

# Effect of LifePak® Supplementation on Antioxidant Status Using BioPhotonic Raman Spectroscopy

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A new, non-invasive BioPhotonic Raman spectroscopy method was used to assess the antioxidant efficacy of the multi-nutrient supplement LifePak® in 25 healthy volunteers for 12 weeks. Raman spectroscopy measures skin (palm) carotenoids as an important indicator of the antioxidant health status. BioPhotonic skin carotenoid readings increased significantly from a baseline of 18,828 units to 32,175 units at the end of the study. Although some individual variability was observed, all subjects experienced an increase in BioPhotonic skin response. Fruit and vegetable consumption was monitored during the study and found to be unchanged throughout. These results suggest that LifePak® supplementation leads to significant strengthening of the body's antioxidant health status as indicated by the BioPhotonic measurement of skin carotenoids.\*

## Introduction

Recent advancements in laser technology have offered new opportunities for the health care industry. The application of BioPhotonic technology increasingly enables non-invasive biological measurements. Raman spectroscopy is a powerful laser spectroscopy that detects the characteristic vibrational/rotational energy levels of a molecule. Inelastically scattered light ("Raman" scattering) originates when energy is exchanged between incident light photons and the scattering molecules, resulting in a characteristic red shift when comparing the incoming with the scattered photon. Raman spectroscopy generates a spectral fingerprint, which depends on a molecule's unique vibrational energy scheme. Since Raman scattering is linear, the intensity of a Raman spectroscopy measurement is directly proportional to the amount of molecules.

Recently, Raman spectroscopy has been applied to the measurement of carotenoids present in the *stratum corneum* layers of human skin (Hata et al., *J Invest Dermatology* 115:441, 2000; and Ermakov et al., *Optics Letters* 26:1179, 2001). Carotenoids play an important role in human health (Gerster, *Int J Vitam Nutr Res* 63:93, 1993), and are believed to confer antioxidant and photo-protective benefits to the skin (Alaluf et al., *J Nutr* 132:399, 2002; Stahl et al., *J Nutr* 131:1449, 2001). Raman spectroscopy allows for a non-invasive, rapid, accurate, and safe assessment of carotenoid levels in the skin. Research suggests that

skin carotenoid levels correlate with levels of carotenoids in the diet and blood (Hata et. al., *J Invest Dermatology* 115:441, 2000).

Carotenoids scavenge singlet oxygen and are an important part of the body's antioxidant defense system (Omaye ST et al., *J Am Coll Nutr* 15:469-74, 1996; Handelman GJ, *Nutrition* 17:818-22, 2001). Serum carotenoid concentrations as well as BioPhotonic skin carotenoid responses are affected by oxidative stress, smoking, sunlight exposure and fruit and vegetable consumption as demonstrated by a large Pharmanex study concluded recently to assess these relationships in 1,375 subjects (unpublished results, Pharmanex, LLC, Provo, UT). Therefore, the skin carotenoid's BioPhotonic response appears to be a convenient and suitable indicator of the body's antioxidant status.

Results of the Pharmanex population study also showed that subjects consuming the antioxidant multi-nutrient supplement LifePak® had significantly higher skin carotenoid levels than subjects not taking antioxidant supplements (unpublished results, Pharmanex, LLC, Provo, UT). The present study was conducted to determine if there is a causal relationship between skin carotenoids and LifePak® supplementation.\*

\* These statements have not been evaluated by the Food and Drug Administration. This product is not intended to diagnose, treat, cure or prevent any disease.

## Materials and Methods

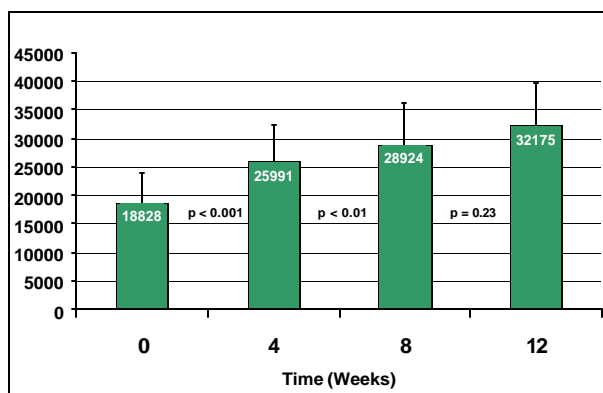
Twenty-five healthy non-smokers between the ages of 18 and 65 years were recruited for this study. Subjects were excluded from the study if they had used antioxidant supplements within the prior three months. Subjects participating in this study were instructed to complete a computer-administered questionnaire to assess demographical, dietary and lifestyle variables. The questionnaire contained a food frequency query asking subjects to record their consumption of foods containing more than 1 mg of total carotenoids per serving according to the USDA carotenoids database. Subjects then underwent the measurement of carotenoid levels in the skin on the palm of the hand using a BioPhotonic Scanner at the University of Utah, Salt Lake City, UT, in the laser laboratory of Werner Gellermann, Ph.D., Department of Physics. On the same day of this initial baseline measurement, subjects were supplemented with LifePak® for 12 weeks at the recommended dosage of two packets daily. The skin carotenoid's BioPhotonic response was measured at 4, 8 and 12 weeks of the study. Statistical significance was examined using appropriate tests (t-test).

## Results

A total of 25 subjects met all study criteria and participated in the study. Retained subjects numbers at 4, 8 and 12 weeks were 22, 17 and 12, respectively. Reasons for dropouts were related to scheduling and compliance problems. No serious adverse reactions were reported. Fruit and vegetable consumption was monitored throughout the study and remained constant at about 2.2 servings daily.

The effects of 12 weeks of LifePak® supplementation on biophotonically measured skin carotenoids are shown in Figure 1.

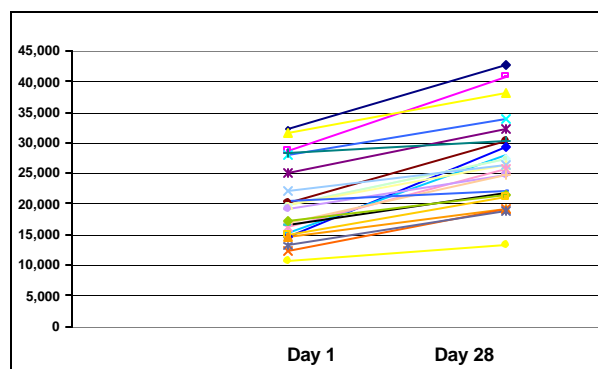
**Figure 1: Changes in Biophotonic Response**



The supplement increased mean scores by 38% ( $p < 0.001$ ) from baseline to week 4, by another 11% by week 8 ( $p < 0.01$ ), and by another 11% by week 12 (n.s.). Changes at any time point were significant compared to baseline ( $p < 0.001$ ).

Figure 2 shows the individual changes observed from baseline to day 28 of the study. All subjects experienced increases in skin carotenoids. The mean increase was  $7,687 \pm 3,212$  units (mean  $\pm$  std. dev.), ranging from 1,500 to 14,600 units. The mean increase for the entire 12-week study period ( $n=12$ ) was  $10,750 \pm 5,865$  units, ranging from 1,900 to 20,300 units.\*

**Figure 2: Individual Changes, Days 1-28**



## Discussion

Antioxidant supplements can improve antioxidant status and this has been shown as well for LifePak® (Pharmanex LLC, Provo, UT) in earlier clinical studies showing increases in serum antioxidant concentrations and improved resistance to *ex vivo* LDL oxidizability (Smidt et al., *FASEB J* 13:A546, 1999). The observed increases in skin carotenoids with LifePak® supplementation in present study at unchanged intake of fruits and vegetables confirm our hypothesis of a cause and effect relationship that was suggested in our earlier population study. LifePak® is a complete vitamin/mineral supplement with additional antioxidant nutrients and provides a total of 15 mg carotenoids daily, as 6 mg beta-carotene, 2 mg alpha-carotene, 5 mg lycopene and 2 mg lutein. The present study not only confirms our earlier studies that the carotenoids of LifePak® appear in blood serum (Smidt et al., *FASEB J* 13:A546, 1999), but also shows that the supplement's carotenoids are delivered to the skin as an important site of action.\*

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The individual variability reported in this study (Figure 2) can be attributed to several factors. Carotenoid absorption may vary due to meal size, the amount of fat consumed with the supplement and perhaps genetic predisposition (Kostic D et al., *Am J Clin Nutr* 62:604-10, 1995; Omaye ST et al., *J Am Coll Nutr* 15:469-74, 1996). It is possible that the low responders in our study did not take the supplement as directed, i.e., with meals, or that the meals were low-fat or fat-free meals. Dietary fat is needed to enable bile secretion and subsequent chylomicron formation in the intestine, which are necessary for carotenoid absorption. In addition, individually different levels of oxidative stress, which again depend on genetic and environmental factors, may have affected skin carotenoid levels.

## Conclusions

These results suggest that LifePak<sup>®</sup> supplementation leads to significant strengthening of the body's antioxidant health status as indicated by BioPhotonic measurement of skin carotenoids.\*

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