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Commentary

Why Vitamin E Therapy Fails for Treatment of Alzheimer's Disease

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INTRODUCTION

Reactive oxygen species (ROS) damage in the Alzheimer's disease (AD) brain is widely reported at the levels of nucleic acids [1], protein [2], and lipids [3]. ROS damage is also associated with heart disease, cancer, and aging. Consequently, numerous clinical trials of the lipid soluble antioxidant, vitamin E, have been conducted for prevention [4] and treatment [5] of age-related morbidity and mortality. The meta-analysis by Miller and colleagues [5] clearly shows that among 19 clinical trials, only the smaller trials show either an increase or a decrease in all-cause mortality and that the overall effect is near zero. By organizing these studies into a dose-response curve, a significant increase in all-cause mortality was observed for vitamin E doses above 400 IU/day. In an earlier trial of vitamin E treatment for AD at the high dose of 2000 IU/day, there was no effect of vitamin E on Mini-Mental Examination Status (MMSE) scores of moderately severe AD patients, despite delays in nursing home placement [6]. In a trial of mild cognitive impairment (MCI) patients, Petersen and collaborators [7] found that even early vitamin E treatment failed to improve cognition. These results prompted Lloret et al. [8] to introduce the novel concept of stratifying AD patients into vitamin E respondents and non-respondents, based on measures of plasma oxidized glutathione (GSSG), the oxidized form of the common antioxidant glutathione (GSH). At the borderline detrimental dose of 800 IU/day [5], about half of the patients failed to respond to vitamin E with lower plasma GSSG; they showed a lower MMSE score after 6 months that suggested a 13% decline in cognitive performance [8]. The other half of the patients for whose GSSG decreased with vitamin E treatment did not significantly change their original MMSE score. Unfortunately, the study was not large enough to detect a decline in MMSE for treatment with placebo. Also provocative from this report was a strong negative correlation between GSSG and MMSE, but most of the effect was due to 4 of 19 AD patients with a 5–10% drop in GSSG in response to vitamin E.

Given a robust rise in oxidized macromolecules with aging and AD, and the assumption that oxidation is causing symptoms, why is the antioxidant vitamin E ineffective? The findings by Lloret and colleagues indicate that vitamin E does not, in fact, lower plasma oxidative stress for half of the AD patients. To understand this result, we need to consider the broader scope of how antioxidants might reduce the load of macromolecular ROS damage. Is it reasonable to expect that a lipid-soluble antioxidant would protect against oxidation of aqueous phase nucleic acids and proteins? The lipophilic vitamin E partitions into membranes where

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it is available to receive an unpaired electron ("oxyradical") from an oxidized conjugated lipid. But if the oxidized vitamin E is not removed, it will either accumulate or pass the electron on to another lipid and further damage the membrane. Oxidized vitamin E can be recycled by passing the extra electron to a water soluble electron acceptor such as ascorbate (vitamin C), urea, pyruvate, NADH, cysteine, or GSH. As elegantly described by Dean Jones (Emory University), whether an antioxidant is recycled depends on the redox potential in the local cellular environment [9], just as pH depends on local buffers. Thus, the half of the AD patients in Lloret's study who showed lower plasma ratio of GSH to GSSG in response to vitamin E could have an oxidized plasma redox potential that energetically blocks removal of oxyradical damage. Unfortunately, the actual redox potential, $E(mV) = -264 - 30\log([GSH]^2/[GSSG])$ [9], was not computed to facilitate comparisons to the literature. A rough calculation of Lloret's 15% increase in GSSG would produce a $+4$ mV shift toward oxidation of plasma GSH/GSSG. It is not certain that this change in potential energy can account for the claimed 15% decline in MMSE (4 points) in the non-responders. Healthy, young adults have a mean plasma glutathione redox potential of -137 mV that begins at age 50 to trend toward a more oxidized -120 mV by age 85 [10]. Similar to other reports from Jose Vina's group, Jones finds an increasingly oxidized plasma redox potential to correlate with frailty, diabetes, hypertension, and cancer [11].

In a larger context, several possibilities arise from these considerations as to why vitamin E therapy is ineffective for some or outright detrimental for others: 1) wrong dose; 2) wrong timing; 3) unbalanced monotherapy; and 4) wrong target.

1) Wrong dose. The dose-response meta-analysis of Miller and colleagues [5] suggests that doses of vitamin E above 400 IU/day may increase mortality. Can we identify those patients with poor response profiles? The work of Lloret et al. [8] suggests that dose may need to be adjusted at least for some individuals to obtain a less oxidized redox potential in plasma.

2) Wrong timing. In cases of clinical AD or possibly even MCI, synapses may be lost and neurons may develop neurofibrillary tangles at a faster rate than they can be replaced. Peterson et al. [7] also failed to improve cognition with vitamin E treatment of patients with MCI. At these stages of the disease, no trial has succeeded in reversing the disease process. This fact alone could explain the failure of vitamin E timing to improve cognitive scores in the study by Lloret et al.

3) Unbalanced monotherapy. As discussed above, vitamin C or other water soluble electron acceptors may be needed in conjunction with vitamin E to facilitate systemic removal of ROS. For these reasons, a current PREADVISE trial is evaluating treatment of AD with vitamin E and selenium, the trace metal required for glutathione peroxidase. Epidemiological studies suggest an AD protective effect for use of vitamin E together with vitamin C, while either alone is not significantly protective [12]. However, since numerous trials of this combination and other antioxidants have failed to show efficacy in prevention of various diseases [4], even this approach may be inadequate. Lloret et al. [8] recommend use of phytoestrogens or Ginkgo biloba extract to stimulate antioxidant defenses, but the new results of a large trial failed to show efficacy of Ginkgo in reducing the incidence of AD or the rate of conversion from MCI [13]. While dietary vitamin E fed to healthy rats caused increases in brain vitamin E, no corresponding increase was seen in brain glutathione levels [14]. Also, feeding senescence-accelerated mice vitamins A + C, L-carnitine and lipoic acid produced no brain changes in glutathione, while feeding vitamins E, C, and 13 additional bioflavenoids, polyphenols, and carotenoids produced large increases in brain glutathione together with a marked reductive shift [15]. Complex mixtures of natural antioxidants including flavonoids and polyphenols found in fruits and berries have shown efficacy in animal models of aging [16] and AD [17,18]. Such a palatable, multimodal approach deserves more attention for prevention and treatment trials of AD.

4) Wrong target. Given ROS damage, but the failure of simple antioxidants to reverse damage, perhaps we need to consider whether we are aiming at the right target. A) The target of ROS damage may be detrimental when ROS signaling is essential to function. ROS signaling is required for processes as diverse as transcription factor activation [19,20], insulin signaling [21], endothelial function [22], and the long-term potentiation model of memory [23]. Hundreds of enzymes with redox-active cysteines (SH groups), including numerous cell surface receptors may require a certain redox potential for activity [24]. B) The target of ROS damage may distract us from the larger issue of redox control. We recently reported an oxidized redox potential in aging rat brain as well as neurons isolated and regenerated from aging brain [25] that could contribute to a larger effect on mitoenergetic failure [26–28]. Therefore, maintaining the glutathione intracellular redox buffer and the extracellular cysteine buffer at physiologically youthful redox potentials around -75 mV [10] may

slow aging and prevent the metabolic redox damage that occurs in AD [29]. C) The target of ROS damage may result in different responses dependent on a variety of co-morbidities. Instead of swamping one arm of biological systems with antioxidants for all subjects, we may need a dose-response targeted reductive shift in the metabolically oxidized redox potential seen in many aging individuals. We also need a better understanding of the cause of this metabolic shift.

Thus, vitamin E therapy has not convincingly failed yet, nor has it succeeded. Future trials are needed in light of the above and other considerations, with a special focus on individual monitoring of redox potential as introduced by Lloret et al. [8] to avoid toxicity and assess biomarkers of efficacy.

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