Fruit Polyphenolics and Brain Aging

Nutritional Interventions Targeting Age-related Neuronal and Behavioral Deficits

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ABSTRACT: Nutritional interventions, in this case, increasing dietary intake of fruits and vegetables, can retard and even reverse age-related declines in brain function and in cognitive and motor performance in rats. Our lab has shown that as Fischer 344 rats age their brains are increasingly vulnerable to oxidative stress. Dietary supplementation with fruit or vegetable extracts high in antioxidants (e.g., blueberry, BB, spinach, respectively) can decrease this vulnerability to oxidative stress as assessed in vivo by examining reductions in neuronal signaling and behavioral deficits and in vitro via H₂O₂-induced decrements in striatal synaptosomal calcium buffering. Examinations have also revealed that BB supplementations are effective in antagonizing other age-related changes in brain and behavior, as well as decreasing indices of inflammation and oxidative stress in gastrocnemius and quadriceps muscles. In ongoing studies we are attempting to determine the most effective BB polyphenolic components. To date, the anthocyanins show the most efficacy in penetrating the cell membrane and in providing antioxidant protection. In sum, our results indicate that increasing dietary intake of fruits and vegetables high in antioxidant activity may be an important component of a healthy living strategy designed to maximize neuronal and cognitive functioning into old age.

KEYWORDS: dietary supplementation; diet; memory; learning; phytochemicals; phytoneutrients; free radicals; oxidative stress; inflammation; behavior; cognitive behavior; blueberry; signal transduction; calcium flux; rats

As humans age they become increasingly likely to display age-related deficits in cognitive and motor performance, even when there is no underlying disease. One strategy to maximize the healthy life span and to retard and/or reverse the impairments associated with normal aging is through nutritional interventions. Research has demonstrated that diets rich in fruits and vegetables are effective at reducing the

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rates of certain types of cancer and cardiovascular disease. Our lab has examined the effects of dietary supplementation with fruits and vegetables on the neuronal and behavioral decrements seen with aging. To date, supplementing a rat’s diet with a fruit or vegetable (e.g., blueberry, strawberry, spinach) has slowed and, in some cases, even reversed deficits in brain function, motor performance, and learning and memory in old animals.

The brain uses a proportionally large amount of the body’s oxygen supply, and with age its endogenous capacity to fight oxidative stress decreases. The free radical theory of aging purports that as the balance between prooxidants and antioxidants shifts in favor of the prooxidants, damage to the brain results in decrements in neuronal and behavioral functioning. We have shown that the fruits and vegetables that are most effective at ameliorating age-related deficits (e.g., signal transduction, motor performance, and cognitive behavior) are those highest in antioxidant capacity. However, cumulatively the multiple polyphenolic compounds present in fruits and vegetables have been shown to have additional effects, including antiinflammatory, antiallergic, antiviral, and antiproliferative, which suggests that their impact on central nervous system (CNS) function and on behavioral parameters may stem from a combination of factors.

A number of neuronal mechanisms and behavioral measures have been shown by us and others to be affected by age in Fischer 344 (F344) rats. Significant decrements occur in neuronal signal transduction, as measured by striatal dopamine release and GTPase activity as well as calcium clearance from striatal synaptosomes and measures of neuronal membrane fluidity, which appear to be age and oxidative stress dependent. Age-related changes in CNS function are also evident in altered psychomotor and cognitive behaviors. Motor performance tests of balance, coordination, strength, and stamina clearly illustrate age-related declines starting at 12 months and continuing through 22 months. A cognitive task assessing spatial learning and memory, the Morris water maze, has also characterized deficits associated with normal aging, changes that parallel decrements in similar abilities in humans. Age-sensitive neuronal and behavioral measures such as these can be expected to be affected by nutritional interventions seeking to retard or reverse the effects of brain aging.

Dietary supplementation with strawberry extract (STB) or spinach extract (SPC) from 6 to 15 months of age (Male F344 rats, source: NIA colony) prevented the onset of age-related deficits in signal transduction as measured by oxotremorine-enhanced dopamine (OX-enhanced DA) release and carbachol-stimulated GTPase coupling/uncoupling in isolated striatal slices, as compared to animals fed a control diet. Additionally, aged animals fed a STB-, SPC-, or blueberry extract (BB)–supplemented diet for 8 weeks (19–21 months) demonstrated similar antiaging effects. In fact, the BB diet reversed the age-related deficits in OX-enhanced DA release.

Efficient regulation of calcium flux and homeostasis is critical for signal transduction and optimal neuronal functioning. Fruit-supplemented diets high in polyphenolics were shown to have an antiaging effect on striatal Ca²⁺ uptake and recovery after depolarization in striatal synaptosomes. The BB diet was especially effective, reversing the age-related decrement in Ca²⁺ recovery in the presence of hydrogen peroxide (H₂O₂)–induced oxidative stress, as compared to control groups. In addition, the BB diet improved cerebellar membrane fluidity, as measured by significantly reduced anisotropy, compared to aged subjects maintained on a control diet.
Behavioral measures showed some of the strongest antiaging effects of the fruit polyphenolic–supplemented diet. On three psychomotor tasks, the rod walk and accelerating roto-rod, which assess balance, coordination, and resistance to fatigue, and the inclined screen, which measures strength, stamina, and balance, the BB diet significantly improved the performance of old animals.

In addition, a measure of cognitive performance was improved by all three fruit- and vegetable-supplemented diets. The Morris water maze (MWM) is a spatial learning task that measures acquisition, working memory, reference memory, the ability to shift set, and spatial learning strategies. The maze is a featureless circular pool of room temperature water (1 m diameter) containing a 10-cm-diameter movable platform hidden just below the surface of the water. Rats swim well and use distal cues to learn to navigate to the hidden platform. This age-sensitive paradigm is a well-accepted measure of hippocampally based learning and memory. Dietary supplementation with either STB, SPC, or BB for eight weeks antagonized the age-related impairments in MWM performance.

As might be suggested by the robust behavioral results, the physiological effects of polyphenolic dietary supplementation is not limited to the CNS. In the periphery, we have found that a BB diet decreases markers of inflammation and oxidative stress in the gastrocnemius and quadriceps muscles (in preparation) and have also shown BB supplementation to significantly increase red blood cell (RBC) membrane fluidity.

Ongoing studies seek to characterize which polyphenolic compounds present in BB and other fruits and vegetables may contribute to the significant antiaging effects described above. Initial studies have determined that anthocyanins and hydroxycinnamic acids (HCA) isolated from blueberries are able to ameliorate in vitro H$_2$O$_2$-induced oxidative stress, with the anthocyanins significantly protecting RBCs against reactive oxygen species even at low doses. Following a 24-hour fast, F344 rats were anesthetized and either anthocyanin or HCA, extracted from blueberry skin or flesh, respectively, were administered by stomach intubation. RBC susceptibility to H$_2$O$_2$-mediated oxidative stress was assessed by dichlorofluorescein assay at 0-, 1-, 6-, and 24-hours postgavage. While the total concentration of plasma anthocyanin and HCA fell off rapidly from 1 hour on, significant protection from reactive oxygen species was seen at 6 and 24 hours. Other experiments have shown that berry anthocyanins penetrate a variety of cell types and that incorporation into the cell’s cytosol protects against H$_2$O$_2$-induced oxidative stress and loss of cell function. Further studies will investigate the protective effects of a variety of polyphenolic compounds in order to clarify the complete profile of phytonutrients found in fruits and vegetables with antiaging effects.

Diets supplemented with fruits and vegetables rich in color and correspondingly high in anthocyanins and other polyphenolic phytonutrients with antioxidant and additional bioactive properties antagonize the effects of aging in our animal model. The amounts of BB, STB, and SPC added to the diets were matched for antioxidant capacity and are roughly equivalent to a daily one cup portion for humans. In sum, research from our lab and others suggests that increasing dietary intake of fruits and vegetables high in antioxidant activity may be an important component of a healthy living strategy designed to maximize neuronal and cognitive functioning into old age.
REFERENCES


