2100 Maury Street • Des Moines, Iowa, USA 50317 • tel: 515.559.5100 • www.kemin.com

AREDS2 SUMMARY: INCLUSION OF LUTEIN AND ZEAXANTHIN ARE THE NEW STANDARD OF CARE FOR AMD PATIENTS

Tatania K. Emmick

KEY CONCLUSIONS

- *AREDS2 results support replacing beta-carotene with 10 mg lutein and 2 mg zeaxanthin to improve the safety and efficacy of AREDS formulations.*
- *The main effect of lutein/zeaxanthin demonstrated a 10% reduction in risk of progression to advanced AMD (AAMD).*
- *There was an 18% reduction in the risk of progression to AAMD with lutein/zeaxanthin in head-to-head comparison with beta-carotene, when used in the AREDS formulation.*
- *AREDS2 results showed a 26% reduction in the risk of progression to AAMD for lutein/zeaxanthin beyond the effects of AREDS supplements in subjects in the lowest quintile of lutein and zeaxanthin dietary intake.*

INTRODUCTION

Age-related eye conditions are estimated to triple over the next 15 years due (*1*) to increasing life expectancies and an aging population. This includes devastating and debilitating eye conditions, such as age-related macular degeneration (AMD). AMD is a degenerative disease that causes progressive loss of central vision, robbing people of their independence. AMD is the leading cause of blindness among people of European descent.(*2*) In the United States alone, it is estimated that AMD is the cause of 50% of the visual impairment and 20% of the blindness among Caucasians.(*3*) In 2004, the direct medical cost of AMD treatment was estimated to be $575 million in the U.S.(*4*) It is estimated that nearly 8 million individuals in the U.S. have AMD with almost 2 million having advanced AMD. This number is expected to double by 2020.(*2*) In addition to AMD, the prevalence of age-related cataract is increasing with an estimated 30.1 million Americans to be affected by 2020.(*5*) It was this projected increase on the socioeconomic burden of age-related eye conditions as well as the existing body of evidence for important nutrients like lutein, zeaxanthin and omega-3 fatty acids on eye health that prompted The National Eye Institute (NEI) of the U.S. National Institutes of Health (NIH) to initiate the Age Related Eye Disease Study 2 (AREDS2). The AREDS2 study was designed with the objective of studying nutritional interventions that might slow the progression from intermediate to advanced AMD (AAMD).

Epidemiological data provide much of the rationale for examining the potential effects of lutein and zeaxanthin on the development of advanced AMD (*6, 7*) and cataract.(*8-11*) Additionally, at the time the original AREDS was initiated, researchers were interested in evaluating the effects of lutein and zeaxanthin on AMD but included beta-carotene in the formulation because lutein and zeaxanthin were not yet commercially available.

Previous reports linking diets low in lutein and zeaxanthin with increased AMD (*6, 7, 12*) and cataract (*9, 11, 13, 14*) risk are further supported by the results of this landmark study.(*15-17*)

THE AGE-RELATED EYE DISEASE STUDY 2

Design. AREDS2 was a five year, multi-center, randomized trial designed to assess whether adding macular xanthophylls (10 mg FloraGLO® Lutein and 2 mg zeaxanthin) and/or long-chain omega-3 fatty acids, 350 mg

docosahexaenoic acid (DHA) and 650 mg eicosapentaenoic acid (EPA) daily to the original AREDS formulation would further slow the progression to AAMD in patients at high risk for developing AAMD. An additional goal of the study was to evaluate the effect of eliminating beta-carotene, lowering the zinc dose, or both in the AREDS formulation.

The study was conducted from 2006-2012 and enrolled 4203 subjects with intermediate to advanced AMD characterized as having either bilateral large drusen or advanced AMD in one eye. Subjects were 55-80 years of age (median 74 years). The study was conducted at 82 different clinical sites. Patients were randomized into four primary treatments: lutein/zeaxanthin (collectively known as xanthophylls), DHA/EPA, combined xanthophyll and DHA/EPA treatment, or placebo. All but 19 subjects received some variation of the AREDS supplement, so there was no true placebo group. Subjects that agreed to participate in an optional secondary randomization were assigned to receive one of four AREDS-type supplements as shown in Table 1. If the participant was a current smoker or a former smoker that had quit within the last year prior to study participation, they were randomized into one of the two arms without beta-carotene. See Figure 1 for a schematic overview of the study's design.

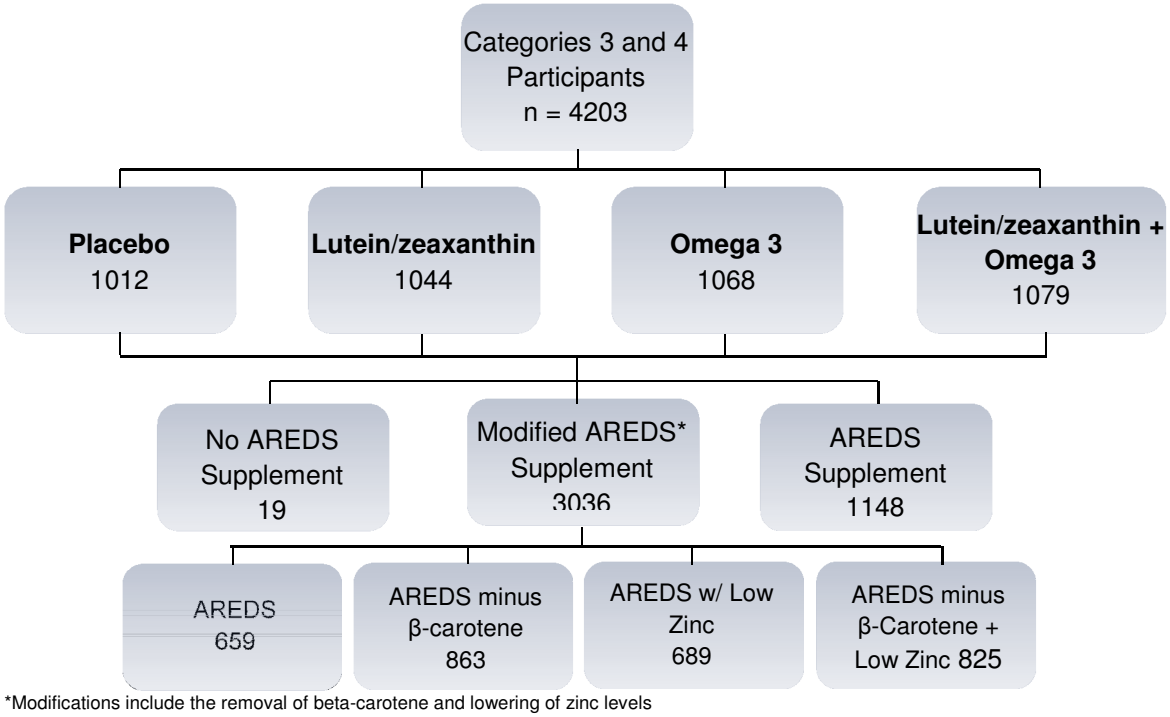

Figure 1. Graphical representation of the treatment groups into which subjects were randomized for AREDS2.

Table 1. AREDS formulations in the secondary randomization

Treatment Group	Vitamin C	Vitamin E	β-carotene	Zinc Oxide	Cupric Oxide
AREDS	500 mg	400 IU	15 mg	80 mg	2 mg
AREDS minus β-carotene	500 mg	400 IU	0 mg	80 mg	2 mg
AREDS with low zinc	500 mg	400 IU	15 mg	25 mg	2 mg
AREDS minus β-carotene + low zinc	500 mg	400 IU	0 mg	25 mg	2 mg

2100 Maury Street • Des Moines, Iowa, USA 50317 • tel: 515.559.5100 • www.kemin.com

RESULTS

Participant Characteristics. Of the 4203 participants enrolled in AREDS2, 97% were white, 57% were female, 13% had diabetes, 50% were former smokers and 7% were current smokers. With regards to AMD status, 65% of the subjects had large bilateral drusen and 35% had advanced AMD in one eye. Nearly 90% of subjects were taking Centrum Silver (with 275 µg FloraGLO Lutein) in addition to study supplements. Some patients admitted to taking lutein/zeaxanthin (3%) or DHA/EPA (11%) on their own. Compliance was very good for a study of this size with 84% of the subjects taking at least 75% of the study supplements. Additionally, attrition rates were very low with only 3% lost to follow-up and 9% died during the course of the study.

A well-nourished cohort. The AREDS2 cohort was very well-nourished as compared to the general population and participants in other studies. The serum levels for lutein and zeaxanthin at baseline were found to be significantly higher than that of the general population age 60+ as sampled in the National Health and Nutrition Examination Survey 2005-2006 (NHANES). In a study evaluating the carotenoid intake of 18 cohorts, the AREDS2 participants' dietary intake of lutein and zeaxanthin was exceeded in only two of these study cohorts as shown in Figure 2.(*18*)

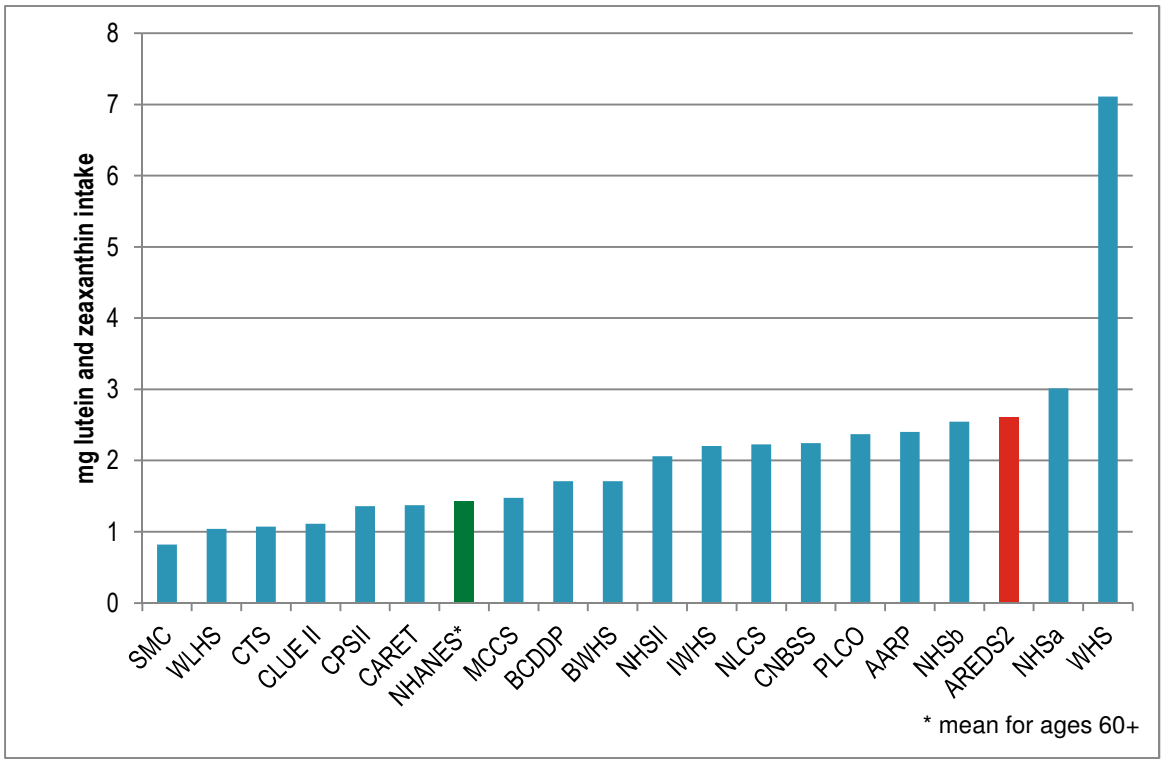

Figure 2. Graphical representation of lutein and zeaxanthin intake of the AREDS2 cohort in comparison with NHANES 2007-2008(*19*) and 18 cohort studies* showing that AREDS2 subjects were better nourished than all but two cohorts.

* SMC – Swedish Mammography Cohort, WLHS – Women's Lifestyle and Health Study, CTS – California Teachers Study, CLUE II – Campaign Against Cancer and Heart Disease, CPSII - Cancer Prevention Study II Nutrition Cohort, CARET – Beta-Carotene and Retinol Efficacy Trial, MCCS – Melbourne Collaborative Cohort Study, BCDDP – Breast Cancer Detection Demonstration Project Follow-up Study, BWHS – Black Women's Health Study, NHSII - Nurses' Health Study II, IWHS – Iowa Women's Health Study, NLCS – Netherlands Cohort Study, CNBSS – Canadian National Breast Screening Study, PLCO – Prostate, Lung, Colorectal, and Ovarian Cancer Screening Trial, AARP – NIH-AARP Diet and Health Study, NHSb – Nurses' Health Study (b), NHSa – Nurses' Health Study (a), WHS – Women's Health Study

Analysis of serum carotenoids indicate competitive absorption when beta-carotene was administered with lutein/zeaxanthin: There was a 2-fold increase in serum lutein levels in the lutein/zeaxanthin supplemented groups. Year 5 serum levels of lutein in those randomized to receive lutein/zeaxanthin and AREDS formulations with beta-carotene (39.1 µg/dL) was significantly lower than in those receiving lutein/zeaxanthin and AREDS formulations without beta-carotene (46.9 µg/dL, p = 0.02) suggesting competitive absorption of carotenoids may have occurred.

Primary outcome. The efficacy of treatments was evaluated by determining the hazard ratios (HR) with a hazard ratio of less than one meaning that the treatment was favored for the given outcome. The primary analysis tested whether adding FloraGLO Lutein and zeaxanthin, DHA/EPA, or a combination of the two to the AREDS formulation reduced the risk of progression to advanced AMD by an additional 25% as compared to study subjects taking the original AREDS supplement, which was the study control arm. The data did not demonstrate a significant reduction in progression to advanced AMD in any of the three treatment arms as compared to the control group. Lutein/zeaxanthin had a HR of 0.90 (p = 0.12) and lutein/zeaxanthin + DHA/EPA had a HR of 0.89 (p = 0.10) whereas DHA/EPA alone had a HR of 0.97 (p = 0.70).

Main effect of lutein and zeaxanthin. The analysis of the main effect of lutein/zeaxanthin compared the rate of progression for the two study arms that were supplemented with lutein/zeaxanthin (L/Z and L/Z/DHA/EPA) to the two study arms that were not supplemented with lutein/zeaxanthin (placebo and DHA/EPA). This comparison showed an additional 10% reduction in risk to progression of advanced AMD with lutein and zeaxanthin (HR of 0.90, p = 0.04). This type of comparison was also conducted for DHA/EPA vs no DHA/EPA, Low Zinc vs High Zinc and No Beta-carotene vs Beta-carotene. No other hazard ratios in these other test groups were significant.

Effect of dietary intake of lutein and zeaxanthin. Stratification of the results by dietary intake of lutein and zeaxanthin resulted in some very notable findings. For individuals in the lowest quintile of lutein and zeaxanthin dietary intake (median 0.7 mg per day), comparison of lutein/ zeaxanthin vs. no lutein/ zeaxanthin showed an additional 26% reduction in the risk of progression to AAMD beyond the effects of an AREDS supplement (HR of 0.74, p=0.01).

Findings with beta-carotene. Comparison of participants assigned to receive lutein/zeaxanthin with AREDS supplements without beta carotene versus an AREDS supplement with beta carotene resulted in an 18% reduction in the risk of progression to advanced AMD (HR = 0.82, p=0.02). When evaluating safety outcomes, it was found that there was an increased risk of lung cancer with beta-carotene supplementation. Subjects taking beta-carotene had a higher incidence of lung cancer (23 cases, 2%) compared to those subjects not taking beta-carotene (11 cases, 0.9%) (p = 0.04). Thirty-one (91%) of those that developed lung cancer were former smokers who quit smoking > 1 year prior to randomization. Lutein/zeaxanthin were not associated with an increased risk of lung cancer.

Cataract results. Subjects in the lowest quintile of lutein and zeaxanthin intake exhibited the greatest benefit from adding lutein/zeaxanthin to AREDS supplementation. When comparing lutein/zeaxanthin vs. no lutein/zeaxanthin, these subjects saw a significant reduction in risk of progression to cataract surgery, any cataract or any severe cataract of 32%, 30% and 36%, respectively (p < 0.05).

CONCLUSION

Based on the results of AREDS2, the NEI recommends adding 10 mg lutein and 2 mg zeaxanthin and eliminating beta carotene in the original AREDS formula to provide a safer, more efficacious supplement.(*20*) Results showed that daily supplementation with a modified AREDS formulation, where 15 mg beta carotene was replaced with 10 mg FloraGLO Lutein and 2 mg zeaxanthin, reduced progression to advanced AMD by 18% when compared to the original AREDS

supplement with beta carotene. Additionally, when comparing subjects taking an AREDS formula with lutein and zeaxanthin to subjects taking an AREDS formula with no lutein and zeaxanthin, an even greater reduction in risk of progression to advanced AMD of 26% was seen in study subjects with the lowest intake of lutein and zeaxanthin in their diet. The lutein and zeaxanthin dietary intake of the lowest quintile is more representative of the general US population, where the dietary intake of lutein and zeaxanthin is typically less than 1 mg per day. This amount is well below the 10 mg lutein and 2 mg zeaxanthin that the study has proven to be effective. In conclusion, the AREDS2 results corroborate previous epidemiological data linking a high dietary intake of lutein and zeaxanthin with a reduction of risk of AMD and cataract, and additionally support the safety and benefits of substituting 10 mg of lutein and 2 mg of zeaxanthin for beta carotene in AREDS formulations as the new standard of care for AMD patients.

REFERENCES

1. Prevent Blindness America. (2008 Update to the 4th Ed.) Vision Problems in the US: Prevalence of Adult Vision Impairment and Age-Related Eye Disease in America.
2. Friedman, D. S., O'Colmain, B. J., Munoz, B., Tomany, S. C., McCarty, C., de Jong, P. T., Nemesure, B., Mitchell, P., and Kempen, J. (2004) Prevalence of age-related macular degeneration in the United States, *Arch Ophthalmol 122*, 564-572.
3. Congdon, N., O'Colmain, B., Klaver, C. C., Klein, R., Munoz, B., Friedman, D. S., Kempen, J., Taylor, H. R., and Mitchell, P. (2004) Causes and prevalence of visual impairment among adults in the United States, *Archives of ophthalmology 122*, 477-485.
4. Rein, D. B., Zhang, P., Wirth, K. E., Lee, P. P., Hoerger, T. J., McCall, N., Klein, R., Tielsch, J. M., Vijan, S., and Saaddine, J. (2006) The economic burden of major adult visual disorders in the United States, *Arch Ophthalmol 124*, 1754-1760.
5. Congdon, N., Vingerling, J. R., Klein, B. E., West, S., Friedman, D. S., Kempen, J., O'Colmain, B., Wu, S. Y., and Taylor, H. R. (2004) Prevalence of cataract and pseudophakia/aphakia among adults in the United States, *Arch Ophthalmol 122*, 487-494.
6. Seddon, J. M., Ajani, U. A., Sperduto, R. D., Hiller, R., Blair, N., Burton, T. C., Farber, M. D., Gragoudas, E. S., Haller, J., Miller, D. T., and et al. (1994) Dietary carotenoids, vitamins A, C, and E, and advanced age-related macular degeneration. Eye Disease Case-Control Study Group, *JAMA 272*, 1413-1420.
7. SanGiovanni, J. P., Chew, E. Y., Clemons, T. E., Ferris, F. L., 3rd, Gensler, G., Lindblad, A. S., Milton, R. C., Seddon, J. M., and Sperduto, R. D. (2007) The relationship of dietary carotenoid and vitamin A, E, and C intake with age-related macular degeneration in a case-control study: AREDS Report No. 22, *Arch Ophthalmol 125*, 1225-1232.
8. Delcourt, C., Carriere, I., Delage, M., Barberger-Gateau, P., and Schalch, W. (2006) Plasma lutein and zeaxanthin and other carotenoids as modifiable risk factors for age-related maculopathy and cataract: the POLA Study, *Invest Ophthalmol Vis Sci 47*, 2329-2335.
9. Moeller, S. M., Voland, R., Tinker, L., Blodi, B. A., Klein, M. L., Gehrs, K. M., Johnson, E. J., Snodderly, D. M., Wallace, R. B., Chappell, R. J., Parekh, N., Ritenbaugh, C., and Mares, J. A. (2008) Associations between age-related nuclear cataract and lutein and zeaxanthin in the diet and serum in the Carotenoids in the Age-Related Eye Disease Study, an Ancillary Study of the Women's Health Initiative, *Arch Ophthalmol 126*, 354-364.
10. Vu, H. T., Robman, L., Hodge, A., McCarty, C. A., and Taylor, H. R. (2006) Lutein and zeaxanthin and the risk of cataract: the Melbourne visual impairment project, *Invest Ophthalmol Vis Sci 47*, 3783-3786.
11. Brown, L., Rimm, E. B., Seddon, J. M., Giovannucci, E. L., Chasan-Taber, L., Spiegelman, D., Willett, W. C., and Hankinson, S. E. (1999) A prospective study of carotenoid intake and risk of cataract extraction in US men, *Am J Clin Nutr 70*, 517-524.
12. Seddon, J. M., Reynolds, R., and Rosner, B. (2010) Associations of smoking, body mass index, dietary lutein, and the LIPC gene variant rs10468017 with advanced age-related macular degeneration, *Mol Vis 16*, 2412-2424.
13. Chasan-Taber, L., Willett, W. C., Seddon, J. M., Stampfer, M. J., Rosner, B., Colditz, G. A., and Hankinson, S. E. (1999) A prospective study of vitamin supplement intake and cataract extraction among U.S. women, *Epidemiology 10*, 679-684.
14. Jacques, P. F., Chylack, L. T., Jr., Hankinson, S. E., Khu, P. M., Rogers, G., Friend, J., Tung, W., Wolfe, J. K., Padhye, N., Willett, W. C., and Taylor, A. (2001) Long-term nutrient intake and early age-related nuclear lens opacities, *Arch Ophthalmol 119*, 1009-1019.

15. Chew, E. Y., Clemons, T., SanGiovanni, J. P., Danis, R., Domalpally, A., McBee, W., Sperduto, R., and Ferris, F. L. (2012) The Age-Related Eye Disease Study 2 (AREDS2): study design and baseline characteristics (AREDS2 report number 1), *Ophthalmology 119*, 2282-2289.
16. AREDS2 Research Group. (2013) Lutein + Zeaxanthin and Omega-3 Fatty Acids for Age-Related Macular Degeneration: The Age-Related Eye Disease Study 2 (AREDS2) Randomized Clinical Trial, *JAMA*, In Press.
17. AREDS2 Research Group. (2013) Lutein/Zeaxanthin for the Treatment of Age-Related Cataract: AREDS2 Randomized Trial Report No. 4, *JAMA Ophthalmol*, In Press.
18. Zhang, X., Spiegelman, D., Baglietto, L., Bernstein, L., Boggs, D. A., van den Brandt, P. A., Buring, J. E., Gapstur, S. M., Giles, G. G., Giovannucci, E., Goodman, G., Hankinson, S. E., Helzlsouer, K. J., Horn-Ross, P. L., Inoue, M., Jung, S., Khudyakov, P., Larsson, S. C., Lof, M., McCullough, M. L., Miller, A. B., Neuhouser, M. L., Palmer, J. R., Park, Y., Robien, K., Rohan, T. E., Ross, J. A., Schouten, L. J., Shikany, J. M., Tsugane, S., Visvanathan, K., Weiderpass, E., Wolk, A., Willett, W. C., Zhang, S. M., Ziegler, R. G., and Smith-Warner, S. A. (2012) Carotenoid intakes and risk of breast cancer defined by estrogen receptor and progesterone receptor status: a pooled analysis of 18 prospective cohort studies, *Am J Clin Nutr 95*, 713-725.
19. NHANES. (2007-2008) What We Eat in America, *http://www.ars.usda.gov/SP2UserFiles/Place/12355000/pdf/0708/Table_1_NIN_GEN_07.pdf Accessed May 22, 2013*.
20. National Institutes of Health National Eye Institute. (2013) NIH study provides clarity on supplements for protection against blinding eye disease, *Press Release* *http://www.nei.nih.gov/news/pressreleases/050513.asp*.