

A Double Blind Placebo Study showing the Effect of lifepak nano supplementation on Skin Carotenoid Scores

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Carsten R. Smidt, Ph. D., FACN, Angela Mastaloudis, Ph. D., Stephen Poole, B.S.
Pharmanex Research Institute, Provo, UT.

Skin Carotenoid Scores (SCS) are a biomarker of overall antioxidant status. This study used Raman Spectroscopy to examine the ability of lifepak nano to increase SCS as compared to a placebo. Lifepak nano increased SCS in all subjects, and was especially effective in raising scores that were above 30,000 Raman Intensity Counts at baseline. The findings of this study suggest excellent bioavailability of lifepak nano's unique CR-6 Liponutrient delivery system including cutting edge nano-carotenoids in fish oil.

Introduction

Carotenoids are an important group of phytonutrients which have been designated as the best biological marker of fruit and vegetable consumption, as well as the best indicator of overall antioxidant status (Institute of Medicine, 2000; Svilaas, 2004). Epidemiological and clinical studies substantiate the protective effects of carotenoids in many areas of health, including cardiovascular, skin and eye health (Smidt, 2005a). As an integral part of the antioxidant network, carotenoids preserve other antioxidants from free radical destruction, thus strengthening the entire antioxidant defense system.

Because of their central role to health, and their ability to accurately predict overall antioxidant status, many techniques have been developed to assess carotenoid levels in human tissues. Previously, blood serum carotenoids have been viewed as the gold standard to assess tissue carotenoid concentrations; recently however, the well established method of Raman Spectroscopy (RS) has been adapted to accurately and non-invasively measure carotenoid concentrations in living human skin. Skin carotenoid concentrations are reported as Raman Intensity Counts and commonly referred to as Skin Carotenoid Score (SCS). The higher the score, the higher the concentration of carotenoid molecules detected at the site of measurement (Ermakov, 2001; Ermakov, 2004; Hata, 2000; Zidichouski, 2004).

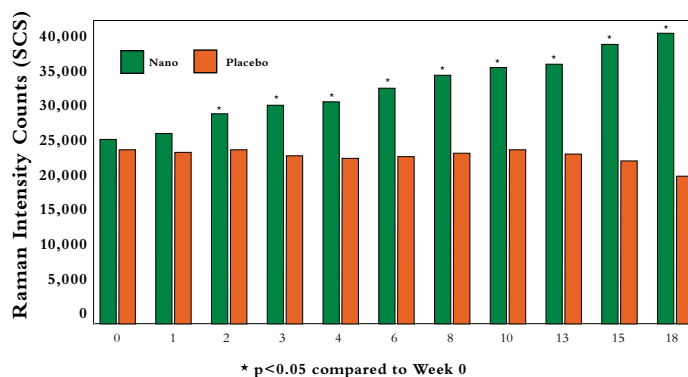
This study used Raman Spectroscopy to assess changes in Skin Carotenoid Scores (SCS) over eighteen weeks of supplementation with lifepak nano or placebo. Lifepak nano is a comprehensive dietary supplement containing vitamins, minerals, and over sixty antioxidants, including a unique delivery system known as CR-6 Liponutrient softgels containing fish oils (omega-3s) and highly absorbable carotenoids which have been enhanced through the application of nanotechnology. Nanotechnology is the highly

sophisticated technique of manipulating single molecules (no larger than 100 nanometers), to enhance their functional behavior. The application of nanotechnology employed by Pharmanex is a mono-molecular encapsulation in which individual nutrients are embedded into single cyclodextrin molecules.

Rationale

Pharmanex has conducted multiple studies to show the ability of various antioxidant preparations to increase SCS, as a biomarker of overall antioxidant status (Smidt, 2002; Smidt, 2005b). As a follow-up to these studies, this study seeks to confirm that the novel preparation enhanced by nanotechnology (lifepak nano), increases SCS as anticipated. Previous nanoized nutrients have shown superior absorption over non-nano preparations (Craft, 2005). The purpose of this study is to demonstrate lifepak nano's ability to increase SCS. Traditional carotenoids are poorly absorbed, and large scale surveys show average diets do not contain sufficient levels of these important antioxidants (Brown, 2004; Block, 1991). Lifepak nano's unique CR-6 Liponutrient delivery system will help to offset low consumption of fruits and vegetables and poor absorption, thus providing more protective benefits.

Figure 1: Average SCS Increase with lifepak nano



This study was conducted by Pharmanex scientists. The study authors, Carsten R. Smidt and Angela Mastaloudis, are employees of Pharmanex, a division of Nu Skin Enterprises, Inc. Pharmanex produces and distributes dietary supplement products, including lifepak nano.

Methods and Materials

Fifty two subjects between 18 and 65 years of age qualified for study participation. Food frequency and health history questionnaires were used to screen for inclusion criteria, and the Pharmanex BioPhotonic Scanner was used to assess skin carotenoid levels. Individuals taking antioxidant supplements, having high exposure to sunlight or tanning bed use, pregnant women, or individuals using sunless tanning products were excluded from the study. All subjects were healthy non-smokers consuming typical U.S. diets containing less than five daily servings of fruits and vegetables, and had baseline Skin Carotenoid Scores between 13,000 and 35,000 Raman Intensity Counts. Participants were instructed to maintain their usual dietary and exercise habits throughout the duration of the study. Subjects (n=52) meeting study criteria were randomly assigned in a double blind manner to one of two groups—lifepak nano (n=27) or placebo (n=25). Additional subject statistics are available in table 1.

Table 1: Subject Statistics

	Lifepak nano	Placebo
Subject per group	(N=20)	(N=22)
Age	40 ± 8.6	35 ± 9.7
Height (cm)	175.3 ± 10.2	172.7 ± 12.7
Weight (kg)	77.6 ± 19.5	80.3 ± 21.3
BMI	26 ± 7	27 ± 5
Baseline	25,599 ± 6,199	23,459 ± 5,686
F/V Intake (servings/day)	2.58 ± .581	2.57 ± .660

Treatments

Study participants were randomly assigned to receive either lifepak nano or a placebo. Subjects were instructed to take lifepak nano or placebo twice daily, i.e., with their morning and evening meals. Additional nutritional information of the study treatments are available below, and a full list of lifepak nano nutrients is available at www.lifepaknano.com.

lifepak nano

Lifepak nano is a comprehensive dietary supplement containing all essential micro and macronutrients including vitamins, minerals, broad spectrum antioxidants to support the entire antioxidant network. A delivery of CR-6 Liponutrient softgel capsules with fish oil and nanoized carotenoids enhances the absorption of nutrients that are known to have poor absorption. Carotenoids cling together in the digestive tract causing poor bio-availability; however nano-encapsulated carotenoids allow for complete molecular dispersion, thus increasing the surface area of these important nutrients and maximizing their contact with the absorptive lining of the digestive tract.

Each serving of lifepak nano contains five capsules (dry ingredients) and two CR-6 Liponutrient softgel capsules (liquid ingredients, including omega-3 fatty acids and nanoized carotenoids).

Placebo

The placebo taken twice daily included: two powder capsules; and two softgel capsules with omega-3 fatty acids (containing no carotenoid antioxidant).

Statistical Analyses

Data are expressed as the mean ± SD (n=42 subjects). Analysis of variance was used to detect statistically significant between and within subject effects. An unpaired t-test was used to analyze differences between sexes with regard to subject characteristics (i.e. age, height, weight). Statistics were calculated using The SPSS System (SPSS Inc. Chicago, IL).

Results and Discussion

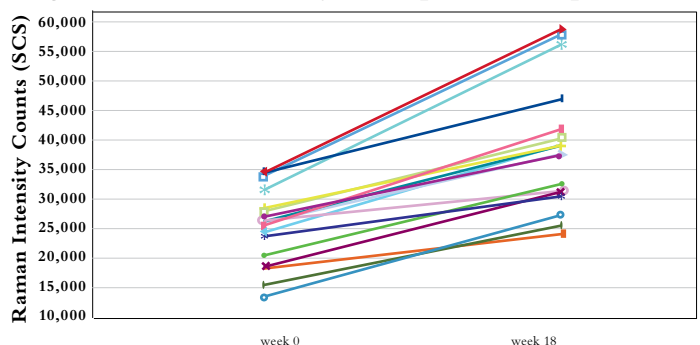
Forty-two subjects completed the study. A summary of subject characteristics are described in table 1. In the lifepak nano group seven subjects were disqualified for non-compliance, and three subjects were dropped from the placebo group also due to non-compliance. All participants in the lifepak nano group experienced dramatic increases in Skin Carotenoid Scores (figure 2). Consumption of lifepak nano increased Skin Carotenoid Scores much faster than expected, showing statistical significance within 2 weeks (figure 1). Skin Carotenoid Scores of the lifepak nano group increased an average of 17,757 +/- 10,113 Raman Intensity Counts in eighteen weeks compared to placebo. Which had no effect on Scanner score at any time (figure 1).

Throughout the eighteen weeks there was a continual increase in scanner score of the lifepak nano group, with no apparent plateau effect (figure 1).

Conclusion

Our findings indicate that lifepak nano effectively increases SCS, a biomarker of overall antioxidant status. All subjects in the lifepak nano group showed dramatic increases in Skin Carotenoid Scores (figure 2), with no signs of plateauing even at the end of the study (figure 1). Carotenoid antioxidant levels increased faster than expected, showing statistically significant increases in two weeks for the lifepak nano group, with no change in the placebo group. While lifepak nano is effective for all subjects, it was especially effective for subjects with scores that were above 30,000 Raman Intensity Counts at baseline (data not shown). We conclude that lifepak nano delivers significant benefit by increasing tissue antioxidants as measured by Biophotonic Scanner.

Figure 2: Individual Subject Response to lifepak nano



References

Block, G. (1991) Dietary guidelines and the results of food consumption surveys. *American Journal of Clinical Nutrition*. 53, 356S-357S.

Brown, MJ, Ferruzzi, MG, Nguyen, ML, Cooper, DA, Eldridge, AL, Schwartz, SJ and White, WS Carotenoid bio-availability is higher from salads ingested with full-fat than with fat-reduced salad dressings as measured with electrochemical detection *AJCN* 2004;80:396-403.

Craft, N.E., Tucker, R.T., Chitchumroonchokchai, C., Failla, M.L., Bhagavan, H.N. Assessment of Coenzyme Q10 Bioavailability Using a Coupled in vitro Digestion/Caco-2 Human Intestinal Cell Model. *FASEB Journal*. 2005; 19(4): Abstract #281.6, A449.

Ermakov, I.V. et al. Noninvasive selective detection of lycopene and beta-carotene in human skin using Raman spectroscopy. *J Biomed Opt*. 2004 Mar;9(2):332-8.

Ermakov, I.V., Ermakova, M.R., McClane, R.W., Gellermann, W. (2001). Resonance Raman detection of carotenoid antioxidants in living human tissues. *Optics Letters* 26, 1179-1181.

Hata, T.R. et al, Non-invasive raman spectroscopic detection of carotenoids in human skin. *J Invest Dermatol*. 2000 Sep;115(3):441-8.

Institute of Medicine, Food and Nutrition Board. Daily Reference Intakes for Vitamin C, Vitamin E, Selenium, and Carotenoids. pg 325. Washington D.C.: National Academy Press, 2000.

Smidt, C.R. Ph.D., FACN, Effect of LifePak Supplementation on Antioxidant Status Using BioPhotonic Raman Spectroscopy. Pharmanex in-house Study. 2002.

Smidt, C.R., Burke, D.S. Nutritional Significance and Measurement of Carotenoids. *Curr Topics Nutraceut. Res*. 2(2):79-91, 2004.

Smidt, C.R., Mastaloudis, A., Effect of G3 and Other Juices on Antioxidant Network Status As Measured by Raman Spectroscopy. Pharmanex in-house study, October 2005.

Svilaas A, Sakhi A.K., Andersen L.F., Svilaas T, Strom E.C., Jacobs D.R. Jr, Ose L, Blomhoff R. Intakes of antioxidants in coffee, wine, and vegetables are correlated with plasma carotenoids in humans. *J Nutr*. 2004 Mar;134(3):562-7.

Zidichouski J.A., Poole S.J., Gellermann W, and Smidt C.R., Clinical Validation of a Novel Raman Spectroscopic Technology to Non-Invasively Assess Carotenoid Status in Humans. *J. Am. Coll. Nutr*. 23(5): 468, 2004