Group #5

Effects of beta-carotene supplementation on blood serum levels of beta-carotene and vitamin E

Summary

This paper describes the results from a randomized double-blind 9-month clinical trial designed to investigate whether beta-carotene treatment affects blood serum beta-carotene levels and serum vitamin E levels. The 48 patients in the study were randomized into groups receiving doses of 0, 15, 30, 45 or 60 mg/dl of Beta-carotene administered daily. Serum beta-carotene and vitamin E measurements were taken at both 3 months and 9 months. Not all measurements were made on all subjects. Since there is no known risk of beta-carotene toxicity at the dose levels given, and the effects of long-term supplementation are more scientifically important, the 9 month measurements are of more interest. The sample was 48% male; the average age was 57.

Based on the analysis it was determined that treatment with beta-carotene increases blood serum levels of beta-carotene, and there is a greater increase in levels with higher doses, and over a longer period of time. There is a negative relationship between beta-carotene treatment and 9 month vitamin E blood serum levels. One plausible explanation for this is that serum levels of beta-carotene must build up before an effect is seen on serum levels of vitamin E.

It was found by use of a T-test that treatment with beta-carotene had a positive and significant effect on serum beta-carotene levels at both 3 and 9 months. The estimated mean beta-carotene was 1411 mg/dl higher at nine months in patients taking any supplements than in patients taking placebo (1597 vs 186 mg/dl). Mean serum beta-carotene levels in those treated with any level of beta-carotene is significant with a p-value of <0.0009, and a 95% CI of 989 to 1833 mg/dl. These findings were similar to those at 3 months although they differed in magnitude. Significantly increased levels of serum beta-carotene were also found when dose level was considered.

Findings about the association between treatment with beta-carotene and blood serum levels of vitamin E were not as straightforward as those for serum beta-carotene. The estimated treatment effect at nine months was to reduce vitamin E levels by -1.12 (7.25 vs. 6.13 mg/L). This difference had a p-value of 0.0405 and a 95% CI -.06 and -2.17 making it marginally significant. Despite this significance, the association over time is not clear and further investigation is required as at 3 months there is no significant association between beta-carotene treatment and serum vitamin E levels.

Due to a small sample size, OLS regression analysis was used to increase precision in the analysis above. Three separate OLS models were fit to the data using 9 month and 3 month serum beta-carotene levels, and 9 month serum vitamin E levels as dependent variables respectively. The variables male, age and cholesterol level at baseline were included in the regressions, as well as the respective baseline measure of serum level. The results were similar to those of the T-tests, but with increased precision.

Background

Beta-carotene is an important dietary carotenoid that is a precursor to vitamin A[1]. There is evidence that increased dietary intake of beta-carotene may be associated with decreased risk for lung cancer, lung diseases and stomach cancer[2]. Although dietary beta-carotene appears to be beneficial, little is known about the effects of beta-carotene supplements. A previous study done by Micozzi et al.[3] has shown that individuals taking beta-carotene supplements are at risk for developing carotenodermia (yellowing of the skin), but there is no known evidence of toxicity[4]. Because of the apparent benefits of increased dietary beta-carotene, and the lack of beta-carotene supplement toxicity, it is important to understand the effects of beta-carotene supplementation in the long term.

In order to better design studies that analyze the potential benefits of beta-carotene supplements, researchers need to understand how different dose levels affect plasma levels of beta-carotene and other blood chemistries over time. There is concern that increased levels of plasma beta-carotene might affect levels of other lipid soluble nutrients such as vitamin E. A study done by Willie et al.[5] found that daily

Comment [A1]: Overall goal? Cancer chemoprevention.

Comment [A2]: I only found 46 patients in the dataset.

Comment [A3]: Probably not in summary, unless a major aspect of your analysis

Comment [A4]: Sort of technical. Why not just say means were compared? And why not start off with the estimates, and then say that the observed differences were significant.

Comment [A5]: I don't understand. How could your prior analysis not considered dose. (Well, okay, I can guess what you mean, but this is not the best way to describe it, and others would probably not understand.)

Comment [A6]: Estimates first. Estimates first. Estimates first. (There are lots of reasons results can be not significant.)

Comment [A7]: Why wasn't this your primary analysis? The only reason I could think of (given the fact that you were going to do it) was that your statistical analysis plan written at the start of the study specified a less precise analysis. Otherwise, I would think you would have just done this analysis.

In any case, it would be important to know how you chose the variables for adjustment. You might have said "in pre-specified secondary analyses, we adjusted for …"

Comment [A8]: The distinction between any effects of dietary beta carotene and supplementation is something that has arisen as the RCTs have been performed, though I will admit that good scientists should have always worried about this issue.

Comment [A9]: presumably in cancer chemoprevention.

Comment [A10]: Willett, as in Walt Willett—one of the main dietary researchers.

beta-carotene supplementation alone increased plasma levels of beta-carotene, but had no effect on plasma levels of vitamin E.

Supplementation did, however, lead to a smaller increase in plasma levels of vitamin E in subjects who received both beta-carotene and vitamin E supplements. Because the study only looked at the effects of 1 dose level (30 mg/day) over a period of 16 weeks, more information is needed on the long term effects of a variety of beta-carotene doses. This study analyzes the effects of 15, 30, 45, or 60 mg/day doses of beta-carotene on plasma levels of beta-carotene and vitamin E for a period of 9 months.

Comment [A11]: Ultimately, we will want to have this quantified: Did "no effect" mean no stat signif" Or was the CI truly of width zero.

Questions of Interest

One objective of the study was to assess the effect of 5 dose concentrations of beta-carotene supplementation on blood serum beta-carotene levels. The study also aimed to find what effect beta-carotene treatment might have on serum vitamin E.

While we address both of these objectives, our primary question of interest is the following: how does serum beta-carotene build up in the body after 3 and 9months of supplementation? We consider the effect of beta-carotene dose on vitamin E blood serum levels at 3 and 9 months a secondary question.

Comment [A12]: And you ight invoke the overall goal of cancer chemoprevention in order to make clear the greatest interest in the longest time period.

Source of the Data

The dataset used for this analysis was downloaded from the Biostatistics 517 website (http://www.emersonstatistics.com/courses/formal/b517_2007/index.asp) on November 16th, 2007. The original source of the data is not known. The dataset contains several subject-specific measures obtained from an experiment designed to address the scientific questions previously mentioned. More specifically, the experimental methods used involved a randomized assignment of 48 volunteers to one of five doses of beta-carotene: 0, 15, 30, 45 and 60 mg/day. The groups receiving dose levels of 0 mg/day and 45 mg/day were each comprised of 8 volunteers. All other dose groups were comprised of 10 volunteers each. The dose group assignment was double-blind meaning neither the study team nor the volunteers were informed of the assigned dose levels.

Comment [A13]: Protecting yourselves against me changing the data, I presume?

Comment [A14]: 46

The volunteers were asked to take their dose of beta-carotene daily for 9 months. Measurements in mg/dl of serum beta-carotene and vitamin E were made at the time of randomization as well as at 3 and 9 months post randomization. Several other measures describing the subjects were taken only at randomization. These include age, sex, weight, body mass index, serum cholesterol level and percent body fat. In our analysis these variables were considered as adding precision. The other variables included in the dataset, time average of serum vitamin E treatment and time average of serum beta-carotene while on treatment, were investigated and determined to not be relevant to this analysis. Table 1 displays summary statistics for these measures by dose group. Taking into consideration the overall scientific objective of the analysis, the beta-carotene and vitamin E measures are classified as outcome variables while dose group is classified as the predictor of interest.

Comment [A15]: This is appropriate when writing to a client, and not so appropriate for putting in a scientific paper.

Comment [A16]: Results

Comment [A17]: Technical jargon. Avoid it.

Due to a small sample size and randomization to dose group, missing values for serum beta-carotene and vitamin E are treated as random and dropped from the analysis. Although there are some differences among subjects with missing outcome variables, there does not seem to be an association between missing values and dose group. This does not, however, guarantee that the missing values would not affect the outcome if present, or that no pattern of missingness could be found if more data were available.

Comment [A18]: I think you mean "missing at random"—a phrase that has specific meaning.

Comment [A19]: presumably the case was dropped from the analysis if the case was missing data

Statistical Methods

Comment [A20]: Sort of a toss-up here as to whether you dismiss the issue in the materials and methods, or whether you report this as results. I tend to report it as results, because it might be hidden toxicity. In a new treatment, it would definitely be a result

Comment [A21]: A lot of the above discussion on missingness was really better placed in Stat Methods.

In order to address the primary scientific question we undertook three statistical tasks: we estimated the effect of different dose levels of beta-carotene on serum levels of beta-carotene as well as the build-up of beta-carotene over time, and we estimated the effect of different dose levels of beta-carotene on serum levels of vitamin E. For the purposes of our analysis, the 3 and 9 month serum levels of beta-carotene and vitamin E are considered the outcome variables. Among these, the serum values at 9 month post randomization are of primary scientific interest. The literature is mainly concerned with long-term medical uses of beta-carotene, and given there are no concerns about toxicity (see background), the 9 month measurement is of most use. The difference between 9 month serum levels and baseline values at the

Comment [A22]: Arguably, we would also be interested in toxicity over this period. We would likely look at cumulative effects.

time of randomization was not used as the outcome measure in order to improve statistical precision by avoiding increased variance. Baseline measurements of both serum beta-carotene levels and vitamin E levels were included as predictors in the final phase of our analysis in order to avoid biasing estimated dose effects when fixed differences in baseline biochemistry between individuals in the dose groups are not controlled for. Stata version 9.1 was used for all statistical analyses.

As an initial analysis we explored an overall treatment effect by defining the treatment group as all subjects receiving a non-zero dose of beta-carotene, and the control as the group of subjects taking the placebo. We conducted a student's t-test both to quantify differences in serum beta-carotene and vitamin E levels between treatment and control, and to determine if any differences were statistically significant. We then explored how the observed effects (differences between treatment and control) differed over each dose concentration. The results of these preliminary t-tests are discussed briefly in the results section.

The primary analysis used was ordinary least squares (OLS) regression to model the effect of dose concentration on serum levels conditional on individual characteristics (equation 1). Given the small sample size, using OLS allowed us to borrow information across close neighbors within independent variable groups. The randomized design of the study mitigates concerns of confounding by variables associated both with dose group, and with serum levels of beta-carotene and vitamin E. Nevertheless, cholesterol, sex and age were included in the analysis for precision. The exact relationship between outcome and the precision variables was not of great importance, and a decision was made not to log transform the data, though non-linear relationships were detected. Although weight, BMI and percentage body fat could also have been included in our analysis we found that this was unnecessary because they were strongly correlated with age, sex and cholesterol. In addition, blood lipids (cholesterol) are known to affect serum levels of lipid soluble vitamins[5]. Robust standard errors were used to correct for heteroscedasticity

In the regression analysis, dose concentration is modeled as the predictor of interest on the serum concentration in the blood, not its levels over time. We achieved this by including dose concentration in the model as binary variables; a value of one meant the subject was assigned to that particular dose concentration, and a value of zero meant the subject was assigned to another. As dose concentration 0 was left out of the model, interpretations of the coefficients on Doses 15, 30, 45 and 60 are made relative to the placebo group.

***Equation 1**:*

$$Y_i = \beta_0 + \beta_1 D_{15,i} + \beta_2 D_{30,i} + \beta_3 D_{45,i} + \beta_4 D_{60,i} + \beta_5 X_{,i} + E_i$$

Dose groups are modeled with binary variables (D_{15}, D_{30}, D_{45}, D_{60}, D_{15}) indicating whether or not subject i belongs to dose group 15, 30, 45 or 60. X is a matrix of independent variables being controlled for in the regression that includes cholesterol, age, baseline serum level and an indicator for being male. The assumption is made that E_i is normally distributed. The results of this regression model (estimated coefficients, standard errors, confidence intervals and p-values) are presented in Table 3.

Following the regression there were several paired t-tests performed to assess the level of serum beta-carotene build-up that seemed evident from both the unpaired t-tests and the regression analysis. The paired t-testes are outlined in table 2 with the preliminary t-tests for treatment effect. The difference between a paired t-test and the unpaired test done between dose groups is that the paired tests are done accounting for individuals, thus allowing for inference on effects of the beta-carotene treatment over time. These tests were performed as secondary analysis to the regression results, and are in support of those findings.

Comment [A23]: The validity of this statement depends on the correlation between baseline and follow-up.

Comment [A24]: Probably a better approach to use with vitamin E than beta carotene, as it is quite likely that there is a dose response for beta carotene

Comment [A25]: What made you choose these variables? Ideally you would make this decision based on knowledge prior to looking at the data.

And the number one precision variable to have included was the baseline variable.

Comment [A26]: Probably a bit technical. Especially if you start talking about a matrix of iindependent variables. And how would that be modeled with a scalar parameter.

Comment [A27]: Paired t tests? How? Within dose groups? What are these answering?

Results

Descriptive statistics

Table 1 provides the means and standard deviations of the precision and outcome variables used in subsequent analyses.

Table 1 Summary Statistics of Analysis Variables, by dose group

	Dose 0 mg/day (N = 8)					Dose 15 mg/day (N = 10)					Dose 30 mg/day (N = 10)				
	Mean	SD	Min	p50	Max	Mean	SD	Min	p50	Max	Mean	SD	Min	p50	Max
Age (yrs)	56.3	4.3	52.0	55.5	64.0	56.3	4.6	50.0	56.5	62.0	57.2	4.1	50.0	57.0	64.0
Proportion Male	0.6	0.5	--	--	--	0.5	0.5	--	--	--	0.3	0.5	--	--	--
Cholesterol	217.8	28.5	190.0	211.5	283.0	223.0	29.7	171.0	223.5	265.0	213.2	33.5	159.0	214.5	268.0
Serum beta-carotene															
At randomization	270.2	136.3	136.3	227.8	476.3	220.1	127.9	64.8	185.6	496.0	219.4	83.8	125.5	205.0	348.5
N missing			*0*					*0*					*0*		
3 months	243.5	94.3	109.3	220.5	384.0	1116.4	317.4	699.0	1203.0	1602.7	1302.3	259.9	854.0	1289.3	1603.3
N missing			*0*					*0*					*1*		
9 months	186.3	87.8	84.5	149.0	323.0	1253.6	570.5	576.8	1250.0	2018.8	1504.6	479.0	849.3	1498.5	2248.5
N missing			*1*					*2*					*1*		
Serum vitamin E															
At randomization	7.88	1.42	6.19	7.60	10.71	7.76	1.21	5.10	7.95	9.24	7.98	1.62	5.12	8.57	9.46
N missing			*0*					*0*					*0*		
3 months	8.27	1.23	6.50	8.40	10.11	8.71	0.91	6.36	8.85	9.74	9.15	0.90	7.12	9.42	10.55
N missing			*0*					*0*					*0*		
9 months	7.25	1.13	5.26	7.23	8.93	5.75	0.50	4.61	5.84	6.28	6.30	1.14	4.31	6.20	7.74
N missing			*1*					*2*					*1*		

	Dose 45 mg/day (N = 8)					Dose 60 mg/day (N = 10)				
	Mean	SD	Min	p50	Max	Mean	SD	Min	p50	Max
Age (yrs)	55.9	3.1	51.0	55.5	60.0	56.5	5.2	52.0	54.5	65.0
Proportion Male	0.5	0.5	--	--	--	0.5	0.5	--	--	--
Cholesterol	213.3	33.5	169.0	212.0	263.0	238.1	38.9	209.0	219.5	312.5
Serum beta-carotene										
At randomization	227.0	105.5	93.3	216.4	395.8	217.8	122.3	48.3	224.3	407.5
N missing			*0*					*0*		
3 months	1236.0	239.3	860.5	1343.3	1440.5	1466.7	251.1	1098.0	1410.3	1959.7
N missing			*0*					*0*		
9 months	1749.1	579.0	950.3	1848.3	2310.4	1877.6	429.9	1233.3	1865.0	2855.0
N missing			*1*					*1*		
Serum vitamin E										
At randomization	8.24	0.95	7.22	8.04	10.05	8.44	1.27	6.32	8.51	10.71
N missing			*0*					*0*		
3 months	8.98	0.63	7.89	8.81	9.78	9.11	0.66	8.07	9.26	10.02
N missing			*0*					*0*		
9 months	6.15	0.88	4.94	5.95	7.05	6.32	1.12	4.87	5.93	8.06
N missing			*1*					*1*		

The average age of the sample was 56.5 years and 47.8 % were male. The mean serum cholesterol level was 221.5 mg/dl. Differences among these precision variables are minimal between dose groups. However, salient differences between dose groups over time do emerge among the statistics summarizing the distribution of serum beta-carotene and, to a lesser extent, vitamin E. These differences are displayed more clearly in Figure 1which provides a visual representation of changes in the values of the outcome measures over time. For beta-carotene (Figure 1), the most striking pattern is the difference in trends between dose group 0 and all other dose groups. While values of beta-carotene fall after randomization for dose 0, values for the treatment dose groups jump significantly after 3 months (between 5 and 7 times the baseline amount) for all other dose groups. This rate of increase is present, but less drastic, between 3 and 9 months for the treatment dose groups. The unexpected pattern of increasing vitamin E serum levels at 3 months, and decreasing vitamin E serum levels at 9 months, suggests there is a complex relationship between vitamin E serum levels and beta-carotene serum levels; this is further investigated in the analysis section. After examining the summary statistics and the distribution of the data in Figure 1 we determined

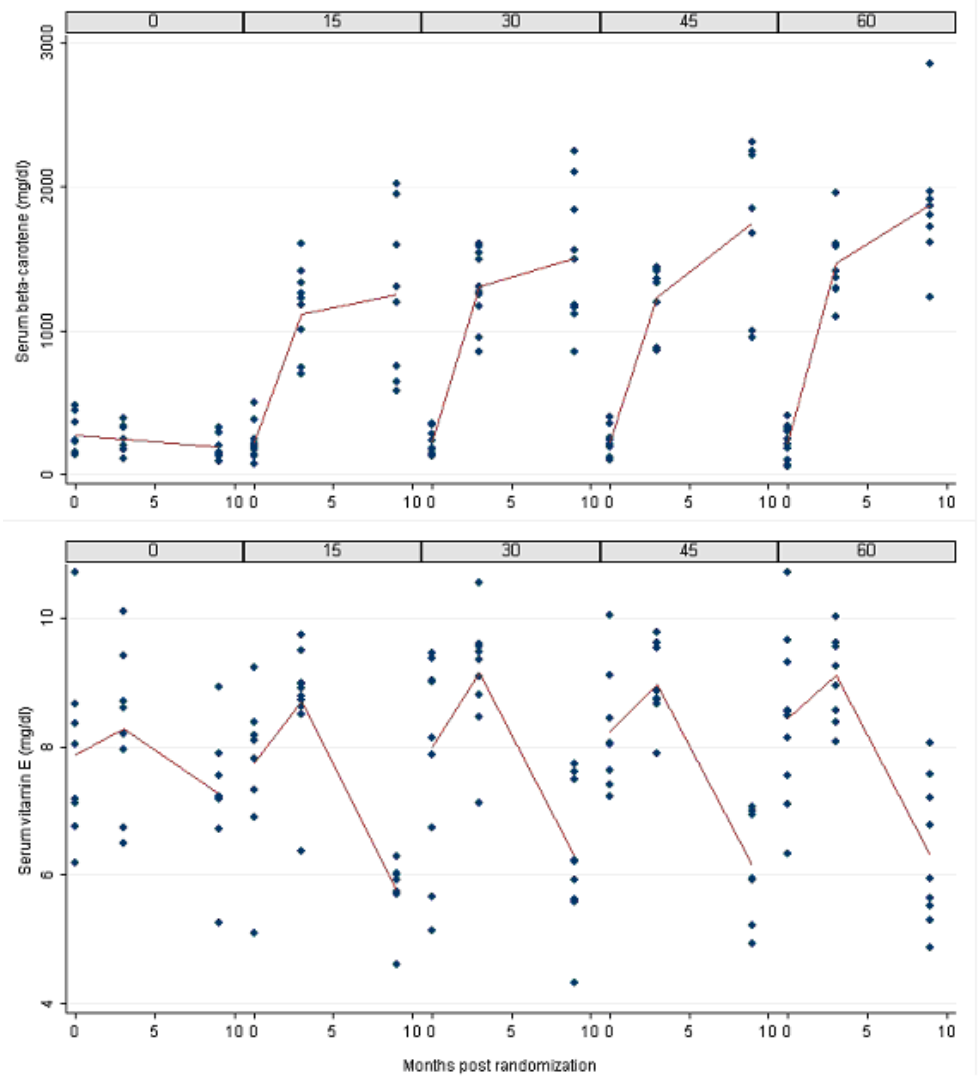
Comment [A28]: Why would they do this?

Comment [A29]: even in the placebo group!

Comment [A30]: not in the placebo group

there weren't any data points sufficiently extreme to merit taking an analysis approach that would mitigate the influence of outliers.

Figure 1. Graphical Results of Outcome variables



Analysis

The results of the many treatment effect T-tests preformed as well as the paired t-test to asses build-up can be seen in Table 2. The estimated mean difference of 9 month beta-carotene levels between the treatment and the control group was 1411 mg/dl. The data are not unusual for differences as small as 1207 or as large as 1614 mg/dl. The p-value of <0.009 obtained in this test provides sufficient statistical evidence to reject the null hypothesis that there is no difference between the treatment and control group. Statistically significant results were also obtained for beta-carotene differences at 3 months (although the estimated difference was about 377 mg/dl smaller). We explored the effects of dose concentration by running several t-tests using both 3 and 9 month serum levels to assess differences between dose groups 15, 30, 45 and 60 mg. relative to the placebo group. All of the differences obtained in this sub-analysis were

Comment [A31]: Undoubtedly you determined your analysis plan prior to looking at the data, so you clearly meant that it did not merit further exploratory analyses that would be different.

Comment [A32]: You could have jittered a tiny bit. But I kind of like this graph, even though you do not have vertical lines separating the dose groups (I probably would have).

I do note that the primary comparison should be between doses, because that is what was randomized. But this looking at time as the primary effect does fit in with your text, and I can judge what is going on by dose with this. And I very much like seeing the individual data.

Prsumably the lines are connecting means. You could tell us that.

Comment [A33]: the combined treatment groups. Make this clear.

Comment [A34]: These analyses would be of far greater interest than the paired t tests. I would have reported them.

significant at the 5% level, and the effect size increased proportionally with dose concentration (with the exception of a 45 mg. dose, whose effect on serum levels at 3 months is smaller than the effect of a 30 mg. dose). These tests are not included in the analysis as they are very similar to those of the regression analysis.

Table 2. T-tests Performed on varying treatment levels

Variable	Diff Est.	P-Value	95% CI		Variable	Diff Est.	P-Value	95% CI	
9 Month Beta-Carortene Serum levels					**9 Month Vitamin E Serum levels**				
Treatment (1, 0), unequal allowed	1411.02	<.0009	1207.53	1614.52	Treatment (1, 0), unequal allowed	-1.12	0.041	-2.17	-0.06
Paired test from baseline on treatment	1374.13	<.0009	1196.66	1551.59	Paired test from baseline on treatment	-1.85	<.0009	-2.26	-1.43
Paired test from baseline on Placebo	-101.56	0.007	-163.32	-39.79	Paired test from baseline on Placebo	-0.86	0.011	-1.44	-0.28
3 Month Beta-Carortene Serum levels					**3 Month Vitamin E Serum levels**				
Treatment (1, 0), unequal allowed	1034.19	<.0009	916.23	1152.14	Treatment (1, 0), unequal allowed	0.71	0.154	-0.33	1.75
Paired test from baseline on treatment	1052.59	<.0009	963.52	1141.66	Paired test from baseline on treatment	0.93	<.0009	0.62	1.23
Paired test from baseline on Placebo	-26.72	0.244	-76.36	22.93	Paired test from baseline on Placebo	0.40	0.141	-0.17	0.97

Comment [A35]: The paired t tests add nothing to me. We had a placebo group. We would not want to presume that the placebo group necessarily had no differences over time, so our real question is how the groups with suppliementation differed from the placebo group.

Estimated differences in vitamin E levels at 9 months were negative and significant between treatment and control. The estimated difference was -1.12 mg/dl with a 95% confidence interval of -0.06 to -2.17 mg/dl. Interestingly, there was no significant association between serum vitamin E and treatment defined broadly, or within dose groups, at 3 months. Based on the information in the paired t-test between 3 month serum vitamin E levels and baseline for those on treatment, there is a significant increase in vitamin E levels. Those not receiving treatment have a non-significant increase in vitamin E levels.

Comment [A36]: and this is important why? (To me, it show the importance of having a placebo group and the relative irrelevance of the paired t tests above).

There was a significant increase from baseline beta-carotene levels at both 3 and months for subjects receiving beta-carotene treatment. There was a significant decrease in beta-carotene in the placebo group after 9 months and a non-significant decrease after 3 months. There was a significant decrease in vitamin E blood serum levels 9 months after baseline for those receiving beta-carotene treatments, as well as for those receiving placebo. The decrease was twice as large in those receiving treatment. There was an increase from baseline vitamin E blood serum levels in both treatment and placebo after 3 months; the increase in the treatment group was significant.

Comment [A37]: same comment

To more precisely explore the relationship between dose of beta-carotene and outcomes we ran three separate OLS models, defined generically in Equation 1. We chose to do only 3 regressions because we did not find that there was a significant difference in the 3 month serum vitamin E levels based on beta-carotene treatment in the t-tests, and so further analysis to increase precision was not advisable. Results for

Comment [A38]: avoid data-driven analyses

The models using 9 and 3 month serum beta-carotene levels as well as 9 month serum vitamin E levels are presented in Table 3. The results reaffirmed what we found in our initial inferential analysis using t-tests.

Comment [A39]: Hardly surprising, if all we were doing was modeling precision variables when comparing means and when already having statistical significance.

9 month serum beta-carotene levels of a volunteer in the group given a dose of 15 mg of beta-carotene daily for 9 months, all else being equal, will tend to result in a 1176 mg/dl increase in blood serum levels over the 9 month blood serum levels of a volunteer in the placebo group. The relative average increase for dose concentrations 30, 45 and 60 mg were 1339, 1640 and 1719 mg/dl relative to placebo. For the 3 month beta-carotene measures, the dose effects are similar in that average serum levels go up per dose group, but the magnitude of the effects are smaller.

Based on our results, the overall effect of beta-carotene dose and 9 month serum vitamin E is negative. The effect size for dose concentrations 15, 30, 45 and 60 mg were -1.24, -.88, -1.21 and -1.11 mg/dl, respectively. Unlike serum beta-carotene there does not seem to be a mediating effect of dose concentration on serum levels, the relation suggested in Figure 1.

Table 3. OLS Regression Results

	9 Month Beta Carotene		3 Month Beta Carotene		9 Month Vitamin E	
Dose 15						
B	1176.73		892.81		-1.24	
SE	203.541		102.553		0.288	
CI	761.6	1591.85	684.62	1101.01	-1.82	-0.65
P>|t|	<0.0009		<0.0009		<0.0009	
Dose 30						
B	1339.27		1053.73		-0.88	
SE	190.6		89.2		0.473	
CI	950.53	1728	872.64	1234.83	-1.84	0.09
P>|t|	<0.0009		<0.0009		0.074	
Dose 45						
B	1640.23		1002.05		-1.21	
SE	172.966		73.196		0.346	
CI	1287.46	1993	853.46	1150.65	-1.92	-0.51
P>|t|	<0.0009		<0.0009		0	
Dose 60						
B	1719.84		1225.36		-1.11	
SE	216.17		114.95		0.38	
CI	1278.96	2160.71	992.01	1458.72	-1.89	-0.34
P>|t|	<0.0009		<0.0009		0.006	
N	*40*		*44*		*40*	
Root MSE	*415.57*		*227.77*		*0.845*	
	B = Coefficient on Dose, SE = Robust Standard Error, CI = 95% Confidence Interval *P>|t| = Two-sided p-value*					
	Other variables include baseline beta-carotene serume indicator for male, baseline Cholesterol and age				*Serum baseline Vitamin E was included rather than beta-carotene baseline*	

Discussion

The effect of increased beta-carotene dose was found to increase mean blood serum beta-carotene concentration for the 9 month measure. This is also true for the 3 month measure. Within individuals on beta-carotene treatment there is a trend of beta-carotene build-up in the serum levels that is supported both by the regression results and the paired t-tests. As stated in the background there does not seem to be a toxic level of beta-carotene, so there is no reason to believe that the clear build-up effect is a problem. One can see the build-up as levels are increasing from 3 months to 9 months in each dose group. Higher doses have larger effects on blood serum levels of beta-carotene. With this information known, studies attempting to see the effect of beta-carotene treatment on some disease, or other blood serum levels, can use dose group rather than testing serum beta-carotene directly, and it can be expected that serum levels will change as dose is changed.

Comment [A40]: is turning orange-yellow a toxic effect?

The negative coefficient values in the 9 months vitamin E regression suggest that vitamin E serum levels decline after 9 months with non-zero doses. The fact that the negative values do not decrease as dose increases implies there is a complicated relationship between dose group and serum vitamin E levels that should be further investigated if more effect information is desired. Individuals in the study both on treatment and not have an increase in vitamin E blood serum levels after 3 months, although only in treatment is it significant. This may suggest that there is a level of beta-carotene that increases absorption of vitamin E and only beyond that point is there an adverse effect of beta-carotene on vitamin E level. The 9 month serum vitamin E levels regression yields interesting results. As dose increases serum vitamin E levels increase slightly, particularly at a dose of 30 mg. This may imply that there is not a linear relationship between treatment with beta-carotene and decreasing vitamin E levels over time, also

Comment [A41]: I agree. Sort of violates one of Koch's postulates, doesn't it.

suggesting that there may be a threshold level of serum beta-carotene needed to have a suppressive effect on serum levels of vitamin E.

Further investigation with a larger sample size would be advised. A longer study period might also be advisable to determine when and if serum levels plateau.

References

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