyanins can even exceed the tuberousroots by several times. The production of valuable di-acylated cyanidine glycosides from callus and cell suspension as well as their regulation towards overproduction and their more desirable profile has been reported (Terahara *et al*, 2000, Terahara *et al*, 2004 and Konczak *et al*, 2005). However, media composition has to be optimized for an intensive biomass increase and the accumulation of the desired metabolite, for efficient sweetpotato callus production (Newell *et al*, 1995; Dhir *et al*, 1998; Lawton *et al*, 2000; and Gong *et al*, 2001) maintained sweetpotato explants onto auxin cytokinin enriched medium as a callus initiation medium. On the other hand, Otani and Shimada 1996; Al-Mazrooei *et al*, 1997; Triqui *et al*, 2007 used initiation medium supplemented with picloram as an auxin source to induce callus from sweetpotato explants. In this study, results indicated that medium supplemented by picloram is favourable to induce callus for the five sweetpotato genotypes used in this study (data are not shown). It could be suggested that the response of cultures to growth regulators supplements is influenced by the endogenous levels of growth regulators within the cells. Moreover, genetic differences among genotypes of sweetpotato could describe the changed response among sweetpotato genotypes to growth regulators.

Antioxidant Activity:

Plant-derived Antioxidant increasingly become important dietary factors (Aruoma 2003 and Ferguson 2001). Due to Boliver, 2003 and Furuta, 1998 who demonstrated that antioxidative and radical scavenging activities are observed in all sweetpotato varities with white, yellow, orange and purple flesh. The influence of a supply of sucrose or sorbitol in different concentrations on antioxidant properties (Anthocyanins, total phenols, total flavonoids and antioxidant activities) of a callus culture derived from five different sweetpotato genotypes were evaluated. Produced callus were incubated on MS medium supplemented with (0.03, 0.1and 0.15 M) of sucrose or sorbitol or without for 4 weeks in dark at 28°C. Antioxidant activity as evaluated using DPPH reagent are shown in Table (1) indicated that, the callus derived from Abees cultivar, and 199062.1 genotype possess the highest antioxidant activity among all tested genotypes. Sorbitol in concentrations of 0.03 M or 0.15 M resulted in increasing the activity of antioxidant in Abees cultivar whilst the sucrose or sorbitol free-media were preferable for 199062.1 genotype which obtained the same antioxidant activity of Abees. This can be due to the interaction between the genotype and its endogenous reducing sugar level. (Miura *et al*, 1998; Luczkiewicz and Cisowski, 2001) indicated that a range of environmental nutritional factors and genotype variations a influence the biosynthetic pathways of the secondary metabolites, thus the media composition has to be optimized for the accumulation of the desired metabolite. Thimann *et al*, 1950 and Wong *et al*, 1974, reported that sugars are one of the most important externally substances for the phenolic biosynthetic pathway, it is also proofed by Solfanelli, *et al.,* 2006 who found that the induction of flavonoid/anthocyanin synthesis genes is sugar specific.

Table 1: Effect of sugar concentration on antioxidant capacity of sweetpotato genotypes.

Concentration (K)		Genotypes (C)					KxO	O	K
Molar	Sugar (O)	1	2	3	4	5	mean	mean	
0.00	Sucrose	45.450^{NP}	64.900^{B}	51.383^{IK}	62.243^{BD}	52.140^{HK}	55.223 B		
	Sorbitol	45.450^{MP}	64.900^{B}	51.383^{IK}	62.243^{BD}	52.140^{HK}	55.223 B		
	KxC	**45.450k**	**64.900a**	**51.383ij**	**62.243ab**	**52.140hj**			**55.223B**
0.03	Sucrose	56.350^{EH}	58.803^{DG}	59.750^{DE}	60.690^{BE}	57.673^{DG}	58.653 A		
	Sorbitol	72.940^{A}	62.127^{BD}	52.807^{HK}	49.030^{KN}	44.127^{OP}	56.206 B		
	KxC	**64.645a**	**60.465bc**	**56.278df**	**54.860eh**	**50.900j**			**57.430 A**
0.10	Sucrose	57.983^{DG}	60.753^{BE}	61.573^{BD}	62.010^{BD}	54.507^{GJ}	59.365 A		
	Sorbitol	64.303^{BC}	55.287^{FI}	46.757^{LO}	52.520^{HK}	38.513^{Q}	51.476 C		
	KxC	**61.143b**	**58.020cd**	**54.165fi**	**57.265de**	**46.510k**			**55.421 B**
0.15	Sucrose	57.607^{DG}	60.000^{CE}	59.183^{DF}	54.343^{GJ}	52.580^{HK}	56.743 B		
	Sorbitol	71.043^{A}	50.547^{JL}	45.587^{MP}	49.743^{KM}	41.597^{PQ}	51.703 C		
	KxC	**64.325a**	**55.273dg**	**52.385gj**	**52.043hj**	**47.088k**			**54.223 B**
OxC	**Sucrose**	**54.347d**	**61.114b**	**57.972c**	**59.822bc**	**54.225 d**		**57.496 A**	
	Sorbitol	**63.434a**	**58.215c**	**49.133e**	**53.384d**	**44.094 f**		**53.652 B**	
Cultivar mean		58.891 a	59.665 a	53.553 c	56.603 b	49.160 d			

- Means followed by the same letters are not significantly different at p=0.05 according to Duncan's multiple range tests.

- 1,2,3,4,5 represent the sweetpotato genotypes (1) Abees cultivar, (2) 1999062.1, (3) 199026.1, (4) 199015.14, (5) 199004.2.

On the other hand, Table (2, 3, 4) shows that there was no relation between the increasment of anthocyanin, total phenolics, total flavonoids and the antioxidant activity, this findings led us to speculate that the highest radical-scavenging activity of Abees is due to another antioxidant substances not detected in our study such as β-carotene and vitamin C, our previous data (not shown) indicated that Abees genotype has high β-carotene contents. This observation come with the findings of (Osamu and Makoto 2002; Oki, *et al*, 2002; Martin *et al*., 2003) whom reported that anthocyanins alone could not account for all the antioxidant activity.

Table 2: Effect of sugar concentration on total anthocyanins content of sweetpotato genotypes (mg/100g).

Concentration (K)		Genotypes (C)					KxO	O	K
Molar	Sugar (O)	1	2	3	4	5	mean	mean	
0.00	Sucrose	10.591^{B}	5.307^{GI}	5.353^{GI}	8.273^{CE}	4.227^{HJ}	6.750 C		
	Sorbitol	10.591^{B}	5.307^{GI}	5.353^{GI}	8.273^{CE}	4.227^{HJ}	7.262 C		
	KxC	**10.591a**	**5.307fh**	**5.353fh**	**8.273bc**	**4.227h**			**6.750 A**
0.03	Sucrose	10.635^{B}	7.487^{DF}	9.517^{BC}	0.000^{K}	8.673^{CE}	8.334 B		
	Sorbitol	3.780^{LJ}	2.920^{J}	4.533^{HJ}	3.270^{J}	3.480^{LJ}	9.559 A		
	KxC	**7.208cd**	**5.203fh**	**7.025de**	**1.635i**	**6.077df**			**5.430 B**
0.1	Sucrose	9.968^{BC}	7.477^{DF}	5.767^{FH}	9.013^{BD}	9.447^{BC}	6.750 C		
	Sorbitol	3.140^{J}	3.033^{J}	3.087^{J}	3.567^{LJ}	4.413^{HJ}	3.597 D		
	KxC	**6.554df**	**5.255fh**	**4.427gh**	**6.290df**	**6.930de**			**5.891 B**
0.15	Sucrose	9.053^{BD}	8.627^{CE}	6.893^{EG}	8.180^{CE}	15.040^{A}	3.448 D		
	Sorbitol	3.220^{J}	4.327^{HJ}	4.513^{HJ}	3.607^{LJ}	3.727^{B}	3.879 D		
	KxC	**6.137df**	**6.477df**	**5.703eg**	**5.893df**	**9.383b**			**6.719 A**
OxC	**Sucrose**	**10.062a**	**7.224b**	**6.882b**	**6.367b**	**9.347a**		**7.976 A**	
	Sorbitol	**5.183c**	**3.897d**	**4.372cd**	**4.679cd**	**3.962d**		**4.418 B**	
Cultivar mean		7.622 a	5.560 c	5.627 c	5.523 c	6.654 b			

- Means followed by the same letters are not significantly different at p=0.05 according to Duncan's multiple range tests.

- 1,2,3,4,5 represent the sweetpotato genotypes (1) Abees cultivar, (2) 1999062.1, (3) 199026.1, (4) 199015.14, (5) 199004.2.

Table 3: Effect of sugar concentration on total Phenolics content of sweetpotato genotypes (mg/g).

Concentration (K)		Genotypes (C)					KxO	O	K
Molar	Sugar (O)	1	2	3	4	5	mean	mean	
0.00	Sucrose	20.976^{M}	32.625^{H}	25.831^{J}	18.770^{O}	36.504^{G}	26.941D		
	Sorbitol	20.976^{M}	32.625^{H}	25.831^{J}	18.770^{O}	36.504^{G}	26.941D		
	KxC	**20.976h**	**32.625c**	**25.831f**	**18.770i**	**36.504b**			**26.941C**
0.03	Sucrose	25.095^{JK}	6.174^{T}	10.000^{RS}	23.322^{L}	23.358^{L}	17.590 F		
	Sorbitol	15.035^{PQ}	46.129^{D}	19.137^{NO}	18.687^{O}	25.032^{JK}	24.804 E		
	KxC	**20.065h**	**26.152f**	**14.569j**	**21.005h**	**24.195g**			**21.197D**
0.10	Sucrose	75.203^{A}	8.968^{S}	24.117^{KL}	9.941^{RS}	40.490^{F}	31.744 A		
	Sorbitol	13.710^{Q}	48.155^{C}	23.037^{L}	20.434^{MN}	18.068^{O}	24.681 E		
	KxC	**44.457a**	**28.561e**	**23.577g**	**15.188j**	**29.279e**			**28.212B**
0.15	Sucrose	51.799^{B}	15.637^{P}	32.292^{H}	24.547^{JL}	28.944^{I}	30.644 B		
	Sorbitol	10.930^{R}	42.270^{E}	32.569^{H}	32.809^{H}	29.100^{I}	29.535 C		
	KxC	**31.364d**	**28.954e**	**32.431c**	**28.678e**	**29.022e**			**30.090A**
OxC	**Sucrose**	**43.268a**	**15.851h**	**23.060f**	**19.145g**	**32.324c**		**26.730 A**	
	Sorbitol	**15.163h**	**42.295b**	**25.144e**	**22.675f**	**27.176d**		**26.490 A**	
Cultivar mean		29.216 b	29.073 b	24.102 c	20.910 d	29.750 a			

- Means followed by the same letters are not significantly different at p=0.05 according to Duncan's multiple range tests.
- 1,2,3,4,5 represent the sweetpotato genotypes (1) Abees cultivar, (2) 1999062.1, (3) 199026.1, (4) 199015.14, (5) 199004.2.

Table 4: Effect of sugar concentration on total flavonoics content of sweetpotato genotypes (mg/g).

Concentration (K)		Genotypes (C)					KxO	O	K
Molar	Sugar (O)	1	2	3	4	5	mean	mean	
0.00	Sucrose	2.464^{HK}	4.570^{CD}	3.511^{EF}	2.452^{HK}	3.480^{EF}	3.295 AB		
	Sorbitol	2.464^{HK}	4.570^{D}	3.511^{EF}	2.452^{HK}	3.480^{EF}	3.295 AB		
	KxC	**2.464cd**	**4.570a**	**3.511b**	**2.452cd**	**3.480b**			**3.295 A**
0.03	Sucrose	2.085^{KN}	0.658^{P}	2.738^{GJ}	2.281^{IL}	2.183^{JM}	1.989 E		
	Sorbitol	1.594^{NO}	6.498^{A}	2.237^{JL}	2.250^{IL}	2.799^{GI}	3.075 B		
	KxC	**1.839g**	**3.578b**	**2.488cd**	**2.265df**	**2.491cd**			**2.532 C**
0.10	Sucrose	5.048^{BC}	0.877^{P}	2.226^{JL}	0.796^{P}	3.222^{FG}	2.434 D		
	Sorbitol	1.790^{LO}	6.195^{A}	2.404^{HK}	3.164^{FG}	0.634^{P}	2.937 C		
	KxC	**3.419b**	**3.536b**	**2.315ce**	**1.980eg**	**1.928fg**			**2.636 C**
0.15	Sucrose	3.600^{EF}	1.488^{O}	3.202^{FG}	1.662^{MO}	2.252^{IL}	2.441 D		
	Sorbitol	1.797^{LO}	5.311^{B}	3.877^{E}	3.544^{EF}	2.913^{GH}	3.488 A		
	KxC	**2.699c**	**3.399b**	**3.540b**	**2.603cd**	**2.583cd**			**2.965 B**
OxC	**Sucrose**	**3.299b**	**1.898e**	**2.919c**	**1.798e**	**2.784c**		**2.540 B**	
	Sorbitol	**1.911e**	**5.643a**	**3.007c**	**2.852c**	**2.457d**		**3.174 A**	
Cultivar mean		2.605 c	3.771 a	2.963 b	2.325 d	2.620 c			

- Means followed by the same letters are not significantly different at p=0.05 according to Duncan's multiple range tests.
- 1,2,3,4,5 represent the sweetpotato genotypes (1) Abees cultivar, (2) 1999062.1, (3) 199026.1, (4) 199015.14, (5) 199004.2.

Anthocyanins Content:

Anthocyanins have been shown to be more effective antioxidant, as Abees showed the highest antioxidant activity among the other tested sweetpotato genotypes, Abees showed also the highest level of anthocyanins accumulation followed by 199004.2 while 199026.1, 199062.1 and 199015.14 represent a low level of anthocyanins content Table (2). The influence of sucrose or sorbitol on the anthocyanin accumulation was low in Abees, while the high sucrose concentration 0.15 M raised the accumulation of anthocyanins of the callus derived from 199004.2, the other three genotypes did not show a good response for anthocyanins content either with the supplementation of sugars to the media nor without. Although, it seems that sugars used in our experiment does not play an important role in anthocyanin enhancement. It was reported that sugars enhances the anthocyanins biosynthesis of different crops (Hara *et al*, 2003; Sheng, *et al*, 2005; Meyer and Staden 1995; Sato *et al*, 1996). Comparing to our results we conclude that the difference of anthocyanins production in response to supplemented sugar to culture media is varying among the different crops

Phenolics Content:

While the anthocyanin-stimulating effect of external sucrose or sorbitol on callus tissue is not clear yet, the simultaneous effect on total phenolics level induced by sucrose or sorbitol in 0.15 has recognized. Table (3) shows that the high concentration of sucrose or sorbitol increases the Phenolics content respectively in the all genotypes used 199004.2, Abees. 199062.1, 199026.1 and 199015.14. The observation that accumulation of phenolics can be induced greatly in the presence of high level of sugar without anthocyanin induction highlights the potential for independent regulation of these two pools which previously indicated by studies on enzymes involved in phenylpropanoid synthesis and flavonoid synthesis in petunia (Solfanelli *et al*, 2006) and parsley (Ranjeva *et al*, 1975 and Hahlbrock *et al*, 1971)

Flavonoids Contents:

Table (4) shows that among tested sweetpotato genotypes, 199062.1 has the highest total flavonoides content followed by 199026.1, 199004.2, Abees and 199015.14. Although the increase of the total flavonoids in 199062.1 did not enhanced in respect of sugars involved in the media, however the effect of 0.15 M sorbitol on the other genotypes was observed (Table 4).

Conclusion:

In general, the antioxidant activity of the genotypes studied would be as follow:199062.1>Abees>199015.14>199026.1>199004.2, for total anthocyanins content, the order would be as follow: Abbes>199004.2>199026.1>199062.1>199015.14, on total Phenolics content the order was: 199004.2>Abees>199062.1>199026>199015.14, however, on total flavonoids, the order inverted to: 199062.1>199026.1>199004.2>Abees>199015.14. It could be notice also that genotype 199015.14 showed always a weak response with the whole determined characters. Our conclusion is some genotypes has the totipotiential for the induction of some secondary metabolites such as anthocyanins without exogenous enhancers, this is was clear with Abees genotype, it was also observed with 199062.1 genotype that showed a high antioxidant activity with high flavonoids content in a control media, however sugars can play a great role for enhancing the production of the secondary metabolites in some other sweetpotato genotypes or even among the other different crops, the role of sugars in stimulating the production metabolites is differ some suggest that it is an osmotic agent (Sato *et al*, 1996), the other propose it is regulate the responsible synthesis genes (Solfanell *et al*, 2006 and Gollop *et al*, 2002).

The future of sweetpotato with the added benefit of having a high antioxidant activity, total Phenolics, total flavonoids and total anthocyanins make it a promising and healthier alternative to the synthetic antioxidant agents. However other antioxidant substances compounds still need to be identified.

ACKNOWLEDGMENT

We thank the International Potato Centre (CIP), Lima, Peru, for providing the sweetpotato genotypes, Dr. Hala Eissa for her help in statistical analysis and Dr. A. Ashoub for critical review of the manuscript.

REFERENCES

Al-Mazrooei, S., M.H. Bhatti, G.G. Henshaw; N.J. Taylor and D. Blakesley, 1997. Optimization of somatic embryogenesis in fourteen cultivars of sweet potato (*pomoea batatas L. Lam*.). Plant Cell Rep., 16: 710-714.

Aruoma, O.I., 2003. Methodological considerations for characterizing potential antioxidant actions of bioactive components in plant foods. Mutat Res., 523-524, pp: 9-20.

Bolivar, A., C. Cevallos and C.Z. Luis. 2003. Stoichiometric and kinetic studies of phenolic antioxidants from Andean purple corn and red-fleshed sweetpotato. J.Agric. Food Chem., 51: 3313-3319.

Brand-Williams, W., M.E. Cuvelier and C. Berset, 1995. Use of a free radical method to evaluate antioxidant activity. Lebensmittel Wissenschaften und Technologi, 28: 25-30.

Callebaut, A., M. Decleire, and K. Vandermeiren, 1993. Ajuga reptans (Bugle): in vitro production of anthocyanins, in: Y.P.S. Bajaj (Ed.), Bioitechnology in agriculture and forestry, Medicinal and Aromatic plants V, 24, Springer, Berlin, pp: 1-21.

Dhir, S.K., J. Oglesby and A.S. Bhagsari, 1998. Plant regeneration via somatic embryogenesis, and transient gene expression in sweet potato protoplasts. Plant Cell Reports, 17: 665-669.

Dong, J.H., C. Der-Lin, H. Jung Chen and Y. Huei Lin, 2004. Antioxidant and antiproliferative activities of sweetpotato (*pomoea batatasL.Lam* 'taining 57') constituents. Bot. Bull.Acad. Sin., 45: 179-186.

Duncan, D.B., 1955. Multiple range and multiple "F-test". Biometrics, 11: 1-42.

El-Bastawesy, A.M., H. Lobna, M.M. El-Far, 2007. Chemical and technological evaluation of some sweetpotato varities, Arab Univ. J. Agric.Sci., Ain Shams Uni., Cairo, 16(1): 87-96.

Ferguson, L.R., 2001. Role of plant polyphenols in genomic stability. Mutation Research, 475: 89-111.

Francis, J.F., 1982. Analysis of anthocyanins in : Markakis P (Ed) Anthocyanins as good colors, pp: 181-208. Academic press, London.

Furuta, S., I. Suda, Y. Nishiba and O. Yamakawa, 1998. High tert-butylperoxyl radical scavenging activities of sweetpotato cultivars with purple flesh. Food Sci. Technol. Int. Tokyo, 29: 33-35.

Gollop, R., S. Even, V. Colova and A. Peri, 2002. Expression of the grape dihydroflavonol reductase gene and analysis of its promoter region. J Exp Bot, 53: 1397-1409.

Gomez, K.A. and A.A. Gomez, 1984. Statistical procedures for agricultural research, pp: 680, 2nd ed j. wiley and sons ltd., New York.

Gong, Y.F., J. Huo, Y.W. Chen and F. Gao, 2001. Effect of NAA and BA on in vitro organogenesis of sweet potato. J. Sou. Chi. Nor. Univ., 26: 443-447.

Hahlbrock, K., J. Ebel, R. Ortmann, A. Sutter, E. Wellman, and H. Grisebach, 1971. Regulation of enzyme activities related to the biosynthesis of flavone glycosides in cell suspension cultures of parsley (*Petroselinum hortense*).Biochim. Biophys. Acta, 244: 7-15.

Hara, M., K. Oki, K. Hoshino and T. Kuboi, 2003. Enhancement of anthocyanin biosynthesis by sugar in radish (*Raphanus sativus*) hypocotyl. Plant Sci, 164: 259-265.

Hayase, F. and H. Kato, 1984. Antioxidative components of sweetpotato. J. Nutr.Sci. Vitaminol, 30: 37-46.

Hertog, M.G.L., E.J. Feskens, P.C.H. Hollman, M.B. Katan, D. Kromhout, 1993. Dietary antioxidant flavonoids and risk of coronary heart disease: the Zutphen Elderly Study. Lancet, 342: 1007-11.

Kamei, H., T. Kojima, M. Hasegawa, 1993. Cyto-static effect of flavonoid from petal on human malignant cells. Proc Jpn Cancer Assoc, 52: 584, (in Japanese).

Konczak, N., M. Terahara, M. Yoshimoto, M. Nakatani, M. Yoshinaga and O. Yamakawa, 2005. Regulation the composition of anthocyanins and phenolic acids in a sweetpotato cell culture towards production of polyphenolic complex with enhanced physiological activity, Trends Food Sci Technol., 16: 377-388.

Lamaison, J.L.C. and A. Carnet, 1990. Teneurs en principaux flavonoids des fleurs de *Crataegeus monogyna* Jacq et de *Crataegeus laevigata* (Poiret D. C) en fonction de la vegetation. Pharm. Acta. Helv., 65: 315-320.

Lawton, R., S. Winfield, H. Daniell, A.S. Bhagsari and S.K. Dhir, 2000. Expression of green-fluorescent protein gene in sweet potato tissues. Plant Molecular Biology Reporter, 18: 139a-139i.

Luczkiewicz, M. and W. Cisowski, 2001. Optimisation of the second phase of a two phase growth system for anthocyanin accumulation in callus cultures of *Rudbeckia hirta*, Plant Cell Tissue Organ Cult., 65: 57-68.

Makkar, H.P.S., K. Baker, H. Abel and E. Pawelzik, 1997. Nutrint contents, rumen protein degradability and antinutritional factors in some color-and white-flowering cultivars of *Vicia faba* beans. J.Sci. Food Agric., 75: 511-520.

Mano, H., F. Ogasawara, K. Sato, H. Higo, and Y. minobe, 2007. Isolation of a sweetpotato gene of anthocyanin biosíntesis in tuberous roots of purple-fleshed sweetpotato. Plant Physiology, 143: 1252-1268.

Martin, P., K. Gould, K. Markham, S. Lewthwaite and L. Ferguson, 2003. Enhanced coloration reveals high antioxidant potential in new sweetpotato cultivars.Journal of the science of food and agriculture, 83: 1076-1082.

Meyer, H.J. and J. Van Staden, 1995. The *in vitro* production of an anthocyanin from callus cultures of *Oxalis linearis*, Plant Cell, Tissue and Organ Culture, 40: 55-58.

Miura, H., Y. Kitamura, T. Ikenaga, K. Mizobe, T. Shimizu and M. Nakamura, 1998. Anthocyanin production of *Glehnia littoralis* callus culture, Phytochemistry, 48: 279-283.

Murashige, T. and F. Skoog, 1962. Arevised medium for rapid growth and bioassays in callus tissue cultures. Physiol. Plant, 15: 473-497.

Namiki, M., 1990. Antioxidants/antimutagens in food. Food Science and Nutrition, 29: 273-300.

Newell, C.A., J.A. Lowe, A. Merryweather, L.M. Rooke and W.D.O. Hamilton, 1995. Transformation of Sweet potato (*Ipomoea batatas (L.) Lam.*) with Agrobacterium tumefaciens. Plant Sci. J., 107: 215-227.

Odake, K., A. Hatanaka, T. Kajiwara, T. Moroi, K. Nishiyama, O. Yamakawa, N. Terahara and N. Yamaguchi, 1994. Evaluation method and breeding of purple sweetpotato "Yamagawa Murasaki" (*Ipomoea batatas*) for raw material of food colorants. Nippon Shokuhin Kogyo Gakkashi, 42: 287-293.

Odake, K., N. Terahara, N. Saito, K. Toki and T. Honda, 1992. Chemical structures of two anthocyanins from purple sweetpotato, Ipomoea batatas. Phytochemistry, 31(6): 2127-2130.

Oki, T., M. Masuda, S. Furuta, Y. Nishiba, N. Terahara and I. Suda, 2002. Involvement of anthocyanins and other phenolic compounds in radical-scavenging activity of purple-fleshed sweetpotato cultivars. J. Food Sci., 67: 1752-1756.

Osamu, Y. and Y. Makoto, 2002. Sweetpotato as food material with physiological functions. Acta Hort, 583: 179-185.

Otani, M. and T. Shimada, 1996. Efficient embryogenic callus formation in sweet potato (*Ipomoea batatas L. Lam.*). Breed. Sci., 46: 257-260.

Patil Bhimanagouda, S., G.K. Jayaprakasha, K.N. Chidambara Murthy, Vikram Amit, 2009. Bioactive compounds: historical perspectives, opportunities, and challenges. Journal of agricultural and food chemistry, 57(18): 8142-60.

Ranjeva, R., A.M. Boudet, H. Harada and G. Marigo, 1975. Phenolic Metabolism in *Petunia* tissues. 1. Characteristicresponses of enzymes involved in different steps of polyphenol synthesis to different hormonal influences. Biochem. Biophys. Acta, 399: 23-30.

Rolland, F., B. Moore and J. Sheen, 2002. Sugar sensing and signalling in plants. Plant Cell (suppl), 14: S185-S205.

Rout, G.R., S. Samantaray and P. Das, 2000. In vitro manipulation and propagation of medicinal plants Biotechnology Advances, 18(2): 91-120.

Sato, K., N. Mamoru and J. Ishi Shigeta, 1996. Culturing conditions affecting the production of anthocyanin in suspended cell cultures of strawberry, Plant Science, 113: 91-98.

Sheng, T., K. Joost, B. Leonie, K. Maarten and S. Smeekens, 2005. Sucrose-specific induction of anthocyanin biosynthesis in Arabidopsis requires the MYB75/PAP1 gene. Plant Physiology, 139: 1840-1852.

Smeekens, S., 2000. Sugar-induced signal transduction in plants. Annu Rev Plant Physiol Plant Mol Biol, 51: 49-81.

Solfanelli, C., P. Alessandra, L. Elena, A. Amedeo and P. Pierdomenico, 2006. Sucrose-specific induction of the anthocyanin biosynthetic pathway in Arabidopsis. Plant Physiology, 140: 637-646.

Terahara, I. Konczak-Islam., M. Nakatani, O. Yamakawa, Y. Goda and T. Honda, 2000. Anthocyanins in callus from purple storage root of *Ipomoea batatas* L, Phytochemistry, 54: 919-922.

Terahara, N., I. Konczak, H. Ono, M. Yoshimoto and O. Yamakawa, 2004. Characterization of acylated anthocyanins in callus induced from storage root of purple-fleshed sweetpotato, *Ipomoea batatas* L, J Biomed Biotechnol, 5: 279-286.

Thimann, K.V., Y.H. Edmondson and B.S. Radner, 1950. The biogensis of anthocyanin. 111.The role of sugars in anthocyanin formation. Arch. Biochem. Biophys, 34: 305-323.

Triqui, Z., A. Guédira, A. Chlyah, H. Chlyah, V. Souvannavong, R. Haïcour and D. Sihachakr, 2007. Effect of genotype, gelling agent, and auxin on the induction of somatic embryogenesis in sweet potato (Ipomoea batatas Lam.). C.R. Biologies.

Vanisree, M. and H.S. Tsay, 2004. Plant cell cultures-an alternative and efficient source for the production of biologically important secondary metabolites, Int J Appl Sci Eng., 2: 29-48.

Verpoorte, A. and J. Memelink, 2002. Biotechnology for the production of plant secondary metabolites, Phytochem Rev., 1: 13-25.

Wong, P.P., M. Zucker and L.L. Creasy, 1974. Induction of phenylalanine ammonialyase in strawberry leaf discs. Plant Physiol., 54: 659-665.

Woolfe, J.A., 1992. Sweetpotato: an untapped food resource. Cambridge, UK., Cambridge university press.

Yi Fang, C. and W. Xianzona, 2002. Antioxidant and Antiprofilactive Activities of Common Vegetables. Journal of Agricultural and Food Chemistry, pp: 381-385.

Yoshimoto, M., S. Okuno, M. Yamaguchi, O. Yamakawa, 2001. Antimutagenicity of deacylated anthocyanins in purple-fleshed sweetpotato. Biosci., Biotechnol. Biochem., 65: 1652-1655.