The Impact of Physical Processes and Chemicals of the Antioxidants (Bioactivity Compounds)

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Executive Summary

Nature has selected and included, in an evolutionary manner, in the design of living bodies, reactions generating free radicals with complex roles: functional, inter-cellular communicational or destructive, etc. The focus of this review is highlighting the effects of physical and chemicals processes on different antioxidants. These changes affect the structures, properties and qualities on activity compounds. The repercussions of these transformations on antioxidants functionality are discussed.

Keywords: Free radicals; Antioxidant activity; Physical and chemicals processes

Background

Free radicals (pro–oxidants) are compounds belonging to the radical groups; they are active biochemically and biologically, and they destroy cell membranes, nuclei, and cytoplasm, producing and maintaining an intense oxidative stress. This is a consequence of the presence of one or several free electrons on the last layer of an atom in the molecule. Free radicals are oxygenated electron–deficit anions that do not make up salts, acids, or bases, but keep this reactive (free) form. The effect of an antioxidant is determined by its availability in a “compartment” to combine with receptors on which it acts to produce a specific effect. The availability of an antioxidant is influenced by transport of molecules through biological membranes, by degree of binding between plasmatic proteins and tissues, by blood flow at level of action site, and by metabolising and elimination of antioxidant [1]. To produce the desired effect, an antioxidant needs to cross cell membranes, i.e. two phospholipid layers. Glycoproteins, lipoproteins, as well as different ion or polar groups on surface of membranes make up membranes. As a result of the interaction of different endogenous or exogenous substances with the receptors on the biological membranes, they can undergo alterations through alterations of spatial orientation of resulting compounds and, consequently, channels/pores measuring up to 8 Å at the level of cell membrane and up to 60–80 Å at the level of capillaries, open up. Besides the opening of ion channels, the effect of interaction mediator–receptor can consist in mobilisation of enzymes mobilising secondary messengers. The potential necessary for transfer through the membranes depends on different factors (Figure 1).

The crossing of the biological membranes by the antioxidant substances depend on:

Factors depending on biological membranes, i.e.: lipid content; existence of specialised transport systems; polarisation of the membrane; presence of pores; membrane physic-pathological state.

Factors depending on the antioxidant substance: molecular mass; chemical structure; ionisation constant (pKa); rate; lipo/hydro solubility of antioxidant substance, etc.

Factors depending on the environment on the surface of the biological membranes: pH; protein bonding; vascularisation; blood flow, etc.

Antioxidant substances can cross biological membranes in two ways: passive transfer and specialised transfer [2]. The physiological role of antioxidants is to prevent the destruction of cell components that occur as a result of chemical reactions involving free radicals. Antioxidant activity depends on that part of molecule with important electron donor properties.

The Impact of Physical Processes

Until recently, antioxidants were considered instruments in fight against oxidation; now that antioxidants are known to maintain and improve health, we need to protect antioxidant nutritional value [3]. High temperatures trigger quick chemical processes. When temperature rises with 10°C, there is doubling or tripling of the chemical reaction speed. Thus, when temperature increases with 1°C,

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reaction speed increases with 10%. Even lower temperature increases result in a shortening of reaction speed.

**Structural integrity**

Structural integrity plays a role in prevention of contact between antioxidants and O₂ thus destroying oxidative potential. Antioxidant is encapsulated in them own protective liposomes or in cell membrane structures and, cannot get into contact with O₂. Intact oil seeds are stable but, once crushed, extracted, warmed in process of refining, oil resistance to oxidation decreases. There is a negative nutrition consequence—the loss of structural integrity, but there is also a positive nutrition consequence—antioxidants become more bioactive [4]. Gastrointestinal absorption of carotenoids from vegetables is inversely proportional with the size of panicle. The treatment procedure of lyocnone at high temperature improves absorption; thus, the amount isomers are not biological equivalents).

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Humidity

Humidity should be carefully controlled. Antioxidant content should be optimised since the diagram of oxidation is U-shaped: it represents the quick rates of oxidation when humidity content is low and when it becomes too high. Most dehydration processes destroy lipoxidases, and when water content decreases below the level allowing the formation of a protective water layer on product surface, self–oxidation potential increases [5]. The high level of water acts as a solvent that mobilises catalysts and reactants towards oxidation. Water can interact chemically through hydrogen bonds with other molecule species.

**Thermal treatment**

Thermal treatment plays an important role due to numerous benefits it plays in preserving foods. It has both positive and negative effects on antioxidants. Positive effects include inactivation of oxidase and breaking of food structures that lead to improved bioactivity. Thermal treatment has an important negative impact on antioxidants in foods. Carotenoids and antioxidants are sensitive to warming: components degrade gradually with time and temperature. Water soluble antioxidants (ascorbic acid) are sensitive to temperature [6]. Another effect of warming is the increase of carotenoid isomerisation in foods. The relevance of this fact is that cis isomers from β–carotene are less absorbed by gastro–intestinal tract and less carried in body (cis and trans isomers from β–carotene are less absorbed by gastro–intestinal tract and less carried in body (cis and trans isomers from β–carotene are less absorbed by gastro–intestinal tract and less carried in body (cis and trans isomers from β–carotene are less absorbed by gastro–intestinal tract and less carried in body (cis and trans isomers from β–carotene are less absorbed by gastro–intestinal tract and less carried in body (cis and trans isomers from β–carotene are less absorbed by gastro–intestinal tract and less carried in body (cis and trans isomers from β–carotene are less absorbed by gastro–intestinal tract and less carried in body (cis and trans isomers from β–carotene are less absorbed by gastro–intestinal tract and less carried in body (cis and trans isomers from β–carotene are less absorbed by gastro–intestinal tract and less carried in body (cis and trans isomers are not biological equivalents).

Storage temperature

Antioxidant losses in fresh fruits and vegetables during storage are significant. At room temperature, 90% of the vitamin C in spinach leaves is lost in three days. At fridge temperature, this loss is about 50%. Freezing can stop and decrease vitamin C losses in peas, broccoli, green beans, and spinach for a period of over three months [7]. Temperature fluctuations have a negative impact on balance between air humidity and products humidity resulting in drying or moisturizing of products. According to the Van’Hoff Law (dependence of chemical reactions on temperature), metabolic activity products decreases at low temperatures to reduce the intensity of biochemical reactions [8,9]. Air temperature oscillations are influenced by environmental air temperature oscillations which are determined by a complex of physical processes.

**Oxygen reduction**

There are processes that allow the removal or reduction of O₂ in foods. This is very important because, for instance, L-ascorbic acid degradation rate in orange juice and canned green peas depends on amount of O₂ in sample. Fruit juices that contain air entrapped are subjected to a vacuum removal that minimise potential of destructive changes on ascorbic acid and on other oxidable components due to the due to molecules to oxygen (O₂) [10,11]. A new approach is use of some enzymes: glucose oxidase (EC 1.1.3.4), thiol oxidase (EC 1.8.3.2), galactose oxidase (EC 1.1.3.9), pyranose oxidase (EC 1.1.3.10) and hexose oxidase (EC 1.1.3.5) related to (EC 1.15.1.1), glutathione transferase (EC 2.5.1.18), catalase (EC 1.11.1.6) and glutathione peroxidase (EC 1.11.1.9) that act by removing O₂ and ROS or by decreasing hydroperoxide lipids [12]. Oxygen lies at basis of many changes as a result of oxidation, which it favours (in presence of light and temperature): it oxidised fats, phenols (colour alterations), liposoluble vitamins (loss of vitamin values), resins, balms, aldehydes, ketones, etc.; it favours polymerisation and resinification of essential oil; it favours rancidisation of vegetal fats; it favours inactivation of antiradical substances acting on active substance.

**Protection against light**

At wave lengths below 500 nm, light generates lipid oxidation and vitamin destruction. Some components behave like photosynthesizers: riboflavin, chlorophyll, myoglobin, haemoglobin pigment, as well as some double–bond systems [13]. Solar rays, particularly those in UV range, produce physical, chemical, etc., alterations. They alter structure and physic–mechanic properties, thus having a negative impact. Under impact of radiations, methionine from protein, together with vitamin B₆, is turned into methylmercaptopropionic aldehyde. A lower or higher permeability to radiations–depending on colour of wrapping, determines destruction of vitamins B₁₂ and C.

**Irradiation**

The effects of irradiation vary depending on antioxidant compounds subjected to irradiation and on strength and length of irradiation time. Irradiation of bell–peppers showed that there is no alteration in content of vitamin C at rates up to 300 Gy. Storage post–irradiation results in increases of amount of vitamin C in fruits and vegetables. At 3 kGy (maximum amount recommended by the Food and Drug Administration, there was a loss of 15% β–tocopherols and 30% α–tocopherols [14]. At 3 GY and 2°C, there is a decrease of 6% in α–tocopherols and no alteration in β–tocopherols.

**Impact of Chemical Processes**

Chemical alterations occur as a result of the action of singular or conjugated factors such as temperature, catalysts, or activation energies [15]. They occur in products as components/substances with properties different from those of the initial products.

**Chemical alterations (reactions)**

Light radiation trigger photochemical reactions that lead to destruction of some valuable elements in products. The effects of photochemical reactions are photolysis and photo–oxidation. Photolysis is photochemical decomposition produced by action of radiations in visible or ultra–violet spectrum. Reaction speed is high in photolytic process. Photo–oxidation takes place in a longer time because, since in photo–oxidative reactions, radiation energy is not
enough for chemical alterations, it must be completed with energy resulted from outer oxidation processes [17]. There are a series of damages, among which corrosion, in which temperature and radiations come after other factors (dry gases) in the triggering and amplification of the damages they produce. Oxygen molecule is the enemy of antioxidants; protecting foods against the interaction with O\textsubscript{2} is the principle on which rely protective antioxidative technologies.

**Enzymes**

Chemico-biological reactions occurring in body are catalysed by enzymes. In living bodies, chemical reactions cannot occur without enzymes. Another characteristic of enzymes is fact that they do not change the reacting reactions, but accelerate them. The best known substances of antioxidant protection system (catalase, glutathione peroxidase, superoxide dismutase) and they achieve the alteration of free radicals. Enzymes are important components of the protection/defence mechanisms. Enzymes eliminate toxic substances from body together with biomolecules, mainly hereditary genetic material and so-called regulation-repair enzymes that repair damage of DNA structure.

The existence of living organisms is conditioned by the activity of some resistance and immunity mechanisms capable of protecting their chemical individuality through recognition and differentiation mechanisms of self-substances from non-self-substances [18,19].

These mechanisms are tolerant to self-molecules, but they activate and react more or less rigorously to remove, neutralise or destroy non-self-substances. Controlling polyphenol oxidation has long been a priority in food science because oxidation phenols are responsible for unwanted brown colour of cut fruits and vegetables and for development of improper colours and flavours in frozen foods. Inactivating these enzymes can be done by a quick treatment of warming (boiling) with preservation of freshness in foods. Quick inactivation of oxidase after apple pressing is a must in establishing apple juice quality [20]. Extrusion, due to the conditions of temperature and pressure, destroy oxidase and favour the bonding of lipids with proteins and carbohydrates in foods, which protects lipid oxidation. Fruit juices that contain a large amount of pulp are more stable than juices with smaller amounts of pulp, due to content of polyphenols and glycoside [21,22].

In the preparation of apple or strawberry juices with less pulp, they often use pectinases. These enzymes hydrolyse the lateral glycosidic chains of flavonoids and isoflavonoids are not completely absorbed by the small intestine, unlike aglycons. The nutritional impact of using pectinases is difficult to quantify: on one hand, polyphenols become more active and, thus, influence health; on the other hand, there are less polyphenols in the products, which can be easily oxidized since they are less stable.

**Supplementing**

For technological reasons, antioxidant nutrients can be added supplementary to foods. Ascorbic acid is used more to increase food resistance to oxidation. Cut fruits and vegetables are saturated in ascorbic acid which is oxidized, thus preventing oxidation of catechin–tannins, and inhibit browning enzymatic reactions. In hydrophobic foods, we use vitamin E is used to protect antioxidants. Since vitamin C "recycles" vitamin E, it is useful to use simultaneously these antioxidants [23,24]. Despite all this, since vitamin C is hydrophobic antioxidants and vitamin E is lipophilic antioxidants lipophilic antioxidants, it is difficult to use them simultaneously in food systems. A solution could be to use ascorbyl–6–palmitate (AP), hydrophobic form of vitamin C which proved to be useful in lipid food systems [25]. The mixture of vitamin E, vitamin C, and phospholipids is an efficient mixture since phospholipids act as emulsifiers allowing contact of two vitamins; phospholipids themselves participate actively in process of anti–oxidation. Both vitamins can be incorporated in liposomes and, thus, they can be used as a feeding system in "oil–in–water" emulsions. Non–nutrient antioxidants such as sulphur dioxide can be used to protect those antioxidants that are nutritionally beneficial [26,27]. Sulphur dioxide prevents carotenoid oxidation but it causes the bleaching of anthocyan.

Supplementary antioxidants from natural sources such as plants and condiments are efficient in protection of foods against oxidation. Plant extracts that have antioxidative properties is: rosemary, sage, thyme, oregano, ginger, cloves, laurel etc. [28] are active in decrease of oxidant impact. Antioxidant mixtures can be obtained from natural sources mixing with oil and pressing, or using extracts in CO\textsubscript{2} from antioxidants. This latter technique is of interest since solvent involved is not toxic; it is flammable and it has a low price.

**Final Remarks**

Free radical action is an inevitable, continuous process, but when it becomes excessive, it is destructive. When the organism’s cells are not protected from oxidative attack, cell damage can be disastrous. Antioxidants are important because they maintain cell and organism systems health. To play a role in destruction of free radicals, antioxidants need: to be present in the proper place and in proper amounts (free radicals have a short life span); to have a non-specific character to react with radical species; to be distributed widely in the intracellular space or to be trans-locatable; to be synthesised by the cells or to be part of nutrition and to be regenerable.

Antioxidant action is due to fact that these substances have an increased ability of bonding oxygen, compared to glycrides, unsaturated fatty acids that bond with more difficulty (rancidity induction period is increased), for instance, α–tocopherol and synthesis substances: propol gallate, octal gallate, duodecil gallate. Certain chemical substances increase the antioxidant effect of these substances (synergic substances): citric acid and ascorbic acid. The synergic effect is due to blocking of metals favouring fat rancidness. Organisms have natural ways of preventing and correcting occurrence of errors and cell atypia–fight against free radicals is carried out with the help of antioxidants who bring cell back to a neuter condition from a redox point of view.

**References**

1. Butnariu M and Samfira I (2012) Free Radicals and Oxidative Stress. J Bioequiv Availab 4: 3.
2. Ferencz A, Juhasz R, Butnariu M, Deer AK, Varga IS, et al. (2012) Expression analysis of heat shock genes in the skin, spleen and blood of common carp (Cyprinus carpio) after cadmium exposure and hyperthermia. Acta Biol Hung 63: 15-25.
3. Gialalone M, Di Sacco F, Traupe I, Topini R, Forfori F, et al. (2011) Antioxidant and neuroprotective properties of blueberry polyphenols: a critical review. Nutr Neurosci 14: 119-125.
4. Bostan C, Butnariu M, Butu M, Ortan A, Butu A, et al. (2013) Allelopathic effect of Festuca rubra on perennial grasses. Rom Biocheln Lett 18: 8190–8196.
5. Gahler S, Otto K, Bohm V (2003) Alterations of vitamin C, total phenolics, and antioxidant capacity as affected by processing tomatoes to different products. J Agric Food Chem 51: 7962-7968.
6. Basu A, Imhan V (2007) Tomatoes versus lycopene in oxidative stress and carcinogenesis: conclusions from clinical trials. Eur J Clin Nut 61: 295-303.
7. Capanoglu E, Beekwilder J, Boyacioglu D, Hall R, de Vos R (2008) Changes in antioxidant and metabolite profiles during production of tomato paste. J Agric Food Chem 56: 964-973.

8. Gliguem H and Birlouez–Aragon I (2005) Effects of sterilization, packaging, and storage on vitamin C degradation, protein denaturation, and glycation in fortified milks. J Dairy Sci 88: 891–899.

9. Schmitt-Hoffmann AH, Roos B, Sauer J, Schleimer M, Kovács P, et al. (2011) Influence of food on the pharmacokinetics of oral alltritoin (9-cis retinoic acid). Clin Exp Dermatol 36 Suppl 2: 18-23.

10. Butnariu M (2012) The oxygen paradox. Pharmacogenomics & Pharmacoproteomics Editorial 3: e104.

11. Somoz V (2008) Physiological effects of thermally treated foods. Mol Nutr Food Res 52: 305-306.

12. Sanjeev K, Ramesh MN (2006) Low oxygen and inert gas processing of foods. Crit Rev Food Sci Nutr 46: 423-451.

13. Ianculov I, Palicica R, Butnariu M, Dumbrava D, Gergen I (2005) Achieving the crystalline state of chlorophyll of the Fir–tree (Abies alba) and the pine (Pinus sylvestris). Rev Chim–Bucharest 56: 441–443.

14. Kumari N, Kumar P, Mitra D, Prasad B, Tiwary BN, et al. (2009) Effects of ionizing radiation on microbial decontamination, phenolic contents, and antioxidant properties of triphala. J Food Sci 74: M109-113.

15. Butnariu M, Sumuleac A, Dehelean C, Chirita R, Saratean V (2006) Studies concerning fertilizer influence (NPK in different doses) on quantity of corn plants chlorophyll. Rev Chim–Bucharest 57: 1138–1142.

16. Zuniga KE, Erdman JW Jr (2011) Combined consumption of soy germ and tomato powders results in altered isoflavone and carotenoid bioavailability in rats. J Agric Food Chem 59: 5335-6341.

17. Casado MF, Cecchini AL, Simao AN, Oliveira RD, Cecchini R (2007) Free radical-mediated pre-hemolytic injury in human red blood cells subjected to lead acetate as evaluated by chemiluminescence. Food Chem Toxicol 45: 945-952.

18. Sorensen M, Jensen BR, Poulsen HE, Deng X, Tygstrup N, et al. (2001) Effects of a Brussels sprouts extract on oxidative DNA damage and metabolising enzymes in rat liver. Food Chem Toxicol 39: 533-540.

19. Matzinger P (2002) The danger model: a renewed sense of self. Science 296: 301-305.

20. Butnariu M (2012) Action and Protection Mechanisms of Free Radicals. J Pharmacogenom Pharmacoproteomics 3: 6.

21. Ozcan ME, Gulec M, Ozzerol E, Polat R, Akyol O (2004) Antioxidant enzyme activities and oxidative stress in affective disorders. Int Clin Psychopharmacol 19: 89-95.

22. Hathwar SC, Bijnu B, Rai AK, Narayan B (2011) Simultaneous recovery of lipids and proteins by enzymatic hydrolysis of fish industry waste using different commercial proteases. Appl Biochem Biotechnol 164: 115-124.

23. Butnariu M and Corneanu M (2012) The Screening of the Analytical Biochemistry Researches Involved In Plant Response to Stress Conditions. Biochem Anal Biochem. 1:7.

24. Malo C, Gil L, Gonzalez N, Martinez F, Caro R, et al. (2010) Anti-oxidant supplementation improves boar sperm characteristics and fertility after cryopreservation: comparison between cysteine and rosemary (Rosmarinus officinalis). Cryobiology 61: 142–147.

25. Venkateswara B, Sirisha K, Chava VK (2011) Green tea extract for periodontal health. J Indian Soc Periodontol 15: 18-22.

26. Zhao F, Liu ZQ (2011) Comparison of antioxidant effectiveness of lipoic acid and dihydrolipoic acid. J Biochem Mol Toxicol 25: 216-223.

27. Barbat C, Rodino S, Petache S, Butu M, Butnariu M (2013) Microencapsulation of the allelochemical compounds and study of their release from different products. Dig J Nanomater Bios. 8: 945-953.

28. Akinci M, Kosova F, Cetin B, Sepici A, Altan N, et al. (2008) Oxidant/antioxidant balance in patients with thyroid cancer. Acta Cir Bras 23: 551-554.