

# “Meta-Analysis, Metaphysics and Mythology”

## Scientific and Clinical Perspective on the Controversies Regarding Vitamin E for the Prevention and Treatment of Disease in Humans

Mark Houston, MD, MSc, FACP, FAHA\*  
 Editor-in-Chief, *Journal of the American Nutraceutical Association*  
 Associate Clinical Professor of Medicine  
 Vanderbilt University School of Medicine

The recent meta-analysis by Miller et al. in the *Annals of Internal Medicine* (2004; 142: Epub,<sup>1</sup>) has ignited a vociferous and polarized controversy about the clinical use of vitamin E in humans to reduce morbidity and mortality in various clinical disorders such as cardiovascular disease and cancer. It is time for scientists on both sides of this debate to accurately assess the facts regarding vitamin E. We still do not know about the efficacy, safety, clinical applications, proper dosage or type of vitamin E that should be utilized long-term by humans for prevention and treatment of disease. Epidemiological data from cross-sectional, case-controlled, prospective studies have shown a robust relationship between the consumption of antioxidant vitamins and minerals, or of foods with high concentrations of these nutrients, and reduction in the incidence of cancer and cardiovascular disease.<sup>2,3,4,5,6,7</sup> However, randomized, placebo-controlled, primary prevention trials using single and paired vitamins

and antioxidant micronutrients consumed in relatively high doses over prolonged time periods have produced conflicting results regarding clinical benefit.<sup>2,3,8,9,10,11,12,13</sup>

Miller's meta-analysis of 135,967 adults from 19 studies who took vitamin E in doses of 16.5 IU to 2000 IU daily vs. placebo for at least one year was seriously flawed. There exists a selection bias, improper recognition and definition of the type of vitamin E (synthetic versus natural alpha tocopherol) and inclusion of studies that used vitamin E in combination with other vitamins (over 50% of studies). Coupled with "statistical complexity" used in the meta-analysis, incorrect results, conclusions and recommendations have been reported. For example, 12 trials with fewer than 10 deaths were excluded from the analysis. The authors stated and assumed a priori that "...we anticipated that many small trials did not collect mortality data". This assumption may well have changed the entire results of the meta-analysis. On the other hand, this type of meta-analysis may be valid in other respects and provides some important information about the type and dose of vitamin E needed for clinical efficacy while avoiding adverse effects. It is possible that some scientists with biased views, preconceived notions and minimal scientific proof in human studies have also made some spurious health claims and ignored potential adverse effects regarding the synthetic d, l alpha-tocopherol (all RAC form), especially when administered in high doses. For example, when administered alone at high doses, all RAC vitamin E may have pro-oxidant effects,<sup>13,14</sup> inhibit glutathione S-transferases that are important in the detoxifi-

\* Correspondence:

Mark C. Houston, MD, MSc, FACP, FAHA  
 Director, Hypertension Institute and Vascular Biology  
 Saint Thomas Medical Group  
 Saint Thomas Hospital  
 4230 Harding Road, Suite 400  
 Nashville, Tennessee 37205  
 Phone: 615-297-2700 Fax: 615-269-4584  
 E-mail: [markh@ana-jana.org](mailto:markh@ana-jana.org)

cation of drugs and endogenous toxins,<sup>15</sup> increase bleeding tendency,<sup>2,16,17,18,19</sup> interfere with the metabolism of vitamin A and other vitamins<sup>17,20</sup> or produce other adverse effects in humans with long term administration.<sup>20</sup>

It should be noted that in many of the studies included in Miller's meta-analysis, the validity and safety of the supplemental vitamin E used could be suspect as has recently been reported by Consumer Labs.

The meta-analysis<sup>1</sup> suggested that doses over 150 IU per day of alpha tocopherol were associated with increased mortality from all causes. However, below this dose the all-cause mortality was slightly, but non-significantly, lower in the primary analysis; but in the secondary analyses based on 4-way data, the pooled risk difference for the low-dosage vitamin E trials was significant at a -33 per 10,000 persons ( $p = 0.021$ ). It was suggested in the meta-analysis that the concomitant use of other vitamins reduced all-cause mortality in both high and low dose vitamin E groups. Unfortunately, the authors do not distinguish between the synthetic and natural forms of alpha tocopherol in their analysis. The authors appear to be attempting to discredit all forms of vitamin E without expressly stating that their meta-analysis refers to only one form of the eight biologically different vitamin E's.<sup>22</sup> It is scientifically irresponsible to misrepresent the results of this study by implying that all forms of vitamin E at any dose may be harmful. The eight forms of vitamin E, four tocopherols (alpha, beta, gamma, delta), and four tocotrienols (alpha, beta, gamma, delta), are not inter-convertible in humans and all have different chemical structures, functions, biological activity and clinical effects.<sup>22</sup>

None of the 19 studies<sup>1</sup> included any of the other seven forms of vitamin E, and, in fact, most of them used the synthetic d, l alpha tocopherol form.<sup>23</sup> Alpha tocopherol is an OTC supplement available as natural d-alpha tocopherol (RRR-alpha tocopherol), which is only about 15% of total vitamin E found in food and synthetic d, l-alpha tocopherol (all RAC alpha tocopherol), which is a racemic mixture of the d and l forms of alpha tocopherol.<sup>23,24,25</sup> Gamma tocopherol is actually the most common form of vitamin E found in food (70%).<sup>25</sup>

It should be noted that of these eight stereoisomers, only one eighth of the total is RRR alpha tocopherol.<sup>22</sup> The other seven eighths are "counterfeit" vitamin E, where the stereoisomers do not exist in nature except in chemical synthesis.<sup>22</sup> This is similar to trans fats, which are due to the chemical hydrogenation (i.e., making of butter-like properties) of vegetable oil; trans fat is not found in the oil prior to the chemical process.

The d- form is the natural form of vitamin E and is more potent in its anti-inflammatory, antioxidant and cell signaling effects.<sup>23,24</sup> The natural d-alpha form has a potency of 1.5 IU/mg, whereas the synthetic d, l form is weaker at a potency of 1.1 IU/mg.<sup>24,26,27</sup> The d, l form is less expen-

sive to produce and is commonly used in clinical trials.<sup>26</sup> The chemically derived d, l form of vitamin E is composed of eight stereoisomers, has minimal vitamin E activity such as anti-inflammatory, antioxidant or cell-signaling effects.<sup>23,24,25</sup> It may actually interfere with the biological activity of d-alpha tocopherol and reduce HDL-cholesterol in humans potentially increasing CV risk.<sup>10</sup> Finally, in the ATBC study this form of vitamin E increased the risk of intra-cranial hemorrhage in patients who were smokers.<sup>2,18</sup> All of the clinical trials on cardiovascular disease prevention that have used the natural d-alpha tocopherol have shown beneficial or neutral effects on morbidity and mortality, but NONE, and none of these trials (CHAOS, HOPE, SPACE, TAPS, HATS, ASAP) showed negative effects, regardless of the dose used.<sup>2</sup> Even those studies that used the all racemic d, l mixture of vitamin E (ATBC, GISSI, PPP, HPS, VEAPS) had at worst neutral results.<sup>2</sup> Finally, there may also be biological differences in the acetate and succinate forms of alpha tocopherol if administered intravenously, but not if given orally (Personal Communication, Barrie Tan, Ph.D.).

Thus, Miller's meta-analysis<sup>1</sup> is suspect because it used some originally flawed studies, all of which used improper methodologies. In addition, the meta-analysis was very selective and excluded many important published vitamin E trials. If these other trials had been included, it may well have changed the results and conclusions. The meta-analysis also involved studies that enrolled primarily older adults with chronic diseases or conditions such as CVD, CHD, MI, smokers, CRF patients on hemodialysis, postmenopausal women on HRT, patients with Alzheimer's and Parkinson's disease, and excluded both older and younger healthier populations. Finally, the study assessed multi-vitamin combinations and/or other substances rather than vitamin E alone (beta carotene, selenium, vitamin C, zinc, copper, garlic, aspirin, fish oils, HRT and various pharmaceutical agents), reviewed data on only one form of vitamin E and had confounding variables that make the best statistical analysis questionable. For example, high doses of alpha tocopherol may also reduce intestinal absorption, cell membrane transport and utilization of other forms of vitamin E, especially gamma tocopherol.<sup>22,28,29</sup>

The hepatic alpha tocopherol transfer protein (TTP) has the greatest affinity for alpha tocopherol compared to gamma tocopherol (10-fold), and is crucial for the relative percent of transport of the various forms of vitamin E in the plasma lipoproteins.<sup>22,30,31</sup> Excessive intake of alpha tocopherol may reduce hepatic transport of other important forms of vitamin E.<sup>22,30,31</sup> This imbalance of the alpha tocopherol/gamma tocopherol levels in the plasma may have significant health consequences.<sup>31,32</sup> The natural abundance of vitamin E in diets suggest that the ratio of gamma/alpha tocopherol should be about 4:1.<sup>25,31</sup> Gamma tocopherol probably has a much more important role in human health than alpha tocopherol.<sup>8,31</sup> The ratio of

gamma/alpha tocopherol in plasma is a much more satisfactory index to measure compliance in clinical trials involving supplementation with alpha tocopherol.<sup>22,33</sup> Over 70% of the vitamin E in the human diet is gamma tocopherol, not alpha tocopherol.<sup>25</sup> The remaining 30% of the dietary vitamin E is 1/3 to 1/2 alpha tocopherol, 1/3 delta tocopherol, but with minimal beta tocopherol. The very basis of activity of alpha tocopherol was originally based on rodent infertility data, which may have little to do with biological activity or clinical effects in humans.<sup>27</sup>

The clinical use of high doses of synthetic beta-carotene has questionable safety and efficacy.<sup>2,18</sup> Beta-carotene has been reported to increase lung cancer in smokers or in those who consume alcohol according to the ATBC study<sup>18</sup> and increase mortality according to HPS.<sup>2</sup> It is important to remember that vitamin E, like beta carotene and other nutrients, does not work alone, but rather symphonically with its cousins.<sup>13,34</sup> To study one species of the family of vitamin E often results in unintended consequences such as those found in the beta carotene smokers study (ATBC).

There may be many reasons for the variable results and conclusions in the clinical trials for the role of vitamin E, other antioxidants and vitamins in the prevention and treatment of human disease. These reasons include inappropriate endpoints or their definitions, inappropriate assessment of the endpoints with clinical or laboratory tools, inappropriate adjuvant therapies, inappropriate patient types (biochemical and oxidative stress inclusion criteria, genetic polymorphisms), incorrect or ineffective type, dose, timing or combinations of vitamins or improper duration of the study.

It would be tempting to conclude that Miller's study provides definitive conclusions and recommendations, but this is not the case. His meta-analysis and the studies that were used in the meta-analysis are not statistically and methodologically sound enough to make such sweeping conclusions. I would offer these recommendations based on present data:

**One:** It would seem prudent and advisable to avoid consumption of synthetic racemic d, l- alpha tocopherol (all-RAC alpha tocopherol) at any dose, at this time, until further human clinical trials demonstrate clinical efficacy with no adverse effects.

**Two:** It would seem prudent and advisable to avoid consumption of single high doses (over 400 IU daily) of natural d-alpha tocopherol (RRR-alpha tocopherol). The level of 150 IU per day suggested as the "upper limit" at which total mortality begins to increase in the meta-analysis is probably not due to any direct toxicity of d-alpha tocopherol, but rather the absence of other tocopherols and/or tocotrienols due to reduced intestinal absorption or decreased hepatic alpha TTP incorporation.

**Three:** Use a mixed tocopherol complex with the correct balance of all four forms of tocopherols. The gamma/alpha tocopherol ratio should be approximately 4:1.

The beta and delta balance should simulate the balance found in natural food as noted previously. This vitamin mixture should be taken each morning at least 12 hours before any ingestion of tocotrienols to avoid interference with intestinal absorption. More clinical studies are needed to determine the correct balance and doses.

**Four:** Use a mixed tocotrienol complex with the correct balance of tocotrienols that simulates that found in natural food. As with most other areas of nutritional science, this should be almost intuitive and is backed by some sound science. Preliminary evidence suggests that the delta and gamma tocotrienol have more biological activity and clinical benefits compared to alpha and beta tocotrienol. This vitamin mixture should be taken at night at least 12 hours after the tocopherol complex. More clinical studies are needed to determine the correct balance and doses.

**Five:** Take the mixed vitamin E complexes above with other antioxidants (vitamin C, mixed carotenoids, B-vitamins, vitamin D, CoQ-10, R- lipoic acid, etc.) to provide nutritional balance, improve total antioxidant effects, avoid potential pro-oxidant effects and enhance recycling of vitamins. Combine this vitamin regimen with 8-10 servings of fresh fruits and vegetables per day with attention to foods with good balanced vitamin E content such as soybeans and wheat germ. Objective testing to determine nutritional deficiencies, oxidative stress profile, antioxidant defenses/capacity and inflammatory status are probably advisable and necessary to prescribe an optimal nutritional and nutraceutical-based regimen for patients.

**Six:** Design and implement scientifically valid prospective clinical trials on the various forms of vitamin E to provide accurate scientific rationale to the type, dose, timing and combinations that improve morbidity and mortality in humans.

The recent SU.VI.MAX study<sup>35</sup> of 13,017 French adults, aged 35-60 years, is an example of a more scientifically valid trial. After 7.5 years of low dose antioxidant supplementation, the total cancer incidence and all cause mortality in men was reduced by 31% and 37% respectively ( $p = 0.004$ ). In this study 30 mg of d-alpha tocopherol (45 IU of d-alpha tocopherol) was used in conjunction with ascorbic acid, selenium, zinc and beta carotene.

Meta-analysis, like meta-physics, is not always scientifically valid, but there may be some hidden truths for the wise observer. One should ask why in this meta-analysis the authors failed to distinguish between studies using the different forms of vitamin E prior to performing their analysis. Studies should always attempt to eliminate as many sources of bias as possible. If an adverse effect occurred only with the synthetic d, l alpha tocopherol, then combining the studies all together with both synthetic and natural vitamin E introduces an obvious source of error. Thus, including different forms of vitamin E in this meta-analysis

clearly introduces a confounding variable leading to potentially erroneous results and conclusions. A re-analysis that separates out this confounding variable should be done.

Opinions and interpretation of science are often blinded by personal prejudice and/or preconceived notions. We should all remember the wise words of Socrates at the Oracle of Delphi when asked what the definition of knowledge and wisdom were, "That which I do not know, I know that I do not know". We have much yet to learn about vitamin E and should keep our minds open as the scientific story of this contentious nutrient continues to unfold.

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