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Dermal carotenoid measurement is inversely related to anxiety in patients with breast cancer

David G Li¹, Gabrielle LeCompte², Lev Golod³, Gary Cecchi⁴, David Irwin⁴, Alden Harken⁵, and Amy Matecki²

¹Predoctoral Clinical Research (TL1), Tufts University School of Medicine, Boston, MA, USA

²Integrative Medicine, Highland Hospital, Oakland, CA, USA

³University of California, Berkeley, Berkeley, CA, USA

⁴Hematology and Oncology, Alta Bates Summit Medical Center, Berkeley, CA, USA

⁵Surgery, Highland Hospital, Oakland, CA, USA

Abstract

Breast cancer is the most prevalent malignancy among women worldwide. Increased oxidative stress and poor subjective health outcomes have been associated with increased risk of cancer recurrence and metastasis, but few studies until now have explored the relationship between oxidative stress and chronic stress/anxiety. This study aims to examine the association between anxiety and a potential dermal correlate of oxidative stress in patients with breast cancer. 102 breast cancer patients were enrolled in a cross-sectional study at Highland Hospital, a county hospital in Oakland, California. Each participant's skin carotenoid score (SCS), a potential dermal correlate of oxidative stress, was recorded via Raman spectroscopy. Patient demographics, breast cancer stage, and subjective health measures (anxiety and self-rated health) were ascertained. Multivariate linear regression analysis was performed to quantify any associations between SCS and the above health correlates. Higher levels of skin carotenoids were associated with decreased severity of anxiety, lower BMI, increased servings of vegetables/fruits in daily diet, Hispanic race, lower educational status, and nonsmoking status. Severity of anxiety as graded by the GAD-7 was inversely associated with dermal carotenoid measurements via SCS. Conclusions. Increased levels of oxidative stress as quantified by SCS is associated with greater severity of anxiety. Because chronic stress has been associated with tumor progression, increased recurrence rates, and

Correspondence to: David G Li, Tufts University School of Medicine, Boston, Massachusetts 02111, USA; david.li@tufts.edu.

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increased metastatic risk in breast cancer, non-invasive dermal carotenoid measurements could be used as a novel objective correlate of subjective health during cancer treatment.

INTRODUCTION

Breast cancer is the most common malignancy in women worldwide and affects 12% of women in the USA.¹² The etiology of breast cancer is multifactorial, with intrinsic propensity (inherent risk and genetic risk factors) and extrinsic offenders (eg, oxidative stress, increased estrogen levels) playing essential roles synergistically.³ Oxidative stress, an imbalance between oxidizing free radicals and antioxidants, can trigger DNA damage and has been widely implicated in breast cancer carcinogenesis.³ DNA damage from oxidative stress can eventually override tumor suppressors gene repair mechanisms, leading to faulty DNA integration and tumorigenesis.⁴

Carotenoids, nutrients with antioxidant properties derived from fruits and vegetables, have been shown to protect against oxidative DNA damage. Previous research has demonstrated decreased carotenoid levels to be associated with decreased fruit/vegetable intake, positive smoking status, and higher body mass index (BMI) measurements.^{5–8} Furthermore, previous studies have suggested that higher serum carotenoid levels are associated with decreased risk of invasive breast cancer⁹ and with reduced risk for breast cancer recurrence in women previously treated for breast cancer.¹⁰

The current gold standard of measuring serum carotenoids is through high performance liquid chromatography, an invasive and costly process.¹¹¹² Skin carotenoid levels, a non-invasive and affordable technique, have been found to correlate with serum carotenoid levels with a correlation coefficient of $R=0.77$ between skin carotenoid levels and serum carotenoid levels.^{13–15} Evidence has suggested that skin carotenoid levels derived from Raman spectroscopy may provide a more accurate representation of true carotenoid status compared with serum carotenoid levels, as skin carotenoids are less affected by acute postprandial fluctuations.^{1316–19}

In addition to objective health outcomes, there has been an increasing body of evidence suggesting a link between subjective health assessments and the risk of cancer. Increased self-reported stress levels can trigger a myriad of changes that could hasten metastasis among patients with cancer. In addition, increased perceived stress is also positively correlated with increased recurrence risk among patients in remission.²⁰²¹

This study has two primary aims: (1) to assess skin carotenoid score (SCS) levels and two measures of subjective health—perceived anxiety and self-reported health and (2) to assess the association between SCS levels and breast cancer stage, fruit/vegetable intake, smoking status, and BMI using SCSs in place of the more invasively acquired plasma carotenoid levels.

METHODS

Cross-sectional study design

Patient recruitment—We asked all current and previous female patients with breast cancer, aged 18–80 years, who came in for treatment or follow-up at Highland Hospital/ Alameda Health System’s Hematology/Oncology Clinic, Infusion Center or Breast Clinic between December, 2015 and April, 2016 to participate in this cross-sectional study. A total of 137 patients (92% response rate) were recruited. The Institutional Review Board of Highland Hospital in affiliation with UCSF approved this cross-sectional study.

SCS measurement—Each patient’s SCS was acquired during one of the routine follow-up clinic visits via a Raman Spectroscopy machine (NuSkin Enterprises, Provo, Utah, USA). The technique of Raman Spectroscopy used to measure SCS in our study has been described, explored, and validated previously in a variety of clinical settings.^{13–15}

Health survey—We administered a survey to collect demographic, treatment, symptom, health behavior, and health status information. Demographic questions inquired about patient age, race (Non-Hispanic Black, Hispanic, Asian, White, Other), educational attainment (less than high school, high school, associate’s degree, bachelor’s degree, master’s degree, doctorate, other), stage of breast cancer (Stage 0, Stage 1, Stage 2, Stage 3, Stage 4), smoking status, diet, weight, and height. Self-rated health (outstanding, good, some chronic issues, or poor) was used to obtain each patient’s own assessment of health according to his/her individual definition of health. Self-reported anxiety was assessed using the standard Generalized Anxiety Disorder 7-item (GAD-7).²² We also acquired a confirmation of patients’ medical information from patient records.

Data analysis—We performed statistical analyses using STATA V.14.2 (Stata). We analyzed associations between SCS scores according to demographic and health-related characteristics using bivariate analyses with analysis of variance. We examined the correlation between SCS and cancer stage, smoking status, BMI, self-reported anxiety, and self-rated health using multivariate linear regression analysis.

RESULTS

Patient demographics

Patients were, on average, 55±10 years of age. All patients were women, with Hispanic patients representing the largest proportion of the study population (33%; table 1). Race/ethnicity was significantly associated with SCS ($F=4.73$, $p=0.0016$). The level of highest education was also significantly associated with SCS ($F=5.73$, $p=0.0182$).

Health-related characteristics

Most patients were overweight, with a mean BMI of 29±7 and a median of 28 (table 2). BMI was inversely correlated with SCS, a finding consistent with previous literature ($r=-0.252$, $p<0.05$). Breast cancer stage, contrary to our hypothesis, did not appear to be associated with SCS ($r=-0.042$, $p=0.67$). Moreover, current medication status (receiving chemotherapy,

radiation therapy, hormonal therapy, etc) did not appear to be associated with SCS ($r=0.06$, $p=0.54$). Of note, 7% of patients were actively receiving radiation therapy, 7% were within 1 year post surgery, 11% were actively receiving chemotherapy and 55% were on some form of hormonal medications. The results of bivariate analysis show that health behaviors were associated with SCS: active smoking status and vegetable/fruit intake (carotenoid source) intake were associated with SCS ($r=-0.2145$, $p=0.030$; $r=0.295$, $p=0.024$, respectively) (table 2).

Multivariate analyses

In multivariate linear regression analyses, BMI ($\beta=-0.46$, 95% CI -0.85 to -0.06 ; $p=0.024$), being an active versus non-active smoker ($\beta=-8.00$, 95% CI -13.83 to -2.18 ; $p=0.008$), and vegetable/fruit intake ($\beta=5.10$, 95% CI 1.28 to 8.93 ; $p=0.010$) were associated with lower SCS levels (see table 2). Self-reported anxiety score ($\beta=-0.48$, 95% CI -0.91 to -0.06 ; $p=0.027$) was associated with SCS (table 4), and self-rated health scores ($\beta=21.09$, 95% CI 18.03 to 49.99 ; $p=0.002$) were associated with SCS (table 5).

DISCUSSION

In this cross-sectional study of 102 patients with breast cancer presenting for routine treatment or follow-up, we found fruit and vegetable intake to be positively associated with SCS, while BMI and educational attainment were inversely associated with SCS. Active smoking status and African American ethnicity were also associated with decreased SCS. Moreover, severity of anxiety via GAD-7 was inversely correlated with SCS measurements. Breast cancer staging, however, was not associated with SCS measurements in this cohort of patients.

The link between dietary carotenoid intake and overall health outcomes has been studied extensively in the past decade, and our findings in this study are consistent with previous findings. BMI has been found to inversely correlate with SCS (BMI 30.1 ± 7.1 kg/m² in SCS $>25,000$ Raman counts (RC) vs 26.7 ± 4.6 kg/m² in SCS $<25,000$ RC; $p=0.0003$) based on a study by Holt *et al*⁸²³ as well as with low plasma carotenoid measurements.²⁴ This finding was illustrated in our study. Moreover, the relationship between smoking and lower plasma carotenoid levels has been extensively documented (13% smokers in SCS group $<25,000$ RC vs 1% in SCS group $>25,000$ RC)⁸²⁵; smoking-induced free radical damage may result in lower carotenoid levels.²⁶²⁷ Our study demonstrated similar findings (25% smokers total with mean SCS of 25,530 RC vs 75% non-smokers total with mean SCS of 32,300 RC). Consumption of fruit/vegetable (measured in servings) has also been shown to be positively associated with SCS levels (3.0 ± 2.1 servings per day in SCS $<25,000$ RC vs 4.7 ± 2.7 servings per day in SCS $>25,000$ RC, $p<0.001$).⁸²³ Likewise, our study demonstrated a SCS of 36,500 RC in those with 4 or more servings of fruit/vegetables per day, whereas those who took less than 4 servings of fruit/vegetables per day had a mean SCS of 29,600 RC.

There was a statistically significant inverse correlation between educational attainment and SCS. We initially hypothesized that less-educated subjects are less likely to be cognizant of the relationship between nutrition and oxidative stress. Therefore, they would be more likely to have higher BMI and thus lower SCS. However, based on our cross-sectional study, an

opposite trend was seen. Finally, Hispanic respondents in our study had the highest SCS, while Non-Hispanic Black respondents had the lowest SCS. This could be explained by dietary habits differing between these cultural groups, but a more expansive and nationally representative sample size may be needed to assess true differences in SCS between racial demographics.

Anxiety severity via the GAD-7 was inversely associated with SCS, suggesting a relationship between subjective health outcomes and dermal carotenoid levels. While this appeared to be a novel finding at the time of the study, a recently published (2017) double-blind, placebo-controlled trial demonstrated macular carotenoid (lutein, zeaxanthin, and meso-zeaxanthin) supplementation to coincide with decreased levels of psychological stress and improved emotional health, lending support to our foremost hypothesis during our study enrolment period.²⁸

Contrary to expectations, self-rated health was inversely associated with SCS. This finding may be explained by a heightened sense of health awareness—an illness-distorted perception of health—among a subset of patients after receiving a breast cancer diagnosis. Plausibly, these patients may be more likely to implement lifestyle changes, such as increasing their fruit/vegetable intake, quitting smoking, and losing weight (lowering BMI), thus being more likely to have higher SCS levels not manifested in self-rated health assessment scores.

Previous studies have documented increased rates of metastasis and cancer recurrence with higher levels of subjectively reported stress, which may suggest a relationship between oxidative stress and anxiety.²⁰²¹ The specific mechanism may be multifactorial, but chronic stress appears to induce immunological alterations leading to changes in leukocyte count, natural killer (NK) cell activity, and stress hormone levels.²⁹ Because patients undergoing chemotherapy have been shown to sustain increased levels of anxiety and decreased quality of life, could the resulting impact on emotional health worsen treatment outcomes and increase the risk of metastasis/recurrence in a positive feedback-like fashion³⁰³¹? While there is a movement toward incorporation of complementary care to improve psychological health in patients with cancer, few evidence-based methods exist to monitor the impact these interventions have on subjective health.³²³³

Limitations

The results from our study do not indicate a significant correlation between breast cancer staging and SCS ($\beta=0.49$, 95% CI -1.82 to 2.80). One explanation for this lack of correlation could be that only 7% and 11% of patients were receiving radiotherapy and chemotherapy, respectively, treatments that may rely on free radical generation for efficacy.³⁴ Patients with higher cancer stages and worse prognosis will often have more aggressive chemotherapy or radiotherapy regimens, and the excess free radical generation may lower overall carotenoid levels. This is consistent with the dynamics of free radical theory as carotenoids function in vivo as free radical quenchers.³⁵

Moreover, despite our finding regarding the association between dermal carotenoid scores and subjective anxiety, the exact causal relationships between SCS and subjective health

correlates could not be demonstrated given the study's cross-sectional nature. Finally, because the study's population was derived from a county hospital, the generalizability of our study's results may be limited by the homogeneity of the studied patient population.

CONCLUSIONS

To our knowledge, no previous study has demonstrated an association between anxiety and dermal carotenoid levels, as measured by the GAD-7 and SCS, respectively. Additionally, our findings further affirm that lower dermal carotenoid scores are associated with higher BMI, positive smoking status, and lower fruit/vegetable intake. Breast cancer staging was not associated with SCS in this cross-sectional study. Future research could involve a larger, prospective study to better assess SCS levels in patients as they progress through cancer treatment to clarify whether SCS is associated with cancer stage or the aggressiveness of treatment regimen. Moreover, randomized controlled studies may be needed to investigate the potential causal mechanisms between anxiety and carotenoids. The findings may facilitate the development of novel tools to quantify subjective anxiety levels in patients with cancer, allowing patients and physicians to mindfully monitor stress and anxiety levels during cancer treatment.

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Significance of this study

What is already known about this subject?

- Carotenoids, nutrients with antioxidant properties derived from fruits and vegetables, may harbor protective effects against oxidative DNA damage.
- Higher body mass index, positive smoking status, and lower fruit/vegetable intake generally correlate with lower levels of serum carotenoid measurements.
- Oxidative stress levels and poor subjective health have been positively associated with the risk of cancer recurrence and metastasis.
- Supplementation with macular carotenoids (lutein zeaxanthin and meso-zeaxanthin) has been found to coincide with lower levels of psychological stress and improved emotional health.

What are the new findings?

- Anxiety severity (quantified by Generalized Anxiety Disorder 7-item) in a cohort of patients with breast cancer is inversely associated with dermal carotenoid measurements via skin carotenoid score (SCS).
- Self-reported health quality measurements were inversely associated with dermal carotenoid measurements, an unexpected finding.
- In our cross-sectional study, there was no significant association between breast cancer stage and dermal carotenoid measurements via SCS.

How might these results change the focus of research or clinical practice?

- Our study lays the foundations for a larger prospective study aimed at investigating and refining the potential utility of dermal carotenoids as an objective biomarker of subjective health during cancer treatment.

Table 1

Dermal carotenoid measurements and subject demographics

Demographics [†]	N	Mean subgroup SCS (SD) [*]
Age	55 (10)	
Race/ethnicity		
White	8	32.75 (21.35)
Non-Hispanic Black	30	22.01 (9.34)
Asian	29	33.92 (14.81)
Hispanic/Latino	34	34.71 (11.42)
Other	1	35
Educational attainment level		
Less than high school	28	37.88 (15.58)
High school graduate	28	29.40 (14.95)
Some college	24	24.89 (8.68)
Associate degree	8	28.87 (14.96)
BA	10	31.4 (9.37)
Masters	4	23 (5.13)
PhD	0	NA

* SCS value is the measurement \times 1000, as per convention.

[†] SCS values did not differ significantly between race/ethnicities and those of different educational attainment levels at $p=0.05$.

SCS, skin carotenoid score.

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Table 2

Dermal carotenoid measurements and physical health correlates

Physical health variable	N	Mean subgroup SCS (SD)*
Body mass index	29.21 (6.69)	30.57
Breast cancer stage		
0	7	29.86 (16.26)
1	24	30.08 (14.81)
2	39	29.96 (11.06)
3	12	33.39 (17.42)
4	20	30.92 (15.52)
Medication status		
Active	56	31.35 (15.51)
Not active	46	29.63 (11.56)
Smoking status		
Active	26	25.53 (14.31)
Not active	76	32.30 (13.32)
Veggie/fruit servings per day		
0–1	31	24.96 (8.27)
2–3	56	32.10 (14.39)
4–5	13	35.33 (16.50)
5+	2	44 (29.23)

* SCS value is the measurement \times 1000, as per convention. p Value ranges are given in table 3.

SCS, skin carotenoid score.

Table 3

Dermal carotenoid measurements and severity of anxiety

SCS range [*]	#Subjects/group	N with GAD	10 [†]	% With anxiety
10–20	26	10		38%
21–30	32	7		22%
31–40	23	9		39%
41–50	13	4		31%
51–60	3	0		0%
61–70	3	0		0%
71–80	2	1		50%

* SCS value is the measurement \times 1000, as per convention. p Value ranges are given in table 3.

[†] Anxiety with 10 or higher on GAD-7 (moderate anxiety and above).

GAD-7, Generalized Anxiety Disorder 7-item; SCS, skin carotenoid score.

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Table 4

Dermal carotenoid measurements and self-rated health

Self-reported health quality level	N	Mean SCS (SD) *
Excellent	10	31.67 (16.06)
Good	45	29.16 (11.79)
Some chronic issues	40	30.18 (13.83)
Poor	7	40.29 (21.04)

* SCS value is the measurement \times 1000, as per convention.

SCS, skin carotenoid score.

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Table 5

Linear regression assessing the association between SCS and patient health characteristics (beta coefficients (β) and 95% CIs are presented)

	β	(95%CI)	p Value
Physical health			
BMI	-0.46*	(-0.85 to 0.06)	0.024
Breast cancer stage	0.26	(-1.92 to 2.44)	0.814
Medication status	3.40	(-1.60 to 8.40)	0.181
Health behaviors			
Smoking status	-8.00*	(-13.83 to 2.18)	0.008
Veggie/fruit servings per day	5.10*	(1.28 to 8.93)	0.010
Subjective health			
Self-reported anxiety score (GAD)	-0.48*	(-0.91 to 0.06)	0.027
Self-rated health (reference=excellent)			
Good	6.77	(-2.55 to 16.08)	0.152
Some chronic issues	9.11	(-0.43 to 18.64)	0.061
Poor		(18.03 to 49.99)	0.002

*
p<0.05,

**
p<0.01.

BMI, body mass index; GAD, Generalized Anxiety Disorder; SCS, skin carotenoid score.