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ORIGINAL ARTICLE

Components of variation in serum carotenoid concentrations: the Polyp Prevention Trial

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Objectives: The intra- and interindividual variations and season and center effects were estimated from a series of serum carotenoid concentrations in the Polyp Prevention Trial (PPT) participants.
Subjects/Methods: Fasting blood was collected annually for 4 years in all 1905 participants, and a subcohort of 901 participants were selected within each (of eight) center(s), by gender and dietary arm of the study, for measurement of five major carotenoid peaks. Using variance of component methods, the variation in serum carotenoid concentrations about the underlying mean was partitioned into explanatory components attributed to various sources.
Results: The contributions of the inter- and intraindividual variances to the overall variation in carotenoid concentrations were in the range of 61–70 and 20–35%, respectively, whereas center and center-by-season effects provided 2.6–9.5 and 0.2–1.4%, respectively. The highest percent (35%) of intraindividual variation was exhibited by lycopene, and the highest percent (70% apiece) of interindividual variation was exhibited by lutein/zeaxanthin and β-carotene. Serum lycopene had the highest ratio of intra- to interindividual variation of 0.57, whereas lutein had the lowest ratio of 0.29. We estimate that the ratio of intra- to interindividual variance around the mean carotenoid concentration can be reduced greatly by collecting 3–4 compared to 1 blood measurement in large-scale trials like the PPT.
Conclusion: In the largest study of components of variation in individuals at high risk for colorectal cancer, the largest contributors to variation in serum carotenoid concentrations were intra- and interindividual effects followed by center and center-by-season effects.
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Keywords: carotenoids; serum; repeated measurements; Polyp Prevention Trial

Introduction

Serum carotenoid concentrations may more accurately reflect a dietary intervention plan that increases or decreases specific vegetables and fruits than one's reports of usual fruit and vegetable intake (Rock *et al*., 1992; Campbell *et al*., 1994; Le Marchand *et al*., 1994; Michaud *et al*., 1998; Polsinelli *et al*., 1998; Forman *et al*., 1999; Smith-Warner *et al*., 2000; John *et al*., 2002; Pollard *et al*., 2003; Jansen *et al*., 2004; Al-Delaimy *et al*., 2005). The association of serum carotenoid concentrations with dietary fruit and vegetable intakes may in part be due to sources of variation in measurement of dietary and serum carotenoids. Season of the year for dietary assessment and for blood collection, inter- and intraindividual variations in intake, and food processing and preparation are contributors to variation in circulating carotenoid concentrations (Castenmiller *et al*., 1999, 2000). Characteristics such as age, body mass, smoking status and alcohol consumption also influence serum carotenoid

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concentrations (Forman *et al*., 1993; Tang *et al*., 2003). Dietary tools, their mode of administration, memory and length of recall, health status of the respondent, food composition databases and laboratory assays are issues that factor in the ability to detect a diet–serum association (Al-Delaimy *et al*., 2005). Indeed, the range of diet–serum carotenoid correlation varies by the individual carotenoid, the carotenoid-rich food sources of the population, as well as the abovementioned characteristics and methodological issues (Romieu *et al*., 1990).

Many factors influence the bioavailability of carotenoids. For example, enzymatic disruption of the food matrix can enhance bioavailability of β-carotene from whole leaf and liquefied spinach, but it has no effect on lutein bioavailability (Castenmiller *et al*., 1999). The administration of a single or combined pharmacological dose of carotenoids can reduce or enhance uptake. An oral dose of lutein impairs zeaxanthin bioavailability and reduces β-carotene uptake in some but enhances uptake in other patients (Kostic *et al*., 1995). An oral dose of β-carotene reduces lutein bioavailability by 54–61% (Thurmann *et al*., 2005). Certain dietary fibers reduce the absorption of carotenoids in women (Riedl *et al*., 1999), whereas lycopene eaten with a lipid-rich food like avocado enhances its absorption from a tomato matrix (Unlu *et al*., 2005). Indeed, the capacity of the digestive system to release carotenoids from the food matrix may be the first step toward determining bioavailability of the carotenoid. Both the small and large intestines are responsible for differential uptake of individual carotenoids (Goni *et al*., 2006).

Earlier research has reported intra- and interindividual variations in serum carotenoid concentrations in small samples (Tangney *et al*., 1987; Yong *et al*., 1994; Talwar *et al*., 2005), in smokers (Block *et al*., 2006) and in one large sample of the European Prospective Investigation of Cancer (EPIC) (Ferrari *et al*., 2005). Yet large-scale multicenter trials, such as the Polyp Prevention Trial (PPT; Appendix), or observational research like the EPIC, contribute additional sources of variation in serum carotenoid concentrations due to potential within- and between-center and seasonal differences in serum levels (Al-Delaimy *et al*., 2005). The optimal data set in a long-term study would have serial measurements of serum carotenoids over time in a cohort to identify contributors to variation in serum levels. These data could provide benchmark estimates of the number of blood collections required to minimize the effect of components of variation to serum measurements.

Knowledge of the sources of variation in serum carotenoid concentrations is fundamental to the development of a long-term intervention trial such as the PPT. Carotenoid concentrations were considered biomarkers for compliance among participants who were randomly assigned to a dietary regimen with enhanced fruit and vegetable intake compared to those who remained on their usual dietary regimen for 4 years. Using annual blood collections to measure serum carotenoid concentrations in the PPT participants, the objectives of this study are (1) to describe seasonal variation in serum carotenoid concentrations by the participating center in the PPT; (2) to examine whether seasonal variation in serum carotenoid levels occurs across centers; (3) to determine the components of variation in serum carotenoid concentrations in PPT participants; and finally (4) to estimate the number of blood collections to reduce variation in serum carotenoid levels in a trial like the PPT.

Materials, subjects and methods

Study design

The PPT was a multicenter, randomized, controlled trial of the effect of a low-fat, high-fiber, high-fruit and high-vegetable dietary intervention on the recurrence of large bowel adenomas. The overall design, rationale, dietary intervention, end point ascertainment and results of the PPT have been reported previously (Lanza *et al*., 1996, 2001; Schatzkin *et al*., 2000). Between 1991 and 1994, the PPT enrolled 2079 men and women, who were aged $\geqslant$35 years and had at least one histologically confirmed large bowel adenomatous polyp removed during a colonoscopy within the previous 6 months at one of eight clinical centers in the United States (listed in Acknowledgements). Of the 2079 participants who were randomized to either the dietary intervention arm or their usual diet, 1905 completed the study after 4 years. All PPT participants completed a modified 106-item Block-National Cancer Institute food frequency questionnaire and a lifestyle questionnaire that included information on non-steroidal anti-inflammatory use, physical activity and smoking status at baseline. The food frequency questionnaire was repeated annually at the same time as the blood collection. Adenoma recurrence was assessed by a colonoscopy at the end of each year of follow-up. Participants were defined as having a recurrence if a polyp appeared during an endoscopic procedure following the colonoscopy after the first year of the trial. The study was approved by the Institutional Review Boards of the National Cancer Institute and the collaborating centers. All subjects provided written informed consent.

After 4 years of annual blood collections, a subcohort of 901 participants was selected proportionately within each center and by gender to represent both arms of the PPT. Each person had at least one blood collection over the 4 years. The percentage of the 901 participants who had 3–4 (all) blood collections were 79% (New York), 87% (Winston-Salem), 88% (Buffalo and Chicago), 91% (Pittsburgh), 94% (Salt Lake City), 95% (Oakland) and 96% (Bethesda). Venous blood samples were taken after an overnight fast.

Serum carotenoid analysis

Concentrations of five carotenoids (α-carotene, β-carotene, lutein/zeaxanthin, cryptoxanthin and lycopene) were measured using high-performance liquid chromatography in

serum samples (Sowell *et al*., 1994). The coefficients of variation for 109 blinded quality control samples from healthy individuals at the National Institutes of Health Clinical Center were 6.9% for α-carotene, 6.4% for β-carotene, 8.2% for lutein/zeaxanthin, 12.4% for cryptoxanthin and 7.3% for lycopene (Steck-Scott *et al*., 2004).

Statistical methods

Fisher's exact test was used to compare the proportion of participants in the intervention and control groups on baseline demographic characteristics (Agresti and Min, 2002). Welch's *t*-test (with unequal variances) was used to compare the means of continuous baseline characteristics in the intervention and control groups (Altman and Dore, 1991). All statistical analyses, except Fisher's exact tests, were performed in the S-Plus 2000 programming language.

Linear mixed models (Searle *et al*., 1992) were computed to test for seasonal effects on carotenoid levels over time. We treated treatment group as a fixed parameter in all models to adjust for the effect (if any) of the shift by the intervention group on the mean serum carotenoid levels. Note that on average, serum carotenoid levels did not appreciably differ by group over the 4 years of the study, as intervention group participants increased intake of non-carotenoid-rich fruit like apples and vegetables like legumes. Therefore, we adjusted for treatment group and yearly effects and included a random-effects term, which accounts for correlation in the repeated measures. Seasonal patterns in mean carotenoid levels were incorporated in the linear mixed model using a series of harmonic terms (based on sines and cosines). We tested for seasonality within each center by comparing models with and without the terms $\sum_{k=1}^{3}[\theta_k \sin(2k\pi t) + \gamma_k \cos(2k\pi t)]$, where t denotes time as a fraction of the year during which the particular measurement was taken, using a likelihood ratio test (χ^2 with 6 degrees of freedom (d.f.)). The incorporation of three harmonic terms is a flexible way to represent complex seasonal or circadian patterns (Albert and Hunsberger, 2005). Next, we tested whether seasonal patterns differed by center (three harmonics for seasonal pattern and eight centers) by using a linear mixed model and testing for a season-by-center interaction using a likelihood ratio test (χ^2 with 42 d.f.). These models were fit using maximum likelihood methods.

Linear mixed models (Searle *et al*., 1992) were used to partition the variance in serum carotenoid levels into explanatory components. Rather than modeling the seasonal patterns with fixed harmonic terms, as was done in the previous analysis, we modeled seasonal patterns with month-within-center random effects. This was done to compare the magnitude of seasonal variation with that of other sources of variation. Specifically, we included treatment group and year of measurement as fixed effects, and subject, center and month within center as random effects. The models were fit using restricted maximum likelihood, and the variance components and the proportion of variance attributed to each source were estimated for each carotenoid for the total population and stratified by gender. Total variance was the sum of all sources of variation, notably the sum of variation due to center, month of blood draw in center, and intra- and interindividual variation. Examination of potential confounding factors such as body mass index and serum cholesterol concentrations did not reveal any significant effects of these factors; therefore, they were not entered in the analysis.

The estimated variance components were used to address the question of the optimal number of repeated measurements for an individual in future similar studies. We used the results of the variance component analysis to calculate the ratios of (a) the variance of a typical subject's mean serum carotenoid levels based on r repeated measurements, compared with (b) the variance of a typical subject's mean serum carotenoid levels without measurement error (that is, the total variance–the analytical variance of the quality control samples). These ratios were estimated for each carotenoid.

Results

Baseline characteristics for all 901 participants with serum carotenoid measurements did not differ by arm of the PPT (all $P>0.20$) (Table 1). One participant in the intervention group did not have a baseline serum carotenoid measurement but had measurements for all subsequent years.

Models of within-center seasonal differences

The *P*-values of significance tests for within-center seasonal differences for serum carotenoids were estimated by mixed effects models (Table 2). Seasonal variation in total serum carotenoids differed markedly in three centers, notably Pittsburgh, Utah and Chicago, and marginally in a fourth center, Bethesda ($P=0.0008$, 0.0001, 0.037 and 0.052, respectively). Significant seasonal variation in total serum carotenoid levels within the four centers remained after adjustment for study year and for treatment arm of the study. Similarly, there was a marked seasonal difference in lutein/zeaxanthin concentrations in these four centers. Cryptoxanthin differed markedly in New York City, Chicago and Bethesda, whereas lycopene differed markedly in six of the eight study centers. α-Carotene differed in Oakland, Pittsburgh, Buffalo and Utah, and β-carotene differed in Pittsburgh, Utah and Chicago.

Models of between-center seasonal differences

Seasonal variation in concentrations of total serum carotenoids, lutein/zeaxanthin, lycopene, α- and β-carotene was significantly different across centers after adjustment for year, center and treatment effects. Serum carotenoid

concentrations did not show a distinct consistent seasonal pattern across the different centers; however, we did find that seasonality was more pronounced in the northern centers than the southern centers. Moreover, there were significant center-by-season interactions for concentrations of lutein/zeaxanthin ($P=0.002$), lycopene ($P=0.005$), α-carotene ($P=0.001$), β-carotene ($P=0.03$) and total carotenoids ($P=0.0005$) (Table 2). The center-by-season interaction for cryptoxanthin was borderline significant ($P=0.069$).

Components of variation

The major sources of variation in total serum carotenoid levels were the inter- and intraindividual variances (Table 3).

Table 1 Mean (±s.e.) or percent for demographic characteristics by treatment arm at baseline: PPT

	Total	*Intervention group*	*Control group*
Number	901	450	451
Age (years)	61.0 ± 0.3	61.1 ± 0.5	60.9 ± 0.5
Male sex (%)	67.7	66.9	68.5
>High school education[a] (%)	76.9	75.5	78.5
Current smoker (%)	12.7	13.3	12.0
Alcohol intake (g/day)	3.5 ± 0.2	3.4 ± 0.3	3.6 ± 0.3
Body mass index (kg/m^2)	27.6 ± 0.1	27.5 ± 0.2	27.6 ± 0.2
Plasma total cholesterol (mg/100 ml)	200.7 ± 1.2	202.0 ± 1.8	199.4 ± 1.6
Serum total carotenoids (μg/100 ml)[b]	91.2 ± 1.4	91.2 ± 1.9	91.2 ± 1.9
Lutein/zeaxanthin (μg/100 ml)	25.1 ± 0.4	24.9 ± 0.5	25.3 ± 0.6
Cryptoxanthin (μg/100 ml)	11.1 ± 0.3	11.0 ± 0.3	11.1 ± 0.4
Lycopene (μg/100 ml)	23.5 ± 0.4	23.3 ± 0.5	23.7 ± 0.5
α-Carotene (μg/100 ml)	6.6 ± 0.2	6.6 ± 0.3	6.5 ± 0.3
β-Carotene (μg/100 ml)	25.0 ± 0.7	25.4 ± 1.1	24.6 ± 0.9

Abbreviation: PPT, Polyp Prevention Trial.
[a]For one participant, education data were missing.
[b]For one participant, all baseline serum carotenoid data were missing.

Interindividual variances contributed between 61 and 70% of the variation in individual and total serum carotenoid concentrations, with the highest percents in lutein/zeaxanthin and β-carotene. Intraindividual variation ranged from 20 to 35% of all variation in individual and total serum carotenoids, with the highest percent exhibited by lycopene. Serum lycopene levels had the highest ratio of intra- to interindividual variance (0.57), whereas lutein/zeaxanthin had the lowest ratio (0.29). Center effects contributed from 2.6 to 9.5% of the variation in individual and total carotenoid concentrations, whereas the variation in month within center (which can be considered a proxy for seasonal effect in this analysis) had a minimal contribution to the overall variation in serum levels (range of 0.2–1.4%). Furthermore, analyses did not reveal gender differences in the percent contribution of the various sources to the components of variation models (data not shown).

The ratios of the variance of a participant's typical estimated mean serum carotenoid concentration with *r* measurements per participant, compared to the variance with no measurement error, appear in Table 4. The ratio is reduced greatly by increasing the number of measurements by 3–4 compared to just 1 measurement; however, a small additional reduction in the ratio is achieved by taking more than 4 or 5 measurements per participant.

Discussion

In the present study, seasonal patterns within centers and a center-by-season interaction appeared for the total and individual serum carotenoids. The major sources of variation in serum carotenoid levels were the inter- and intraindividual variances. Interindividual variances contributed between 61 and 70% of the variation in individual and total serum carotenoid concentrations, with the highest percents in lutein/zeaxanthin and β-carotene. Intraindividual variation ranged from 20 to 35% of all variation in individual and

Table 2 *P*-values for tests of seasonal variation in total and individual serum carotenoid concentrations by study center

Center[a]	*Individual and total carotenoids*					
	Lutein/zeaxanthin	*Cryptoxanthin*	*Lycopene*	*α-Carotene*	*β-Carotene*	*Total carotenoids*
Oakland	0.20	0.65	0.019	0.002	0.79	0.88
Pittsburgh	0.043	0.21	0.048	0.006	0.0004	0.0008
Winston-Salem	0.25	0.33	0.003	0.29	0.61	0.14
Buffalo	0.65	0.80	0.65	0.017	0.41	0.85
New York city	0.78	0.038	0.004	0.061	0.65	0.32
Salt Lake city	0.009	0.002	0.005	0.002	0.006	0.0001
North Chicago	0.047	0.037	0.26	0.096	0.047	0.038
Bethesda	0.001	0.099	0.007	0.20	0.73	0.052
Interaction[b]	0.002	0.069	0.005	0.001	0.034	0.0005

[a]Tests for seasonal trends within center were based on likelihood ratio tests for the inclusion of three pairs of harmonic terms (χ^2 with 6 d.f.).
[b]Global test for interaction, based on likelihood ratio tests of whether the seasonal patterns were the same across all centers (χ^2 with 42 d.f.).

Table 3 Variance component analysis, the percentage of total variation in serum carotenoid levels attributed to each component and the ratio of intra- to interindividual variation: PPT

	Interindividual		*Center*		*Month-in-center*		*Intraindividual*		*Intraindividual/interindividual*
	Variance	*%*	*Variance*	*%*	*Variance*	*%*	*Variance*	*%*	*ratio*
Lutein/zeaxanthin	0.15	70.5	0.017	8.1	0.0013	0.6	0.045	20.7	0.29
Cryptoxanthin	0.24	66.6	0.022	6.1	0.0008	0.2	0.098	27.1	0.41
Lycopene	0.17	61.0	0.008	2.6	0.0040	1.4	0.10	35.0	0.57
α-Carotene	0.38	67.5	0.054	9.5	0.0061	1.1	0.12	21.9	0.32
β-Carotene	0.40	70.7	0.044	7.8	0.0026	0.5	0.12	21.0	0.30
Total carotenoids	0.13	67.5	0.019	9.5	0.0013	0.7	0.044	22.3	0.33

Abbreviation: PPT, Polyp Prevention Trial.

Table 4 Ratios of the variance of a participant's estimated mean serum carotenoid concentration with *r* measurements per participant compared to the variance with no measurement error per participant

	Number of measurements per subject (r)							
	1	*2*	*3*	*4*	*5*	*6*	*7*	*8*
Lutein/zeaxanthin	1.30	1.15	1.10	1.07	1.06	1.05	1.04	1.04
Cryptoxanthin	1.38	1.19	1.13	1.10	1.08	1.06	1.05	1.05
Lycopene	1.68	1.34	1.23	1.17	1.14	1.11	1.10	1.08
α-Carotene	1.34	1.17	1.11	1.08	1.07	1.06	1.05	1.04
β-Carotene	1.35	1.17	1.12	1.09	1.07	1.06	1.05	1.04
Total carotenoids	1.34	1.17	1.11	1.09	1.07	1.06	1.05	1.04

These ratios are given by the formula $R(r) = (v_b + v_e/r)/v_b$, where v_b denotes the estimated interindividual variance, v_e denotes the estimated intraindividual variance and r denotes the number of measurements per individual.

total serum carotenoids, with the highest percent exhibited by lycopene. Serum lycopene had the highest ratio of intra- to interindividual variance (0.57), whereas lutein/zeaxanthin had the lowest ratio (0.29). Center effects contributed from 2.6 to 9.5% of the variation in individual and total carotenoid concentrations, whereas the variation attributed to season (that is, the month-within-center variation) had a minimal contribution to the overall variation in serum levels (range 0.2–1.4%).

Our findings can be compared to the results from three earlier reports (Tangney *et al*., 1987; Talwar *et al*., 2005; Block *et al*., 2006). Talwar *et al*. reported carotenoid levels weekly for 22 weeks in 14 men and women, aged 20–54 years (Tangney *et al*., 1987; Talwar *et al*., 2005). Their range in the intra- and interindividual variances was from 0.13 (lutein) to 0.24 (α-carotene) and from 0.31 (lutein) to 0.65 (α-carotene), respectively. The ratio of the two variances was from 0.38 in β-carotene to 0.69 in lycopene. Block *et al*. (2006) reported two sets of carotenoid concentrations from the blood of 206 individuals, collected 2–4 weeks apart. The patients were aged 45 years on average, 39% men, 54% White and 65% smokers. The range in the intra- and interindividual variances was from 0.117 (cryptoxanthin) to 0.665 (lycopene) and from 0.26 (cryptoxanthin) to 1.33 (lycopene), respectively. The ratio of the two variances was from 0.317 (β-carotene) to 0.499 (lycopene). In a study of 24 men, similar patterns of variation in serum carotenoids appeared using two fasting bloods collected 1 week apart (Tangney *et al*., 1987; Talwar *et al*., 2005). In both investigations, the authors estimated 3–4 carotenoid measurements to limit attenuation of regression coefficients to 10% (Tangney *et al*., 1987; Block *et al*., 2006). The range in the intra- and interindividual variances for the PPT was comparable to previous research (Tangney *et al*., 1987; Talwar *et al*., 2005; Block *et al*., 2006). Yet PPT individuals had an adenomatous polyp within 6 months before the trial, thereby providing for the first time the components of variation in serum carotenoid concentrations in a high-risk population for colorectal cancer.

To what extent does season influence serum carotenoid concentrations? Seasonal patterns in reported fruit and vegetable intake or carotenoid intake occur in markedly different magnitudes by population, even after adjustment for age, body mass index, alcohol, smoking and other recognized covariates (Bates *et al*., 1984; Ziegler *et al*., 1987; Rautalahti *et al*., 1993; Olmedilla *et al*., 1994; Cooney *et al*., 1995; Forman *et al*., 1999). Seasonal effects are relatively strong in countries without extensive food preservation and transportation systems (Bates *et al*., 1984; Forman *et al*., 1999), and therefore access to carotenoid-rich foods may be season dependent. Economic diversity would also increase between-person variation (Willett, 1998) and day-to-day variation may be particularly large in developing countries if expensive carotenoid-rich foods are affordable occasionally. Although season of dietary and blood collections may not affect population estimates, it may contribute to the misclassification of individuals if intake/blood levels are treated categorically or in a continuous manner (Subar *et al*., 1994). The effect of season was significant in 3–6 PPT centers depending on the carotenoid, but seasonal contribution to variation in serum levels was more minimal than other sources. The fairly homogenous socioeconomic status of PPT participants and the availability of carotenoid food sources throughout the year may have reduced the effects of seasonality.

Carotenoids are colorful fat-soluble pigments that are synthesized in nature by photosynthetic microorganisms

and plants. Several biomarkers of carotenoid intake have been used to examine the carotenoid–disease association. Serum carotenoid concentrations are sensitive to dietary intake, as they are not closely regulated by homeostatic mechanisms. Significant diet–serum β-carotene correlations range from 0.13 (Goodman *et al*., 1966) to 0.51 (Yong *et al*., 1994). Plasma levels of other carotenoids are also reasonably correlated with dietary intakes: 0.58 for α-carotene, 0.49 for β-cryptoxanthin, 0.31 for lutein/zeaxanthin and 0.50 for lycopene (Yong *et al*., 1994). Adipose tissue carotenoid concentrations reflect storage depot levels and have low correlations with dietary intake assessed by a food frequency questionnaire in Costa Rica ($r = -0.06$ to 0.20) (Kabagambe *et al*., 2005) and in the Netherlands ($r = 0.20$) (Kardinaal *et al*., 1995). Indeed, dietary carotenoid intake had higher correlations with serum than adipose tissue concentrations in a comparative study (Irwig *et al*., 2002). In the PPT, adipose tissue samples were not collected to compare the components of variation in adipose tissue with blood levels.

The results of our variance component analysis can assist in the design of future studies and, in particular, offer the ability to measure a person's nutritional biochemical status within an array of study subjects. Specifically, one can determine the number of measurements per participant to reduce the variance of the participant's mean serum carotenoid concentration. Once the intra- and interindividual variances are estimated, the ratio of the variance of the typical participant's estimated mean with r measurements per participant, compared to the variance with an infinite number of measurements, that is, no measurement error per participant, can be calculated. For most carotenoids, approximately four measurements per participant are required to have a variance of the typical participant's estimated mean no more than 10% larger than the variance with no measurement error; however, seven serum measurements of lycopene are required using the 10% cutoff. Based on our analysis, for the majority of carotenoids, there is a small payoff in taking more than four or five measurements per participant. Investigators may consider other factors, including the degree of accuracy in their studies, and the cost of and participants' burden from repeated measures to determine the number of biospecimen collections. For many epidemiologic studies, obtaining a highly accurate measure of individual intake and serum concentrations by using repeated measures is simply beyond practical possibilities.

In summary, components of variation in serum carotenoid concentrations were calculated from a subcohort of 901 PPT participants, all of whom had an adenomatous polyp before entry in the trial. Among high-risk individuals for colorectal cancer, intra- and interindividual variations in specific and total carotenoids had the largest contributions to variation in carotenoid concentrations. Season and center-by-season effects provided less, albeit significant, contributions to the variation in serum carotenoid concentrations than person effects. Similar research would be of interest in cohort studies of populations with limited economic resources and other gastrointestinal disorders than in the PPT, especially using more recent laboratory assays that can detect additional minor carotenoid peaks than our findings.

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Appendix

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