



ANTIOXIDANT POTENTIAL OF TAMA - AN ETHNIC FOOD OF SIKKIM

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ABSTRACT

One ethnic food of Sikkim is tama. People of ethnic group in Sikkim use tama as food for diabetics, pregnant & lactating mother. They believe that tama controls hypertension and prevents inflammation and gastrointestinal disorders. These health benefits may be related with antioxidant activity. Objective of the present study was, therefore, to evaluate antioxidant potential of tama. *In vitro* antioxidant activity of tama was measured by superoxide anion generation with the help of xanthine-xanthine oxidase assay, linoleic acid peroxidation assay and by DPPH photometric assay as well as by nitric oxide radical scavenging assay, hydroxy radical scavenging assay and by ABTS radical scavenging assay. At the same time total antioxidant capacity of tama was determined by phosphomolybdenum method and by ferric reducing power (FRP) method. Antioxidant compounds like carotenoid, ascorbic acid, flavonoids and polyphenol were also estimated in tama. Results showed that tama has strong antioxidant activity which was due to high content of polyphenol, ascorbic acid and flavonoids. Health benefits of tama may, therefore, be explained through its antioxidant potential.

KEYWORDS: Tama, Anti oxidant potential; Ascorbic acid; Polyphenol; Flavonoid; Carotenoids.

1. INTRODUCTION

Ethnic food has many definitions. Church et.al, (2006) define ethnic food as foods that originate from other countries with a different food culture.^[1] Verbeke et.al (2005) define ethnic food as the expression of food in terms of attitude, values, behavior and beliefs of a culture.^[2] The Swedish Chamber (2010) defines ethnic food as food stemming from customs and traditions.^[3]

In general, ethnic foods are prepared by the ethnic group utilizing knowledge of local ingredients of plants/herbs and/or animal sources through their heritage and culture. Ethnic foods are available in almost all countries of world. Few examples are, Maori food of New Zealand, Masai food of Kenya, Pizza of Italy, Hindu food of India etc.^[4,5] Ethnic foods possess some important health-benefits compounds like bio-nutrients, probiotics, antimicrobial, antioxidant etc.^[6]

Sikkim, the land of mystical splendour, is a landlocked state of India. Several ethnic foods like falki, suzom, thukpa, phaparko roti, nakima, nya cham, piranlu, phulaurah, chatamari, ponguzom gyuma, khapjay, khoreng, kegu, chhwelaa, wachipa, kachila, ponguzom, kasalok, bauwa, siltimbur, nakima, nya cham, piranlu, bhatmas ko achar, dheroh, nya cham, piranlu, kodoko roti, kwanti, chiura, chambray, chakho,

dheroh, gyathuk, ghario, koirala, ghartarul, philingo, ken-tsong, lakhamari, lauwa, lapsi, moongarbuk, phashyagyari, phando, phituk, etc. are available in Sikkim.^[7]

Tama is one such ethnic food of Sikkim. Tama is a non-fermented bamboo shoot product. Some varieties of bamboo shoots commonly grown in the Sikkim Himalayas are *Dendrocalamus hamiltonii*, *Dendrocalamus sikkimensis* and *Bambusa tulda* locally known as 'choya bans', 'bhalu bans' and 'karati bans' respectively are edible when young. These young bamboo shoots are commonly sold in the local markets during the months of June to September. Bamboo shoots are collected, defoliated and boiled in water with turmeric powder for 10-15 min to remove bitter taste of bamboo. Now the bamboo shoots are ready for consumption. These bamboo shoots along with methi, turmeric, green chilli etc. are the major ingredients for preparation of tama curry which is prepared by frying methi seeds in oil, adding bamboo shoots, turmeric powder, sliced round chili and salt and then stirring followed by cooking for 3 minutes. Tama curry is ready to serve with cooked rice.^[7]

Ethnic groups of Sikkim use tama as food for diabetics, pregnant & lactating mother. It is their belief that tama could control hypertension as well as prevent inflammation and gastrointestinal disorders. Considering these health benefits it was thought worthwhile to evaluate antioxidant potential of tama.



Figure 1: Tama.

2.2 Test sample

Using lyophilizer samples of tama were freeze dried and then made powder. The powder was extracted with water for 10 minutes at 37°C. 100 µg / ml was chosen as test dose.^[8]

2.3 Studies of Antioxidant potential of tama

Antioxidant potential of tama was checked by doing antioxidant assays and estimation of antioxidant compounds.

2.3.1 Antioxidant assays

Antioxidant assays were done by noting superoxide anion generation through the followings.

2.3.1.1 Xanthine-/xanthine oxidase assay – by the method of Chang *et al.*^[9]

2.3.1.2 Linoleic acid peroxidation assay- by the method of Chang *et al.*^[10]

2.3.1.3 DPPH photometric assay – by the method of Mensor *et al.*^[11]

Antioxidant potential of tama was also checked by,

2.3.1.4 ABTS radical scavenging assay – by the method of Shirwaikar *et al.*^[12]

2.3.1.5 Hydroxy radical scavenging assay – by the method of Halliwell *et al.*^[13]

2.3.1.6 Nitric Oxide radical scavenging assay – by the method of Panda *et al.*^[14]

Total antioxidant activity of tama was measured by,

2.3.1.7 Phospho molybdenum method as developed by Prieto *et al.*^[15]

2. METHODOLOGY

2.1 Collection of samples

39 Samples of tama were collected from different houses of Saramsa, Nanduk, Barbing, Patuk, Sajong and Shotek villages of east Sikkim.

2.3.1.8 Ferric reducing power (FRP) method as suggested by Oyaizu.^[16]

2.3.2 Estimation of Antioxidant compounds

Antioxidant compounds present in tama were estimated for,

2.3.2.1 Flavonoids content

Flavonoids content of tama was determined using Aluminum chloride colorimetric method.^[17]

2.3.2.2 Polyphenol content

Polyphenol content of tama was measured by Folin Ciocalteu reagent.^[18]

2.3.2.3 Ascorbic acid content

Ascorbic acid content of tama was determined by the method of Cakmak and Marschner.^[19]

2.3.2.4 Carotenoids content

Carotenoids content of tama was measured by the method of Jensen.^[20]

2.4 Chemicals and reagents

Chemicals and reagents required for the study were of analytical grade and of highest purity.

They were purchased from Loba Chem., Himedia Lab, India, Merck, Germany, Sigma-Aldrich and from St. Louis, MO, USA.

3. RESULTS

In vitro antioxidant activity of tama through superoxide anion generation by linoleic acid peroxidation assay, xanthine-xanthine oxidase assay and by DPPH photometric assay is presented in Figure – 2. Percent

inhibitions of xanthine oxidase, linolenic acid peroxidation and DPPH by tama were found 91, 86 and 89 respectively. In case of standard antioxidant quercetin the same values came as 100, 95 and 98 respectively.

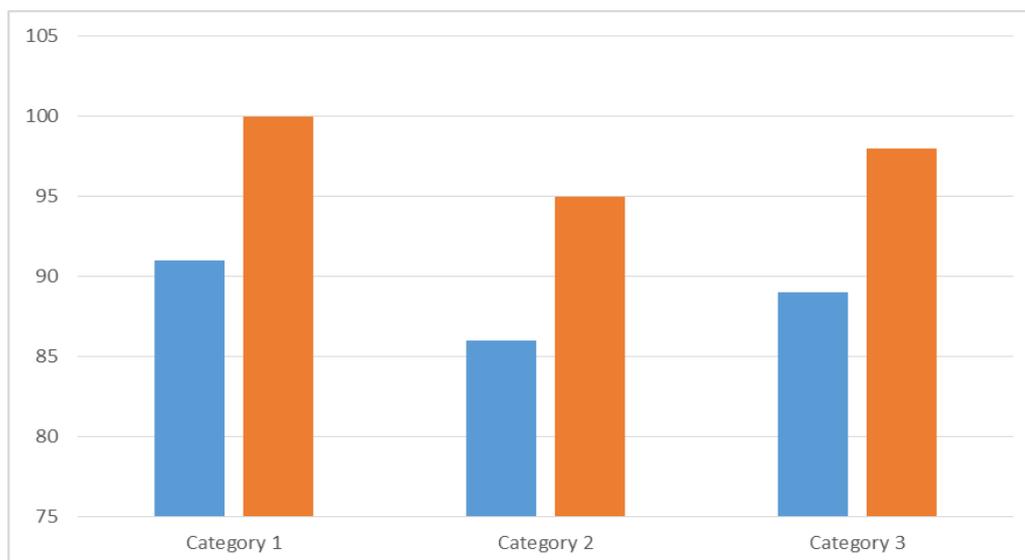


Figure 2: Showing in vitro antioxidant activity of tama through superoxide anion generation by xanthine-xanthine oxidase assay, linoleic acid peroxidation assay and by DPPH photometric assay.

Category 1: Xanthine oxidase Category 2: Linoleic acid Category 3: DPPH

Results, in terms of percent inhibition, were the mean of triplicate experiments.

Quercetin was used as standard antioxidant

■ Tama ■ Quercetin

Figure – 3 showed results of in vitro antioxidant activity of tama through ABTS scavenging assay, hydroxy radical scavenging assay and nitric oxide radical scavenging assay. Tama gave.

IC₅₀ values for ABTS, hydroxyl radical and nitric oxide radical as 30 $\mu\text{g} / \text{ml}$, 2.2 $\mu\text{g} / \text{ml}$ and 60 $\mu\text{g} / \text{ml}$ respectively but when ascorbic acid was used as standard antioxidant, the said values came 16 $\mu\text{g} / \text{ml}$, 2.1 $\mu\text{g} / \text{ml}$ and 52 $\mu\text{g} / \text{ml}$ respectively.

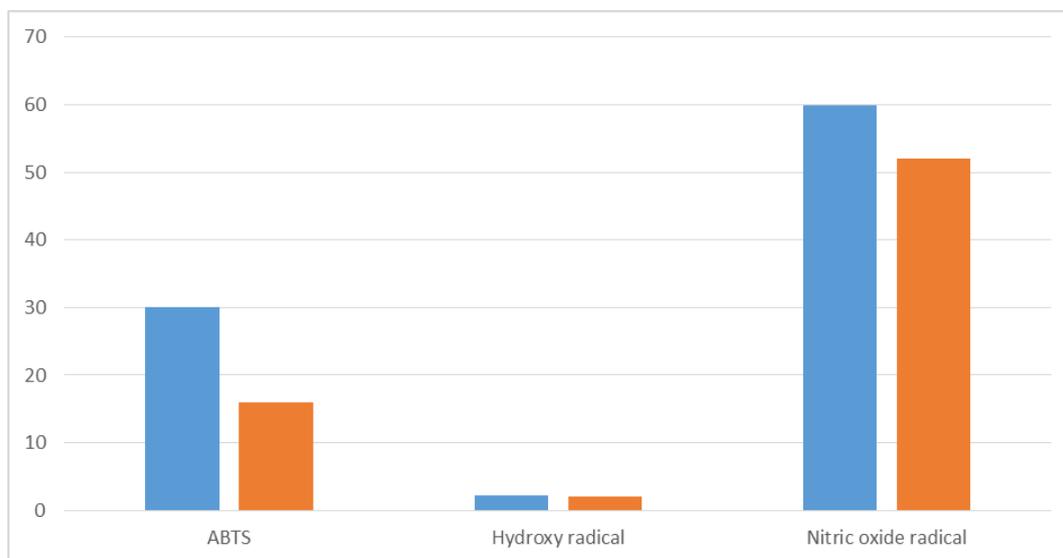


Figure 3: Showing in vitro antioxidant activity of tama through ABTS scavenging assay, hydroxy radical scavenging assay and nitric oxide radical scavenging assay.

Results, in terms of $\mu\text{g}/\text{ml}$, were the mean of triplicate experiments.

Ascorbic acid was used as standard antioxidant

■ Tama ■ Ascorbic acid

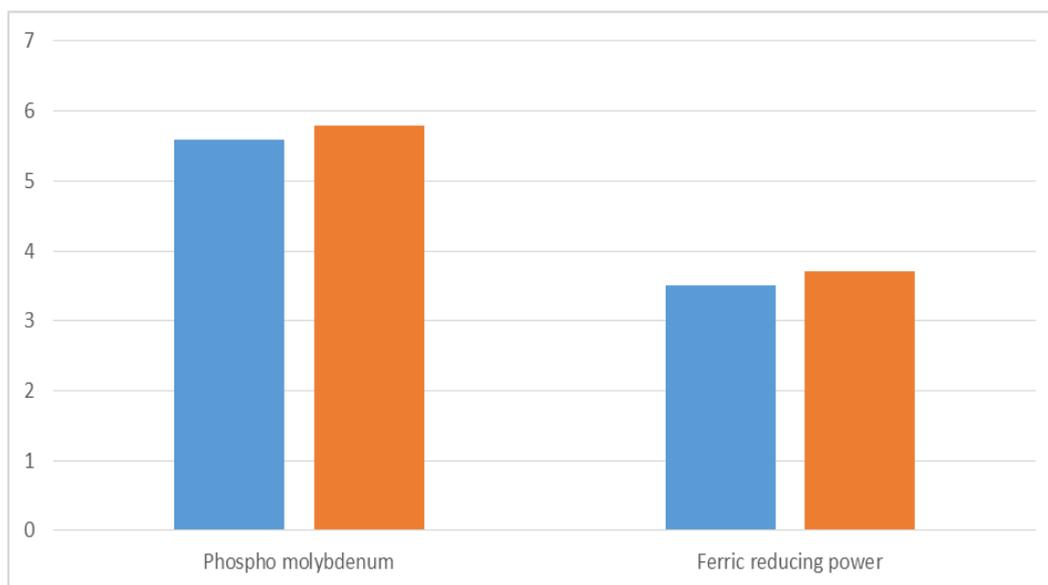


Figure 4: Showing result of total antioxidant activity of tama checked by phospho molybdenum method and ferric reducing power (FRP) method.

Results, in terms of M AAE/ g, were the mean of triplicate experiments.

Butylated hydroxyanisole (BHA) was used as standard antioxidant.

■ Tama ■ BHA

Figure – 4 showed total antioxidant activity of tama checked by phospho molybdenum method and by ferric reducing power (FRP) method. By phospho molybdenum method and by ferric reducing power method Total antioxidant activity of tama came as 5.6 and 3.5 respectively. In case of butylated hydroxyanisole (BHA), the standard antioxidant, the same values were 5.8 and 3.7 respectively. Results were expressed in terms of M AAE/ g.

Amount of antioxidant compounds such as flavonoids, polyphenol, ascorbic acid and carotenoids present in tama are shown in Figure – 5. In terms of mg/g of dry weight, tama contained poly phenol, flavonoids, ascorbic acid and carotenoids as 69.4, 42.8, 25.2 and 18.3 respectively.

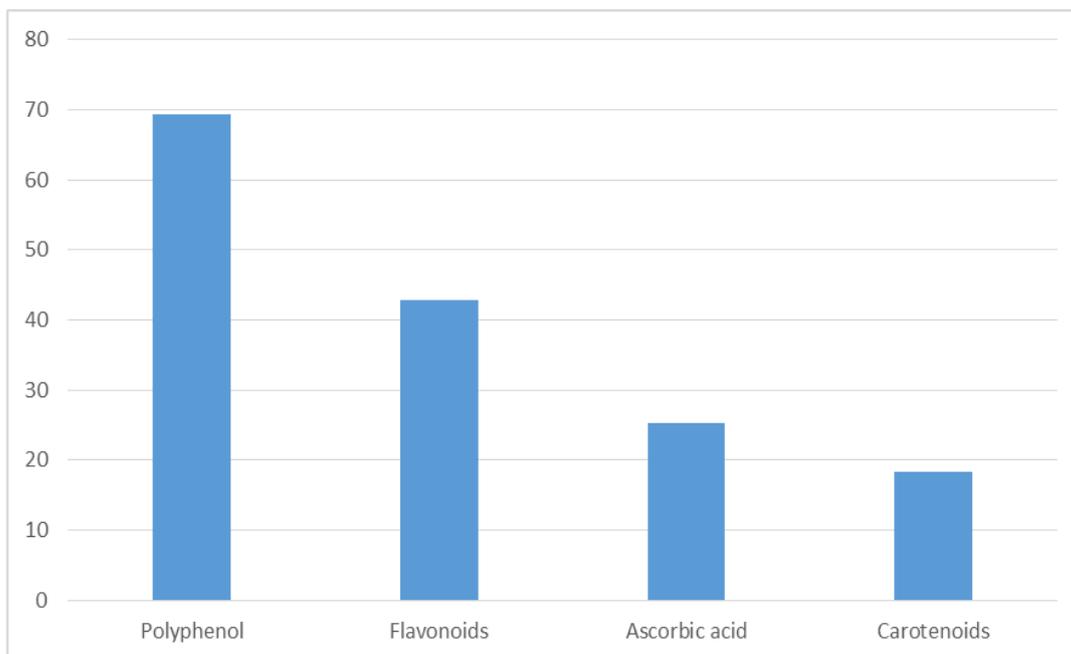


Figure 5: Showing amount of antioxidant compounds like polyphenol, flavonoids, ascorbic acid and carotenoids present in tama.

Results (mg/g of dry weight) were the mean of triplicate experiments.

4. DISCUSSION

Free radicals such as hydroxyl ion, singlet oxygen, superoxide ion, hydrogen peroxide etc. are generated in human cells under normal metabolic activities.^[21] Free radicals are also generated from exogenous sources. These free radicals, in turn, cause oxidative damage to enzymes, proteins, lipids and nucleic acids. As a result body suffers from oxidative stress which is responsible for induction of many chronic and degenerative diseases like cancer, ischemic heart disease atherosclerosis, ageing, diabetes, immunosuppression, etc.^[22]

Antioxidants can break free radical chain reaction thereby minimize oxidative stress. Antioxidant defense mechanism is present in normal human body. Still human body requires more antioxidants. Therefore, there is always high demand of antioxidant compounds. Several antioxidants have been synthesized. Examples are, butylated hydroxytoluene, butylated hydroxyanisole, propyl gallate, TBHQ (tert-butylhydroxyquinone) etc. These synthetic antioxidants are not safe. Their toxicity is a matter of concern. It is often claimed that these antioxidants have many side effects including carcinogenicity.^[23] Therefore, there is high demand for naturally occurring antioxidants. Search for natural antioxidants is continuously going on in many sources such as oilseeds, legumes, cereals, animal products, fruits, vegetables, nuts, etc. Even herbs and medicinal plants were also taken as source of search of natural antioxidants.^[24]

In the present study investigation was carried out on antioxidant potential of tama, one of the ethnic foods of Sikkim. As different antioxidant compounds have different mechanisms of action, several methods were used to assess the antioxidant efficacy of tama.

Results showed that tama has good antioxidant activity as revealed by superoxide anion generation with the help of linoleic acid peroxidation assay, xanthine-xanthine oxidase assay and by DPPH photometric assay. Range of percent inhibition came 86-91% which was more or less close to that of standard antioxidant quercetin, 95 – 100% (Figure – 2). Antioxidant activity of tama was also checked by hydroxy radical scavenging assay, ABTS radical scavenging assay and nitric oxide radical scavenging assay. In all cases tama showed high antioxidant activity and results were comparable to the effect of ascorbic acid (Figure -3). Total antioxidant capacity of tama was further checked by phospho molybdenum method and by ferric reducing power method. Results were expressed in terms of M AAE/ g. In both the two cases tama showed antioxidant activity which was comparable to that of the standard antioxidant butylated hydroxyanisole (Figure – 4).

Antioxidant compounds like polyphenol, flavonoids, ascorbic acid and carotenoids present in tama were estimated. Tama contained high amount of polyphenol [69.4 mg/gm], flavonoids [42.8 mg/gm], ascorbic acid

[25.2 mg/gm] (Figure – 5). Antioxidant properties of various solvent extracts of potato peel, sugar beet pulp and sesame cake were checked. It revealed that high antioxidant activity of the materials was due to its high content of phenolic compounds.^[25] Antioxidant potential of tama may therefore be explained due to its high content of polyphenols, flavonoids and ascorbic acid.

5. CONCLUSION

Present study showed that tama, one of the ethnic foods of Sikkim, has in vitro antioxidant property. Perhaps this antioxidant property of tama helps ethnic group of Sikkim to remain fit and active by inhibiting various oxidation reactions inside their body. In vivo studies are now needed to through more light in this direction.

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Conflict of interest: The authors declare that they have no conflict of interest.

REFERENCES

1. Church, S., Gilbert, P., and Khokhar, S, 2006. *Ethnic Groups and Foods in Europe*, Eurofir Synthesis Report No 3: Available at: <http://www.eurofir.net/>
2. Verbeke. W. and López G.P. Ethnic Food Attitudes And Behavior Among Belgians And Hispanics Living In Belgium. *British Food Journal*, 2005; 107(11): 823-840.
3. Swedish Chambers, 2010. Market Brief. *Focus On The Swedish Ethnic Market*. Published by The Swedish Chambers of Commerce, East Sweden. (http://www.cci.se/_upload/marketbriefs/Fish_nov06.pdf)
4. Dae Young Kwon. What is ethnic food? *Journal of Ethnic Foods*, 2015; 2: 1.
5. Dae Young Kwon. Ethnic foods and their taste: salt and sugar, *J Ethn Foods*, 2017; 4: 133-134.
6. Tamang Jyoti Prakash, Okumiya Kiyohito, Kosaka Yasuyuki. Cultural Adaptation of the Himalayan Ethnic Foods with Special Reference to Sikkim, Arunachal Pradesh and Ladakh, *Himalayan Study Monographs*, 2010; 11: 177-185.
7. Tamang Jyoti Prakash, Thapa Namrata. Some nonfermented ethnic foods of Sikkim in India *J Ethn Foods.*, 2014; 1: 29-33.
8. Mitra Prasenjit, Ghosh Tanaya, Mitra Prasanta Kumar. Effect of extraction solvents on in vitro anti oxidant activity of *Mentha spicata* L. leaves. *World J Pharmacy and Pharmaceutical Sciences*, 2018; 7(4): 1727 – 1736.
9. Chang WS, Chang YH, Lu FJ, Chiang HC. Inhibitory effects of phenolics on xanthine oxidase, *Anticancer Res.*, 1994; 14: 501-506.
10. Chang W, Choi ab, Sei C, Kim ab, Soon S, Hwang a, Bong K, Choi a, Hye J, Ahn a, Min Y, Lee a, Sang H, Park b, Soo K, Kim C. Antioxidant activity

- and free radical scavenging capacity between Korean medicinal plants and flavonoids by assay-guided comparison. *Plant Science*, 2002; 163: 1161-1168.
11. Mensor LL, FMenezes FS, GLeita GG, Reis AS, Dos Santos TC, Coube CS, Leitaõ SG. Screening of Brazilian plant extracts for antioxidant activity by the use of DPPH free radical method, *Phytother Res*, 2001; 15: 127-130.
 12. Shirwaikar A, Shiwaikar A, Rajendran K, Punitha ISJ. *In vitro* antioxidant studies on the benzyl tetra isoquinoline alkaloid berberine. *Biol pharm Bull.*, 2006; 29: 1906-1910.
 13. Halliwell B, Gutteridge JM, Aruoma OI. The deoxyribose method: a simple "test tube" assay for determination of rate constant for reaction of hydroxyl radicals. *Anal Biochem*, 1987; 165: 215-219.
 14. Panda BN, Raj AB, Shrivastava NR, Prathani AR. The evaluation of nitric oxide scavenging activity of *Acalypha indica* Linn Root. *Asian Journal Research, Chemistry*, 2009; 2(2): 148-150.
 15. Prieto P, Pineda M, Aguilar M. Spectrophotometric quantitation of antioxidant capacity through the formation of phosphomolybdenum complex: Specific application to the determination of vitamine E. *Analytical Biochemistry*, 1999; 269: 337-341.
 16. Oyaizu, M. Studies on products of browning reaction: antioxidative activities of products of browning reaction prepared from glucosamine. *Japanese Journal of Nutrition and Dietetics*, 1986; 44(6): 307-15.
 17. Chang C, Yang M, Wen H, Chern J. Estimation of total flavonoid content in propolis by two complementary colorimetric methods. *J. Food Drug Analysis*, 2002; 10: 178-182.
 18. McDonald S, Prenzler PD, Autolovich M, Robards K. Phenolic content and antioxidant activity of olive extracts. *Food Chemistry*, 2001; 73: 73-84.
 19. Cakmak I, Marschner H. Magnesium deficiency and high light intensity enhance activities of superoxide dismutase, ascorbate peroxidase and glutathione reductase in bean leaves. *Plant Physiol.*, 1992; 98: 1222-1227.
 20. Jensen A. Chlorophyll and carotenoids. In: Hallebust JA, Craigie JS. (eds). *Handbook of Physiochemical and Biochemical Methods*. Cambridge University Press, Cambridge, UK, 1978; 5-70.
 21. Halliwell B, Gutteridge JMC. Role of free-radicals and catalytic metal ions in human disease: an overview. *Methods Enzymol*, 1990; 186: 1-85.
 22. Young IS, Woodside JV. Antioxidants in health and disease. *J. Clin. Pathol.*, 2001; 54: 176-186.
 23. Branen AL. Synthetic anti oxidants. *Journal of American Oil Chemist Society*, 1975; 52: 59-63.
 24. Rimbach G, Fuchs J, Packer L. Application of nutrigenomics tools to analyze the role of oxidants and antioxidants in gene expression. In: Rimbach G, Fuchs J, Packer L eds.), *Nutrigenomics*, Taylor and Francis Boca Raton Publishers, FL, USA, 2005; 1-12.
 25. Mohdaly AA, Sarhan MA, Smetanska I, Mahmoud A. Antioxidant properties of various solvent extracts of potato peel, sugar beet pulp and sesame cake. *J Sci Food Agric.*, 2010; 90(2): 218-26.