Journal of Applied Science and Agriculture, 9(6) May 2014, Pages: 2419-2424

AENSI Journals

Journal of Applied Science and Agriculture

ISSN 1816-9112

Journal home page: www.aensiweb.com/jasa/index.html

Comparative Phenolic Content and Antioxidant Activities of Four Wild Raspberries in Iran

[1]Aezam Rezaee Kivi, [2]Nasrin Sartipnia, [3]Maryam Babai Khalkhali

[1]*Department of Biology, Faculty of Science, Islamic Azad University, Khalkhal, Iran.*
[2]*Department of Biology, Faculty of Science, Islamic Azad University, Eslamshahr, Iran.*
[3]*Department of Chemistry, Faculty of Science, Islamic Azad University, Khalkhal, Iran.*

ARTICLE INFO

Article history:
Received 20 March 2014
Received in revised form 20 April 2014
Accepted 15 May 2014
Available online 1 June 2014

Key words:
Antioxidant capacity, raspberry, total phenolics, correlation

ABSTRACT

Four wild raspberries (R. hyrcanus, R.anatolicus, R.hirtus, R. caesius) from Iran were analyzed comparatively in order to determine the total phenolics, anthocyanins, flavonoids, vitamin C content and measuring the antioxidant activity using two different assayes. Ferric reducing ability (FRAP), and 2, 2-diphenylpicrylhydrazil radical scavenging capacity (DPPH). Total phenolic content ranged from 379.67-681.33mg GAE/100g fresh weight (FW). The highest total anthocyanin was observed in R. hirtus. The range of ascorbic acid content of species was 18.22-34.23 mg/100g FW. R. hyrcanus had the highest total flavonoid (297.33 mg QE/100gFW). There are linear relationships between the antioxidant capacities with total phenols and total flavonoid. But, No statistically significant correlation was observed between antioxidant activity and total anthocyanins. The present study demonstrates the potential of certain raspberry species, notably R. hyrcanus and R.hirtus, for improvement of nutritional value through germplasm enhancement programs.

To Cite This Article: Aezam Rezaee Kivi, Nasrin Sartipnia, Maryam Babai Khalkhali., Comparative Phenolic Content and Antioxidant Activities of Four Wild Raspberries in Iran. ***J. Appl. Sci. & Agric.,*** *9(6): 2419-2424, 2014*

INTRODUACTION

Berries contain high concentration in bioactive compounds such as polyphenols, including anthocyanins, phenolic acids, tannins, carotenoids, vitamin A, C, E, folic acid and minerals such as calcium, selenium and zinc (Kresty *et al*., 2001; Pineli *et al*., 2011). Among them, raspberries became well known and contain high level of anthocyanin and phenolic compounds with high invitro antioxidant capacities compared with other berries (Poiana *et al*., 2010). The consumption of horticultural products plays an important role in the maintenance of health and in disease prevention, such as inflammation, cardiovascular disease, cancer and ageing-related disorders (Rimm *et al*., 1996; Terry *et al*., 2001). It has been already demonstrated that a wide diversity of phytochemical levels and antioxidant capacities exist within and across genera of small fruit (Moyer *et al*., 2002). Fruits of raspberry are characterized by a high nutrition value (Poiana *et al*., 2010). Fruits and vegetables are a good source of natural antioxidants, which containing many different radical scavenger components that provide protection against harmful-free radicals and therefore associated with lower incidence and mortality rates of cancer and heart diseases in addition to a number of other health benefits (shui and leong., 2006). Among natural compounds, phenolics and in particular flavonoids were found to be an important part of human diet and are considered as active principles in many medicinal plants (Cooper-Driver., 2001). In addition, increasing recent interest in nutraceuticals and functional foods has led plant breeders to initiate selection of crops with higher than normal phenolic antioxidant contents, such as raspberries (Shiow *et al*., 2000), plums and peaches (Cavallos_Casajs *et al*., 2006), sea buckthorns (Ercisli *et al*., 2007), and strawberries and apples (Scalzo *et al*., 2005). All these programs aim to set the base line for establishing breeding efforts, with the intention of adding value to fruits, with respect to the level and diversity of health benefits that such crops could impart. In recent years increasing attention has been paid by consumers to the lesser known fruits such as raspberry, honeysuckle, hardy kiwifruit, elderberry, sea buckthorn, bilberry, strawberry, etc., which have unusual flavors and qualities, and many of which are rich with antioxidants and anthocyanins (Ercisli *et al*., 2007). Therefore, detailed information about the health-promoting components of more raspberry species could lead to a better understanding and an increased consumption of this fruits, including its use in functional foods and as ingredients in pharmaceuticals, nutraceuticals, and medicine.

Corresponding Author: Aezam Rezaee Kivi**,** Department of Biology, Faculty of Science, Islamic Azad University, Khalkhal, Iran.
E-mail: Azam_rezaee_k@yahoo.com

Journal of Applied Science and Agriculture, 9(6) May 2014, Pages: 2419-2424

Raspberry fruit are widely grown in some regions of Iran. Despite its wide usage in this country, there have been no standardised studies on the fruit as the case is for other fruit species. The objective of this study was to determine antioxidant capacity, total anthocyanins, total phenolic, ascorbic acid, and total flavonoids of a number of selected raspberry species in Iran. We propose, in this study, that selected raspberry species, rich in antioxidant, phenolic and anthocyanin compounds will yield fruits with enhanced functional properties, such as antioxidant, colorant and antimicrobial properties.

MATERIAL AND METHODS

Collection and preparation of raspberry fruits samples:

Iranian raspberries that were evaluated in this study (*R. hyrcanus, R.anatolicus, R.hirtus, R. caesius*) were collected from the north (Heiran, Gilan province) and northwest (Ardebil province) regions of Iran.

Approximately 500 g of ripe raspberry fruits were harvested manually in July 2012. The fruits were sorted according to uniformity of shape and color and then immediately transported to lab and free zed with liquid nitrogen and kept at -80 ºC, until needed for analysis.

Extraction and measurement of total ascorbic acid:

Total ascorbic acid content was determined using the dinitrophenylhydrazine (DNPH) method (Terada *et al*., 1978). Five grams of homogenized fruit tissue was added to 100 ml of a mixture of 6% metaphosphoric acid in 2 $moll^{-1}$ acetic acid. The mixture was centrifuged at 17,000 × g for 15 min at 4ºC and supernatant was filtered through Whatman filter paper. One milliliter aliquot of the supernatant was mixed with 0.05 ml of 0.2% 2, 6-dichlorophenolindolphenol (DCIP) and the solution was incubated at room temperature for 1 h. After that, 1 ml of 2% thiourea in 5% metaphosphoric acid and 0.5 ml of 2% DNPH in 4.5 $moll^{-1}$ sulfuric acid were added to the solution, and then incubated at 60ºC for 3 h. The reaction was stopped by placing the tubes in an ice bath and slowly adding 2.5 ml of cold 90% sulfuric acid. Total ascorbic acid was measured by absorbance at 540 nm using a standard curve. The concentrations were expressed as ascorbic acid on a fresh weight basis, mg per 100 g of fruit.

Extraction and measurement of total anthocyanins:

Some of frozen tissue was ground to a fine powder under liquid nitrogen by cold mortar and pestle and 1g of the resultant powder was added to 10 ml of methanol containing HCl (1%, v/v) and held at 0ºC for 10 min (Cordenunsi *et al*., 2003). The slurry was centrifuged at 17,000× g for 15 min at 4 ºC and then the supernatant was used. Total anthocyanins content was measured with the pH differential absorbance method, as described by Cheng and Breen (1991). Briefly, absorbance of the extracts were measured at 510 and 700 nm in buffers at pH 1.0 (hydrochloric acid-potassium chloride, 0.2 M) and 4.5 (acetate acid- sodium acetate, 1 M). Anthocyanin content was calculated using a molar extinction coefficient of 29,600 (cyaniding-3- glucoside).

Absorbance (A) = $(A_{510} - A_{700})_{pH1.0} - (A_{510} - A_{700})_{pH4.5}$

Results were expressed as mg cyaniding 3-glucoside equivalent per 100g of fresh weight.

Extraction and measurement of total phenolic content:

Total phenol in the methanol extracts was determined with Folin-Ciocalteu reagent by the method of Slinkard and Singleton (1972). Gallic acid (GAE) was used as a standard and results were expressed as mg Gallic acid equivalents per 100 g fresh weight.

Extraction and measurement of total flavonoid:

Some of frozen tissue was ground to a fine powder under liquid nitrogen by cold mortar and pestle. One gram of the resultant powder was added to 10 ml of methanol containing HCl (1%, v/v) and held at room temperature for 24 h (Cordenunsi *et al*., 2003). The slurry was centrifuged at 4000× g for 15 min at 4ºC, and the supernatant was used. The total flavonoid contents were determined by a colorimetric assay (Yanping *et al*., 2004). One milliliter aliquot of appropriately diluted sample was added to a 15 ml tube containing 4ml of deionized water. Then 0.3 ml of 5% $NaNO_2$ was added to this mixture, which was allowed to stand for 5 min at room temperature, and 0.6 ml of 10% $AlCl_3.6H_2O$ was added. The mixture was allowed to stand for 6 min at room temperature, and 2 ml of 1 mol l^{-1} NaOH was added, and the total was made up to 10 ml with deionized water. The absorbance of the solution was measured immediately at 510 nm. Quercetin was used as a standard compound for the quantification of total flavonoid.

Determination of the antioxidant capacity by DPPH radical scavenging method :

The antioxidant capacity of the raspberry fruits were evaluated by free radical 2, 2-dipheynl-1-picrylhydrazyl (DPPH) methods. For the determination of free radical scavenging capacity, raspberry samples were extracted with methanol. Then, they were centrifuged (Sigma 3K30, Germany) at 15,000× g for 10 min.

Journal of Applied Science and Agriculture, 9(6) May 2014, Pages: 2419-2424

The supernatants were concentrated under reduced pressure at 40º C. The dried extracts were dissolved in methanol. Free radical scavenging activity was measured according to the principle of Nakajima *et al*. (2004) with some modifications reported by Chiou *et al*. (2007). Fifty microliters of the diluted extracts (concentrations 2-20 mg ml^{-1}) were added to 1 ml 0f 6× 10^{-5} mol l^{-1} DPPH (free radical, 95%, sigma-Aldrich Chemie GmbH, Steinheim, Germany) in methanol. The mixture was shacked and left at room temperature for 30 min; the absorbance was measured spectrophotometrically at 515 nm. Methanol was used as an experimental control. The percent of reduction of DPPH was calculated according to the following equation

$$\%\ \text{inhibition of DPPH} = \frac{\text{Abs control - Abs sampele}}{\text{Abs control}} \times 100$$

Determination of the antioxidant capacity by FRAP assay:

The FRAP assay (Benzie and Strain 1999) was conducted using three aqueous stock solutions containing 0.1 $moll^{-1}$acetate buffer (pH 3.6), 10 $mmoll^{-1}$ TPTZ [2, 4, 6-tris (2-pyridyl)-1, 3, 5-triazine] acidified with concentrated hydrochloric acid, and 20 $mmoll^{-1}$ferric chloride. These solutions were prepared and stored in the dark under refrigeration. Stock solutions were combined (10:1:1, v/v/v) to form the FRAP reagent just prior to analysis. For each assay laboratory duplicate, 2.97 ml of FRAP reagent and 30µl of sample extract were mixed. After 10 min, the absorbance of the reaction mixture was determined at 593 nm on a spectrophotometer. The result was compared with the standard curve obtained by using different concentrations of $FeSO_4$. $7H_2O$.

Statistical analysis:

Experiment was a completely randomized design with three replications and four points. In order to determine the significant differences between values, analysis of variance (ANOVA) and Duncan‘s multiple range tests were performed. Significance of difference was defined at the 5% level ($p<0.05$).

Results:

The comparative data about total phenolics, flavonoid, anthocyanins and vitamin C in species raspberries are presented in Tab. 1. The differences in total phenolic content (TPC) among raspberry species was statically significant ($p<0.05$). The TPC of raspberry species was in the range of 379.67-681.33 mg gallic acid per 100 g FW basis. Among all the species analyzed, the *R. hyrcanus* revealed the highest TPC at 681.33 gallic acid equivalents/100 g FW followed by *R. hirtus* (529 mg gallic acid per 100 g FW).

Table 1: Total anthocyanin(TA), total phenolic (TP), total flavonoids content(TF) and ascorbic acid (AA) 0f raspberry fruits.

species	DPPH (%)	FRAP (µmolFe^{+2})	TA (mg/100g)	TP (mgGAE/100g)	TF (mgQE/100g)	AA (mg/100g)	TF/TP
R. hyrcanus	73.46a	40.80a	35.19c	681.33a	297.33a	22.56c	2.29
R. hirtus	67.23b	34.58b	48.11a	529b	259.67b	34.23a	2.03
R. anatolicus	60.18c	30.49c	42.68b	423.67c	207c	18.22d	2.04
R. caesius	52.34d	26.20d	46.75a	379.67d	165d	32.13b	2.30

Values in the same column with different lower-case letters are significantly different at $p<0$.

The total flavonoids content ranged from 165-297.33 mg Quercetin per 100 g FW basis. Results indicated that the difference in flavonoid content among raspberry species were statiscally significant ($p<0.05$). Ratio of total flavonoid / phenolics in the raspberry fruits are presented in Tab. 1. The highest ratio total flavonoids / phenolics were observed in *R. caesius* at 2.30. The low ratio in some species is probably a result of the rich spectrum of phenolic acids (Marinova *et al*., 2005).

The content of total anthocyanins of raspberry species were in the ranged of 35.19-48.11mg, expressed as cyanidin3-glucoside equivalents per 100 g FW basis (Table 1).The highest total anthocyanins content was found in *R. hirtus* (48.11mg/100gFW). There were significant differences ($p<0.05$) in anthocyanins content between *R. hirtus*, *R. anatolicus* and *R. hyrcanus*. However, significant differences in the total anthocyanin content were not observed between *R. hirtus* and *R. caesius* ($p<0.05$). A wide variation was found among raspberry species in terms of ascorbic acid content, ranging from 18.22 to 34.23 mg /100 g. The *R. hirtus* had the highest ascorbic acid content in its fruits (34.23 mg /100g). Ascorbic acid content of raspberry was previously reported as being between 16.8 and 32.4 mg /100 g (Deighton *et al*., 2000; Mullen *et al*., 2002; Pantelidis *et al*., 2007).

The antioxidant activity results using DPPH method in raspberry species are shown in Tab. 1. A statistical significant difference ($p<0.05$) was found among species. All raspberry species showed high antioxidant activity. The highest antioxidant activity was observed *R. hyrcanus* (73.46%). The FRAP assay showed greater variability between species (Table 1). The raspberry species had FRAP values in the range 26.20 -40.80 µmol Fe^{+2}/ g FW. *R. hyrcanus* species was significantly more active than other species ($P<0.05$).

Journal of Applied Science and Agriculture, 9(6) May 2014, Pages: 2419-2424

Table 2: Pearson's correlation coefficients for quantitative determinations in raspberry species.

Variables	DPPH	FRAP	TA	TP	TF	AA
DPPH	1	0.98**	0.66*	0.95**	0.99**	0.21ns
FRAP	0.98**	1	0.74**	0.98**	0.98**	0.25ns
TA	0.66*	0.74**	1	0.75**	0.64*	0.71**
TP	0.95**	0.98**	0.75**	1	0.96**	0.17ns
TF	0.99**	0.98**	0.64*	0.96**	1	0.16ns
AA	0.21ns	0.25ns	0.71**	0.17ns	0.16ns	1

[a]95% confidence interval, ns, no significant; *, significant at $p<0.05$; **, significant at $p<0.001$.

Discussion:

The TPC data's obtained are comparable to previous findings which reported values between 1280-2116 mg gallic acid equivalents per g DW basis (Pantelidis *et al*., 2007) and 278.6-496.1 mg gallic acid equivalents per 100 g FW basis (Rumune *et al*., 2011). Our TPC results were higher than those reported elsewhere. The phenolic content and composition of fruits depend on environmental factors as well as post-harvest processing conditions (Benvenuti *et al*., 2004; Kadir *et al*., 2009).

The fruits of raspberry revealed the presences of considerable amounts of flavonoids. Thus, results of the present study supported the antioxidant and nutraceutical potential of this plant species. The differences in the composition of the fruits could be due to the growing conditions, such as soil, geographical and environmental conditions during the fruit development, degree of maturity at harvest and genetic differences (Agata *et al*., 2009). Considerable data suggests that higher content of total phenolics, flavonoids, and anthocyanins in raspberry fruits contribute to their higher antioxidant activity (Liu *et al*., 2002; Wang and Lin., 2000). Although ascorbic acid has been considered as important antioxidant, it accounts only for 6% of the total antioxidant activity in raspberry (Kalt *et al*., 1999). Measuring the antioxidant capacity in order to evaluate the potential health benefits of breeding material or various agronomic factors can be a tedious task. Thus, the determination of indirect parameters, such as the content of the total phenolics, flavonoids or anthocyanins, for describing the potential health benefits, may be a more appropriate objective.

Although anthocyanins seem to contribute to the antioxidant capacity (Mullen *et al*., 2002; Wang and Lin., 2000) of raspberry fruit, their other health benefits seem unclear. Mullen *et al*. (2002) suggests that raspberry anthocyanins provide only little cardioprotective vasodilatory effects, and their role in the inhibition of tumour cells may also be less important than that of other phytochemicals in raspberry (Liu *et al*., 2002).

In this study, high level of antioxidant activity obtained for *R. hyrcanus*, by two methods used, could be due to its high level of total phenolics and total flavonoids content.

A correlation analysis was done among total phenolics, flavonoids, anthocyanins , ascorbic acid and the antioxidant activity values obtained (Table 2). The total phenolic content and flavonoids exhibit a significant correlation ($p<0.01$). These results are in agreement with other studies (Wang and Lin., 2000; Pantelidis *et al*., 2007; Bunea *et al*., 2011). The high correlation between the FRAP and total phenolics content and flavonoids can be attributed to the fact that both assayes rely on the same reaction mechanism. The highest pearson's coefficient between was obtained when it has been compared DPPH and flavonoids (0.995). In this study, no statistically significant correlation was observed between antioxidant activity and ascorbic acid (0.21, 0.25). These finding are in keeping with previous observation. Sara *et al*. (2008), Deighton *et al*. (2000) suggested that associations between the antioxidant properties and the proportion of phenols present as ascorbic acid are generally not very evident in raspberry.

As a conclusion, this investigation clearly shows the potential value of raspberry germplasm. Raspberry fruits are a significant source of phenolic compounds, anthocyanins, total flavonoids and ascorbic acids. Antioxidant activity was high in fruits and varied greatly among the species. Therefore raspberry could be considered a good source of natural antioxidants. They can potentially be used in food and nutraceutical supplement formulations as well. Moreover, since commercial raspberry cultivars do not exist, these results could be important to use these species as breeding materials in future traditional breeding or advanced biotechnology studies. In addition, a wide range of agronomic characteristics, such as high yield and pest and disease resistance of these selected species could be incorporated into an improved raspberry cultivar.

REFERENCES

Agata, M.P., C. Fabian, B. Alessandra, 2009. Quali-quantitative analysis of flavonoids of Cornus mas L. (Cornaceae) fruits. Food Chemistry, 115: 450-455.

Benvenuti, S., F. Pellati, M. Melegari, D. Bertelli, 2004. Polyphenols, anthocyanins, ascorbic acud and radical scavenging activity of Rubus, Ribes, and Aronia. Food Science, 69(3): 164-169.

Benzie, I.F., J.J. Strain, 1999. Ferric reducing/antioxidant power assay. Direct measure of total antioxidant activity of biological fluids and modified version for simultaneous measurement of total antioxidant power and ascorbic acid concentration. Methods Enzymol, 299: 15-27.

Bunea, A., O.R. Dumitrita, M. Adela. Z. Pintea, C.I. Sconta, C.S. Bunea, 2011. Comparative polyphenolic content and antioxidant activities of some wild and cultivated blueberries from Romania. Notulae Botanicac Horti Agrobotanici, 39(2): 70-76.

Cavallos-Casals, B.A., D. Byrne, W.R. Okie, L. Cisneros-Zevallos, 2006. Selecting new peach and plum genotypes rich in phenolic compounds and enhanced functional properties. Food Chemistry, 102: 516-522.

Cheng, G.W., P.J. Breen, 1997. Activity of phenylalanine ammonia-lyase (PAL) and concentrations of anthocyanins and phenolics in developing strawberry fruit. Journal of the American Society for Horticultural Science, 116(5): 865-869.

Chiou, A., V.T. Karathanose, A. Mylona, F.N. Salta, F. Preventi, N.K. Andriopoulos, 2007. Currants (Vitis vinifera L.) content of simple phenolics and antioxidant activity. Food Chemistry, 102: 516-522.

Cordenunsi, B.R., J.R.O. Nascimento, F.M. Lajolo, 2003. Physico-chemical changes related to quality of five strawberry fruit cultivars during cool storage. Food Chemistry, 83: 167-173.

Cooper-Driver, G.A., 2001. Contribution of Jeffrey Harborne and co-workers to the study of anthocyanins. Phytochemistry, 56: 229-236.

Deighton, N., R. Brennan, C. Finn, H.V. Davies, 2000. Antioxidant properties of domesticated and wild Rubus species. Journal of Science and Food Agriculthure, 80: 1307-1313.

Ercisli, S., E. Orhan, O. Ozdemir, M. Sengul, 2007. The genotypic effects in the chemical composition and antioxidant activity of sea buckthorn (Hippophae rhamnoides L.) berries grown in Turkey. Scientia Horticulthure Amesterdam, 115(1): 27-33.

Kadir, U.Y., E. Sezai, Z. Yasar, S. Memmmune, Y.K. Ebru, 2009. Preliminary characterization of cornelian cherry (Conus mas L.) genotypes for their physic-chemical properties. Food Chemistry, 114: 408-412.

Kalt, W., C.F. Forney, A. Martin, R.L. Prior, 1999. Antioxidant capacity, vitamin C, phenolics, and anthocyanins after fresh storage of small fruits. Journal of Agricultural and Food Chemistry, 47: 4638-4644.

Kresty, L.A., M.A. Morse C. Morgan, P.S. Carlton, J. Lu, A. Gupta, M. Blackwood, G.D. Stoner, 2001. Chemoprevention of esophageal tumorigenesis by dietary administration of lyophilized black raspberries. The Journal of Cancer Research, 6: 6112-6119.

Liu, M., X.Q. Li, C. Weber, C.Y. Lee, J. Brown, R.H. Liu, 2002. Antioxidant and antiproliferative activities of raspberries. Journal of Agriculthural and Food Chemistry, 50: 2926-2930.

Marinova, D., F. Ribarova, M. Atanassova, 2005. Total phenolics and total flavonoids in Bulgarian fruits and vegetables. Journal of the University of Chemical Technology and Metallurgy, 40(3): 255-260.

Moyer, R.A., K.E. Hummer, C.E. Finn, B. Frei, R.E. Wrolstad, 2002. Anthocyanins, phenolics, and antioxidant capacity in diverse small fruits: Vaccinium, Rubus, and Ribes. Journal of Agriculthure and Food Chemistry, 50(3): 519-525.

Mullen, W., A.J. Stewart, M.E.J. Lean, P. Gardner, G.G. Duthie, A. Crozier, 2002. Effect of freezing and storage on the on the phenolics, ellagitanins, flavonoids, and antioxidant capacity of red raspberries. Agriculture and Food Chemistry, 50: 5197-5201.

Pantelidisa, G.E., M. Vasilakisa, G.A. Manganaris, G. Diamantaidisa, 2007. Antioxidant capacity, Phenol, anthocyanin and ascorbic acid contents in raspberries, black berries, red currants, goose berries and cornelian cherries. Food Chemistry, 102: 777-783.

Pineli, L.L.O., C.L. Moretti, M.S. Santos, A.B. Campos, A.V. Brasileiro, A.C. Cordova, M.D. Chiarello, 2011. Antioxidants and other chemical and physical characteristics of two strawberry cultivar at different ripeness stages. Journal of Food Composition and Analysis, 24(1): 11-16.

Poiana, M.A.,Moigradean, D., Raba, M.L., Popa, M, 2010. The effect of long-term frozen storage on the nutraceutical compounds, antioxidant properties and color indices of different kinds of berries. Journal of Food Agriculture &Environment, 8(1): 54-58.

Rimm, E.B., A. Aschiero, E. Giovannucci, D. Spiegelman, M.J. Stampfer, W.C. Willet, 1996. Vegetables, fruit, and cereal fiber intake and risk of coronary heart disease among men. The Journal of the American Medical Association, 275(6): 447-451.

Rumune, B., V. Pranas, R.V. Petras, 2011. Variation of total phenolics, anthocyanins, ellagic acid and radical scavenging capacity in various raspberry (Rubus spp.) cultivars. Food Chemistry, 137(11): 456-462.

Sara, T., M. Burno, C. Franco, B. Stefano, B. Jules, D. Chris, C. Ezra, B. Arnauda, B. Maurizio, 2008. Antioxidant phenol compounds, and nutritional quality of different strawberry genotypes. Agriculture and Food Chemistry, 56: 696-704.

Scalzo, J., A. Politi, N. Pellegrini, B. Mezzetti, M. Battino, 2005. Plant genotype affects total antioxidant capacity and phenol contents in fruit. Nutrition, 21: 207-213.

Shiow, Y., Wang, R.B. James, 2007. Free radical scavenging capacity and antioxidant enzyme activity in deerberry (Vaccinium stamineum L.). Food Science and Technology, 40(8): 1352-1361.

Shui, G. and L.P. Leong, 2006. Residue from star fruit as valuable source for functional food ingredients and antioxidant nutraceuticals. Food Chemistry, 97(2): 277–284.

Slinkard, K., V.L. Singleton, 1977. Total phenol analyses: automation and comparison with manual methods. American Journal of Enol Viticult, 28: 49-55.

Terada, M., Y. Watanabe, M. Kunitoma, E. Hayashi, 1978. Differential rapid analysis of ascorbic acid and ascorbic acid 2-sulfate by dinitrophenilhydrazine method. Annal Biochemistry, 84: 604-608.

Terry, R.D., E. Masliah, D.P. Salmon, N. Butters, R. Deteresa, R. Hill, L.A. Hansen, R. Katzman, 1991. Physical basis of congnitive alteration in alzheimer's disease: synapse loss is the major correlate of cognitive impairment. Annals of Neurology, 30(4): 572-580.

Wang, S and H. Lin, 2000. Antioxidant activity in fruits and leaves of blackberry, raspberry and strawberry varies with cultivar and developmental stage. Journal of Agricultural Food Chemistry, 48 (2): 140-146.

Yanping, Z., A. Yannua, W. Dongzhi, 2004. Antioxidant activity of a flavonoid rich extract of Hypericum perforatum L. in vitro. Journal of Agriculture and Food Chemistry, 52: 5032-5039.