Aspects of antioxidant foods and supplements in health and disease

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Free radicals generated as byproducts of normal metabolism can damage biologically relevant molecules. When their generation is increased, damage can also be increased, resulting in the development of many pathological conditions. Antioxidant defenses protect the body from the detrimental effects of free radicals. Dietary fruits and vegetables provide a reasonable amount of compounds that act as physiological antioxidants. Although existing knowledge does not allow a final and conclusive assessment of the relevance of antioxidants for health, it does provide the basis for its rational consideration. This paper addresses the specific aspects of antioxidant supplementation in health and disease.

AGING, EXERCISE, AND ANTIOXIDANT SUPPLEMENTS

The 57th World Health Assembly (Geneva, 17–22 May, 2004) of the World Health Organization recognized the importance of both a healthy diet and regular and adequate physical activity as major factors in the promotion and maintenance of good health throughout the entire life course. One currently recognized characteristic of a healthy diet includes components that counteract oxidative stress, which, besides being involved in the etiopathogeny and progression of chronic diseases, contributes to the process of aging.[1,2,3] Since antioxidant defenses decrease with aging and the production of reactive oxygen species (ROS) increases with both aging and exercise, the concurrent effect of aging and exercise could exacerbate oxidative stress.[4,5,6,7,8,9]

To determine whether this deleterious process could be decreased or palliated by dietary antioxidant treatment, the group of Dr R Jiménez studied the potential effect of both exercise and antioxidant treatment in healthy aged subjects. The work was a long-term (2 years) and large-scale (400 aged subjects, 58–86 years old) intervention study based on both moderately intense exercise and concurrent antioxidant treatment consisting of daily intake of a functional antioxidant beverage, Funciona™, a blend of antioxidant-rich fruit extracts enriched with antioxidant vitamins. The subjects were randomly incorporated into one of three groups: 1) control (no antioxidant treatment, no training), 2) training (3 sessions per week, 50-min sessions), and 3) training plus antioxidant treatment (330 mL/day of Funciona™). After 10 months of these interventions, it was found that even though exercise caused oxidative stress, the antioxidant treatment prevented this effect.[5,10] These findings demonstrate that although moderate training elicits a number of favorable responses and improves physical health in elderly people, even if initiated in later years, regular exercise might be accompanied by antioxidant supplementation to attain an optimum level of defense against the free radical generation associated with aging and exercise.[2]

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OXIDATIVE STRESS AND BRAIN FUNCTION

The brain is particularly vulnerable to ROS for several reasons: 1) it consumes approximately 20% of the total body oxygen despite the fact that it comprises less than 2% of total body weight, 2) it contains high levels of polyunsaturated fatty acids and may contain low proportions of endogenous antioxidants, 3) iron accumulates in brain-specific regions, and 4) iron-binding proteins (i.e., ferritin) may be relatively deficient in the brain. These characteristics and evidence from both direct and indirect studies have associated neurodegenerative diseases with oxidative stress conditions and emphasize the use of antioxidants as prophylactic agents in their management.[11,12,13,14]

Dr OI Aruoma discussed the view that neurodegenerative disorders are associated with various degrees of behavioral impairments that significantly decrease the quality of life. Bioactive compounds (in particular, those possessing antioxidant activities) in traditional medicine and food plant extracts may be particularly important in protecting against a number of neurological diseases in which oxidative stress is implicated.[12,13,15] Elucidation of the intracellular pathways associated with free radicals in neuronal cells is essential in order to gain insight into the pathophysiologic basis for neuronal death and to enable pharmacologic strategies to ameliorate such neuronal degeneration.[15,16] In addition, the potential neuroprotective effects of flavonoids against the neuronal deficits associated with aging or age-related neurodegenerative diseases is of increasing interest.

Cellular studies examining the potential mechanisms of neuroprotection by flavonoids in preventing neuronal cell death caused by oxidative stress have identified three different mechanisms of flavonoids:[15,16] 1) they can act as scavengers for ROS, 2) they maintain the correct glutathione levels, and 3) they inhibit the Ca^{2+} influx, which represents the last step in the cell death cascade. Thus, these qualities coupled with the anti-inflammatory properties of certain flavonoids could render these compounds suitable for application where oxidative stress, inflammation, and antioxidant defense depletion take place, such as in Alzheimer's and Parkinson's disease. However, emerging evidence suggests that the antioxidant activity of phenolic compounds is unlikely to be the sole mechanism responsible for their protective action.

Flavonoids can exert modulatory actions in cells by interacting with a wide spectrum of molecular targets central to the cell-signaling machinery. These include activation of mitogen-activated protein kinase, protein kinase C, serine/threonine protein kinase Akt/PKB, and phase II antioxidant detoxifying enzymes; downregulation of proinflammatory enzymes (COX-2 and iNOS) through the activation of peroxisome proliferator-activated receptor gamma; regulation of calcium homeostasis; inhibition of phosphoinositide 3-kinase, tyrosine kinases, NFκB, c-JUN, and antioxidant response element pathways, as well as modulation of several cell survival/cell-cycle genes.[15,17,18] Therefore, in addition to their antioxidant capacity, phenolic compounds may exert a protective effect by selectively inhibiting or stimulating key proteins in the cell-signaling cascades, a point further discussed by Dr C Fraga.

ANTIOXIDANT VITAMINS AND CANCER

The protective efficacy of supplementation with a combination of antioxidant vitamins and minerals in reducing the incidence of cancer and ischemic vascular disease in the general population has been proposed.[19,20] Dr I Sánchez-García reviewed the nutritional factors involved in the genesis of cancer, since they may increase the risk of cancer or act as chemoprotective agents. In fact, two-thirds of human cancer could be prevented by modifying lifestyle habits, specifically, nutritional habits. Thus, the consumption of fruits and vegetables is associated with a lower incidence of certain types of cancer, with the molecular mechanism(s) involved in such an association being largely unknown.

Dr S Hercberg summarized the results of the SU.VI.MAX study, [19] which used a combination of antioxidants at nutritional doses typically attainable with a balanced diet rich in fruits and vegetables. A total of 12,741 apparently healthy adult subjects of both sexes were assigned at random to one of two groups: group 1 received a single daily capsule of a combination of 120 mg vitamin C, 30 mg vitamin E, 6 mg beta-carotene, 100 μg selenium (as selenium-enriched yeast), and 20 mg zinc (as gluconate); group 2 received a placebo. The follow-up time was 7.5 years. At baseline, plasma levels of beta-carotene and vitamin C were higher in women than in men. A total of 271 subjects developed ischemic vascular disease but no differences were detected between the two groups and no gender-group interaction was detected. Moreover, although there was no difference between the groups in the incidence of cancer, a significant protective effect of antioxidants was found in men but not in women. Concerning mortality, no overall significant difference was observed for all-cause mortality between groups. Again, a gender-group interaction was observed, showing a significant protective effect of antioxidants on the development of cancer in men. It is proposed that the gender difference observed in the study could be due to the lower baseline status observed in men for certain antioxidants, especially beta-carotene.

Thus, the efficiency of supplementation in reducing cancer incidence may be related to the ability to correct antioxidant status with an adequate dose of antioxidant

nutrients in subjects with suboptimal antioxidant status (such as men in the SU.VI.MAX study). This hypothesis is supported by the results of a trial conducted in Linxian, China,[21] and performed in a general population of males with very low baseline micronutrient status. The study used a combination of nutritional doses of antioxidants, including beta-carotene. This has been the only trial that found a statistically significant reduction in total and specific cancer incidence and overall mortality after supplementation with antioxidants. However, results of other large primary intervention trials were consistent with the lack of effect of supplementation of antioxidants on cardiovascular diseases, whereas the protective effect on cancer was variable. Two studies did not find a positive effect (Physicians' Health Study[22] and Women's Health Study[23]), and two others actually found a deleterious effect (Alpha-Tocopherol Beta-Carotene Cancer Prevention Study,[24] and β-Carotene and Retinol Efficacy Trial[25]), especially on lung cancers. The main difference between the SU.VI.MAX study and these others is likely to be related to the doses and types of antioxidants used, as well as recruitment methods, selection criteria, and population characteristics.

From these trials, it may be concluded that antioxidant supplementation may have a beneficial effect on cancer incidence only in healthy subjects who are not exposed to cancer risk and who have a particularly low baseline status. High doses of antioxidant supplements may be deleterious in high-risk subjects without any clinical symptoms in whom the initial phase of cancer development has already started. Moreover, antioxidant supplementation could be ineffective in well-nourished subjects with adequate antioxidant status. The results of the SU.VI.MAX study therefore suggest that an adequate and well-balanced supplementation of antioxidant nutrients, at doses that might be reached by a healthy diet with a high consumption of fruits and vegetables, has protective effects against cancer in male subjects. The ineffectiveness of supplementation in women may be due to their better baseline antioxidant status.

The results of the SU.VI.MAX study reinforce the general recommendations of a life-long diversified diet containing foods that are rich sources of antioxidant nutrients. On the other hand, the benefit of long-term supplementation with high-doses of antioxidants, especially in subjects exposed to risk factors for cancers or those with infra-clinical cancers, is still lacking convincing evidence.

POLYPHENOLS AS ANTIOXIDANTS

Dr C Fraga discussed the potential role of plant polyphenols (PPs) as antioxidants for human health, especially as related to cardiovascular disease. It has been proposed that plant PPs can contribute to the health benefits associated with the consumption of fruits and vegetables.[26] Flavan-3-ols (catechin, epicatechin), flavonols (quercetin), isoflavones (genistein), stilbenoids (resveratrol), and curcuminoids (curcumin), are examples of PPs that have been studied extensively in the last decade with regard to the prevention of cardiovascular disease, cancer, neurodegeneration, and other diseases. In a significant part of this research, the free radical scavenging capacity has been considered the major biological activity of PPs.[27] However, the very limited tissue bioavailability of PPs hardly supports the in vivo relevance of such systemic free radical scavenging action. On this basis, the research on PPs has been refocused to evaluate alternative molecular mechanisms that explain the antioxidant effects observed in vivo. In this regard, the interactions of PPs with cell membranes[28] and proteins, along with the modulation of cell signals and gene transcription,[29] have been indicated as potential mechanisms underlying the beneficial health effects of PPs. Importantly, these interactions can occur at physiologically relevant concentrations of PPs.[17] In the case of cardiovascular disease, several epidemiological studies have related a high consumption of PPs with a decreased risk of cardiovascular disease and hypertension.[30,31,32,33,34]

Considering that cardiovascular pathologies, including hypertension, have been consistently associated with ROS-mediated adverse effects, the beneficial effects of flavanol consumption could be easily linked to an antioxidant action, but as mentioned, the actual levels of these compounds found in the body do not underscore such interpretation. The interactions of polyphenol membranes and of polyphenol proteins (enzymes, transcription factors) that were demonstrated in vitro and in cell cultures can occur within the expected levels of PPs in the body. Clearly, the occurrence of such mechanisms does not preclude a direct free radical or metal-chelating action. Furthermore, the actual occurrence of these potential interactions in vivo should be considered when linking an antioxidant action to a PP or to any other molecule whose metabolic fate and biological actions are not firmly established.

CONCLUSION

In conclusion, although the role of oxidative stress in aging, neurodegenerative and vascular diseases, cancer, diabetes, and other related diseases is largely accepted, the value of antioxidant strategies is still debatable. This becomes more important when, apart from foods or reasonable lifestyle changes, antioxidant supplements are considered. Well-defined long-term trials are still needed to asses the efficacy of antioxidant strategies or of antioxidant-rich nutritional intervention. Any persistent

reduction in oxidative stress should be monitored using appropriate, reliable, and sensitive biomarkers, and the real health benefits of lowering the oxidative stress should be assessed critically. Meanwhile, a well-balanced diet rich in fruits and vegetables is highly advised.

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REFERENCES

1. Harmar AJ. Aging: overview. Ann NY Acad Sci. 2001;928:1–21.
2. Jiménez R. Aging and oxidative stress. The role of antioxidants. In: Marroni NP, ed. *Estresse oxidativo e antioxidantes*. Porto Alegre: Editora da ULBRA; 2002:131–152.
3. Sachidanandam K, Fagan SC, Ergul A. Oxidative stress and cardiovascular disease: antioxidants and unresolved issues. Cardiovasc Drug Rev. 2005;23:115–132.
4. Chevion S, Moran DS, Heled Y, et al. Plasma antioxidant status and cell injury after severe physical exercise. Proc Natl Acad Sci USA. 2003;100:5119–5123.
5. Galán MI, Palacios E, Ruiz F, et al. Exercise, oxidative stress and risk of cardiovascular disease in the elderly. Protective role of antioxidant functional foods. Biofactors. 2006;27:167–183.
6. Hawkins S, Wiswell R. Rate and mechanism of maximal oxygen consumption decline with aging: implications for exercise training. Sports Med. 2001;33:877–888.
7. Jessup JV, Horne C, Yarandi H, Quindry J. The effects of endurance exercise and vitamin E on oxidative stress in the elderly. Biol Res Nurs. 2003;5:47–55.
8. Palomero J, Galán AI, Muñoz ME, Tuñón MJ, Jiménez R. Effects of aging on the susceptibility to the toxic effect of cyclosporin A in rats. Changes in liver glutathione and antioxidant enzymes. Free Radic Biol Med. 2001;30:836–845.
9. Rousseau AS, Margaritis I, Arnaud J, Faure H, Roussel AM. Physical activity alters antioxidant status in exercising elderly subjects. J Nutr Biochem. 2006;17:463–470.
10. Jiménez R, Delgado MA, Muguerza B, et al. Physical activity and antioxidant vitamins for the elderly: beneficial effects of a long-term dietary treatment with antioxidant vitamins on the prooxidant effects of physical activity in a large-scale study in aged healthy people. Free Radic Biol Med. 2004;36(Suppl 1):16.
11. Joseph JA, Shukitt B, Denisova NA, et al. Reversals of age-related declines in neuronal signal transduction, cognitive, and motor behavioural deficits with blueberry, spinach or strawberry dietary supplementation. J Neurosci. 1999;19:8114–8121.
12. Mandel S, Youdim M. Catechin polyphenols: neurodegeneration and neuroprotection in neurodegenerative diseases. Free Radic Biol Med. 2004;37:304–317.
13. Markesbery WR, Carney JM. Oxidative alterations in Alzheimer's disease. Brain Pathol. 1999;9:133–146.
14. Sutherland BA, Rahman RMA, Appleton I. Mechanisms of action of green tea catechins, with a focus on ischemia-induced neurodegeneration. J Nutr Biochem. 2006;17:291–306.
15. Aruoma OI, Sun B, Fujii H, et al. Low molecular proanthocyanidin dietary biofactor Oligonol: Its modulation of oxidative stress, bioefficacy, neuroprotection, food application and chemoprevention potentials. Biofactors. 2006;27:245–265.
16. Aruoma OI, Bahorun T, Jen L-S. Neuroprotection by bioactive components in medicinal and food plant extracts. Mutat Res. 2003;544:203–215.
17. Mackenzie GG, Carrasquedo F, Delfino JM, Keen CL, Fraga CG, Oteiza PI. Epicatechin, catechin, and dimeric procyanidins inhibit PMA-induced NF-kappaB activation at multiple steps in Jurkat T cells. FASEB J. 2004;18:167–169.
18. Williams RJ, Spencer JPE, Rice-Evans C. Flavonoids: antioxidants or signalling molecules. Free Radic Biol Med. 2004;36:838–849.
19. Hercberg S, Galan P, Preziosi P, et al. The SU.VI.MAX study: a randomised, placebo-controlled trial of the health effects of antioxidant vitamins and minerals. Arch Intern Med. 2004;164:2335–2343.
20. Hercberg S, Preziosi P, Briançon S, et al. A primary prevention trial of nutritional doses of antioxidant vitamins and minerals on cardiovascular diseases and cancers in general population: the SU.VI.MAX Study. Design, methods and participant characteristics. Control Clin Trials. 1998;19:336–351.
21. Blot WJ, Li JY, Taylor PR, et al. Nutrition intervention trials in Linxian, China: supplementation with specific vitamin/mineral combinations, cancer incidence, and disease-specific mortality in the general population. J Natl Cancer Inst. 1993;85:1483–1492.
22. Hennekens CH, Buring JE, Manson JE, et al. Lack of effect of long-term supplementation with beta-carotene on the incidence of malignant neoplasms and cardiovascular disease. N Engl J Med. 1996;334:1145–1149.
23. Lee IM, Cook NR, Manson JE, Buring JE, Hennekens CH. Beta-carotene supplementation and incidence of cancer and cardiovascular disease: the Women's Health Study. J Natl Cancer Inst. 1999;24:2102–2106.
24. The Alpha-Tocopherol BCCPSG. The effect of vitamin E and beta carotene on the incidence of lung cancer and other cancers in male smokers. N Engl J Med. 1994;330:1029–1035.
25. Omenn GS, Goodman GE, Thorquist MD, et al. Effects of a combination of beta-carotene and vitamin A on lung cancer and cardiovascular disease. N Engl J Med. 1996;334:1150–1155.
26. Stainmetz KA, Potter JD. Vegetables, fruit and cancer prevention: a review. J Am Diet Assoc. 1996;96:1027–1039.
27. Fraga CG. Plant polyphenols: how to translate their in vitro antioxidant actions to in vivo conditions. IUBMB Life. 2007;59:308–315.
28. Verstraeten SV, Keen CL, Schmitz HH, Fraga CG, Oteiza PI. Flavan-3-ols and procyanidins protect liposomes against lipid oxidation and disruption of the bilayer structure. Free Radic Biol Med. 2003;34:84–92.
29. Surh YJ, Kundu JK, Na HK, Lee JS. Redox-sensitive transcription factors as prime targets for chemoprevention with anti-inflammatory and antioxidative phytochemicals. J Nutr. 2005;135(Suppl):S2993–S3001.
30. Arts IC, Jacobs DR, Harnack LJ, Gross M, Folsom AR. Dietary catechins in relation to coronary heart disease death among postmenopausal women. Epidemiology. 2001;12:668–675.
31. Buijsse B, Feskens EJ, Kok FJ, Kromhout D. Cocoa intake, blood pressure, and cardiovascular mortality: the Zutphen Elderly Study. Arch Intern Med. 2006;166:411–417.

32. Geleijnse JM, Launer LJ, Hofman A, Pols HA, Witteman JC. Tea flavonoids may protect against atherosclerosis: the Rotterdam Study. Arch Intern Med. 1999;159:2170–2174.
33. Hertog MG, Kromhout D, Aravanis C, et al. Flavonoid intake and long-term risk of coronary heart disease and cancer in the Seven Countries Study. Arch Intern Med. 1995;155:381–386.
34. Mennen LI, Sapinho D, de Bree A, et al. Consumption of foods rich in flavonoids is related to a decreased cardiovascular risk in apparently healthy French women. J Nutr. 2004;134:923–926.