Title

Starving for a Degree: The Relationship Between Diet, Cognition, and Food Insecurity in the College Student Population

Author

Radtke, Marcela DeVon

Publication Date

2023

Peer reviewed|Thesis/dissertation

Starving for a Degree: The Relationship Between Diet, Cognition, and Food Insecurity in the College Student Population

By

MARCELA DEVON RADTKE

DISSERTATION

Submitted in partial satisfaction of the requirements for the degree of

DOCTOR OF PHILOSOPHY

in

Nutritional Biology

in the

OFFICE OF GRADUATE STUDIES

of the

UNIVERSITY OF CALIFORNIA

DAVIS

Approved:

_____

Francene M. Steinberg, PhD, RD, Chair

_____

Rachel E. Scherr, PhD

_____

Nancy Keim, PhD

Committee in Charge

2023

Acknowledgements

A close mentor once told me “Life doesn’t pause while you’re in graduate school.” With the loss of my dad, the COVID-19 pandemic, and every moment in-between, big or small, good or bad, it has been my family, friends, and mentors that have motivated and encouraged me to become the now, Dr. Marcela D. Radtke. Life didn’t pause, but these special people along the way made it possible to keep moving forward, and for that I thank you all.

To my dissertation committee, Dr. Rachel Scherr, Dr. Francene Steinberg, and Dr. Nancy Keim, thank you. Your constructive feedback and support throughout this process has been invaluable. It has been an honor to work with and learn from such amazing scientists and opinionated scholars throughout my PhD experience. I would also like to thank my Qualifying Exam Committee, Dr. Carl Keen, Dr. Nancy Keim, Dr. Suzanna Martinez, Dr. Jon Ramsey, and Dr. Patricia Oteiza for challenging me to think critically and prepare me for a future career as an independent researcher. Nutrition is a multidisciplinary science and each of you contributed to my understanding of the application of nutrition through different scientific lens and approaches.

Mentors

To my mentor, Dr. Rachel Scherr, thank you. To say you have been an influential person in my life would be an understatement. You have helped shape every aspect of the person I am today. You saw a potential in me that I was unable to see for myself. You have challenged me, supported me, and advocated for me for close to a decade. You supported my desire to bring something new to the GGNB by integrating the fields of nutrition, medicine, and public health. You stopped at nothing to leverage the funding and resources needed to complete my research goals. You allowed me to have a voice in the study design process, supported my ideas for incorporating technology and industry into my research, and developed my skills in the art that is

scientific writing. It was you who prepared me to enter the rigorous field of academia, and as a mentor, I hope I am able to provide my future students with the mentorship you provided me. In addition to all of your academic and research expertise, there are so many personal qualities you possess that I hope to embody half as well as you do. I admire your loyalty, perseverance, and dedication to those around you. Seeing you become a wife, a mother, a business owner, among the many other incredible life and career accomplishments, you truly embody the phrase "everything happens for a reason." You will forever be my mentor, my advocate, my confidant, and my family.

To my mentor, Dr. Francene M. Steinberg, thank you. You have welcomed me with open arms and supported every academic, extracurricular, and personal endeavor throughout my graduate experience. The knowledge, expertise, and support you have provided me with are attributes I will take with me to every stage of my life. You approach both leadership and mentorship with the utmost grace, and I hope I can follow in your path as I navigate a career in academia. You have been a phenomenal advocate for me in every aspect of this journey and it has been an absolute pleasure to learn from you, Francene.

To my mentor, Dr. Sheri Zidenberg-Cherr, thank you. You created a lab culture of rigorous nutrition science, supported by positivity and inclusivity, not to mention well-dressed and well-celebrated. You have provided me with insightful feedback on academic and personal endeavors, encouraging me to think critically every step of the way. As I navigate creating a lab of my own, I will absolutely be implementing things "the Sheri way." Thank you for supporting me always.

To my mentor, colleague, friend, and so much more, Dr. Lyndsey D. Ruiz, thank you. We have been through it all! I would not have survived this experience without you; You

supported me in ways I never even knew I needed. You were the first person I called when my world shattered, and you went above and beyond to put the pieces of me back together. Watching you pursue your passion to help graduate students navigate this journey has been an absolute inspiration, as I can speak firsthand to how many lives you will change for the better, as mine has been one of them already. You have been such a pivotal piece in my life, far beyond the PhD experience, and I cannot thank you enough for everything, Lyndsey.

To the one and only Dr. Debbie S. Fetter, thank you. I cannot imagine what this experience would have been like if I had not met you! You introduced me to the world of nutrition science and your love, passion, and dedication made it impossible to leave. It has been a highlight of my graduate experience to work with you and I have learned so much about the type of instructor and mentor I want to become from you. Thank you for embracing the UC Davis SEED Scholars and for promoting an inclusive learning environment.

To my Davis mom, Beth Foraker, thank you. I like to think our paths crossed for a reason so many years ago – you and your family have truly made Davis my home. You have showed me and so many others the true definition of unconditional love. Working with you and the UC Davis SEED Scholars is one of my proudest life accomplishments. You have provided a voice and sense of belonging for so many individuals who have been previously silenced. The experience and knowledge you have shared with me over my time at UC Davis has helped me become a multifaceted, adaptable, and compassionate instructor, researcher, mentor, and advocate for my future students.

To the women of the Zidenberg-Cherr/Scherr Lab Group and the Steinberg Lab Group, thank you. You are all such wonderfully unique people and it was an absolute privilege to work with you over the years. A special thanks to Melanie Gerdes, Dr. Anna M. Jones, and Dr. Britt

Loofbourrow – you have all been such monumental role models for me in different ways and I cannot express the gratitude I feel toward this group in shaping me to the person I am today. I could not have completed this work without my amazing undergraduate interns, to whom I immensely thank.

Friends

To my roommate (and legally defined "domestic partner"), Dani, thank you. Living with you throughout this experience has brought such a light to my life. Your positive energy is contagious, and you constantly ground me in remembering the aspects of life that are important. You helped me prepare for the Preliminary and Qualifying Exams, you have served as my editor-in-chief for every application, and celebrated all of my successes as your own. You have endured some of life's most challenging moments by my side and somehow, we made it out that much stronger. You are a vibrant ray of sunshine and have made my PhD experience that much brighter.

To my resident "Chateau" guest, Bianca, thank you. You are wise beyond your years, B. Our car commute chats were the best part of so many days. You served as an amazing listener, prompted thought-provoking questions, and challenged me to be the best version of myself always. You care so deeply about those around you, and I am so unbelievably grateful to have had your support throughout this experience.

To my long island queen and PhDiva, Michelle Zuelch, thank you. It has been an absolute dream to learn and work alongside you. You find the positive in every situation, both inside and outside the lab, and I genuinely cannot imagine going through this experience with anyone else. Our happy hours were the highlight of my week and I know that you will be a forever friend, Michelle.

Family

"Radtke's don't give up." A motto I learned from you Mom - I love you and thank you. You have supported me in every stage of this incredibly challenging journey. You have been there to celebrate every fellowship, award, job opportunity, and manuscript submission, while also being there to help guide me through every obstacle. You constantly reminded me to think and act with empathy, as everyone is going through their own version of hardship. You have inspired so many of my past, present, and future goals. Your patience and advocacy for inclusion motivated my work in developing an inclusive opportunity for people with intellectual disabilities. Your ability to connect with people from diverse racial and sociodemographic backgrounds excited me to conduct research in underserved, marginalized communities. You have and will continue to be the most influential person in my life, and I am so proud to be your daughter.

To my sister Caitlin, thank you. Words cannot describe how much I cherish our relationship. Not only am I lucky enough to call you my sister, but you are also my best friend. You pushed me to pursue my goals and never let a day go by where you did not tell me how proud you were of my accomplishments. I look up to you in every way, sis. I also want to thank my brother-in-law, Matt, for everything you have done and currently do for our family. To the light of my world, Cora and Robbie, thank you for making Titi's day better each and every day.

To my person, Jacob, thank you. If actions speak louder than words, I wish I could equally express to you the infinite ways you have shown me unconditional love and support throughout this experience. You have been my biggest supporter, showing up to all my events, even when I tell you it's not important. Thank you for allowing me to practice my phlebotomy skills on you, thank you for listening (and re-listening) to me practice presentations, and thank

you for supporting my success in every way. You have encouraged me to be a better me, and because of that, I am a better researcher, mentor, friend, scholar, author, partner, and so much more.

And lastly, to my dad, you would be so proud. There have been so many moments throughout this journey that I wish I could have shared with you. With every major accomplishment, I have felt you there, cheering me on. Your love of food and cooking is now eternally represented through my degree in Nutritional Biology and I could not be more proud to carry on your name as Dr. Radtke.

TABLE OF CONTENTS

ABSTRACT

College students are at an increased risk for experiencing acute or chronic food insecurity (FI) due to the underlying economic pressures of higher education, including the expensive cost of tuition and inflated cost of living in proximity to a college campus. Driven by the newfound autonomy over dietary choices and purchasing behaviors, and often perpetuated by limited nutrition knowledge and financial literacy, the prevalence of FI in the college student population is four times higher than the national average. The experience of FI may be accompanied by negative physiological and mental health outcomes, such as nutrient deficiencies, overweight or obesity, poor sleep, and increased stress, anxiety, and depressive symptoms, among others. With the emergence of FI as a public health concern, research in this field is limited, and populations of interest have predominately focused on children and older adults. Therefore, the objective of this dissertation was to explore the diet-related and cognitive outcomes of FI in a diverse population of university students using innovative biomarkers and novel technologies. This dissertation provides novel insight into the potential for strategic community-based interventions aimed at improving food security status and health outcomes in a population at increased risk for experiencing FI.

The first aim of this dissertation was to validate assessment tools for identifying changes in diet-related biomarkers with the experience of FI. The Veggie Meter®, a device that implements pressure-mediated reflection spectroscopy to identify carotenoids in adipose, and Diet ID™, a novel image-based dietary assessment tool, were assessed against different biomarkers of dietary intake. After conducting a systematic review of existing literature (n = 29) comparing methods of reflection-spectroscopy against plasma carotenoids and/or validated tools for dietary intake, the Veggie Meter® produced moderate to strong correlations (average p =

0.76; $p < 0.001$), whereas other modes of spectroscopy were weakly correlated. Similarly, Diet ID™ was compared to other forms of dietary intake, including plasma carotenoids, skin carotenoids, and 24-hour NDSR dietary recalls in a diverse population of university students (n = 42). Diet ID™ was correlated to nearly all nutrients of interest, including macronutrients (diet quality, calories, carbohydrates, protein, fiber, and cholesterol), micronutrients (Vitamin A, calcium, folate, iron, sodium, potassium, Vitamins B_2, B_3, B_6, C, and E), and phytonutrients (carotenoids). When compared to plasma carotenoids and skin carotenoid scores, controlling for BMI, carotenoid intake predicted by Diet ID™ was also correlated (Adjusted $R^2 = 0.37$, $p = 0.0001$; Adjusted $R^2 = 0.41$, $p < 0.0001$, respectively).

Following validation of the Veggie Meter® and Diet ID™, these studies served as the impetus to use these assessment tools to evaluate the impact of food access resources on diet-related biomarkers as the second aim. This observational cohort study consisted of university students (n = 132) with varying food security statuses. Pre- and post-measurements of skin carotenoids, plasma carotenoids, and dietary carotenoids were collected to determine changes with the use of food access resources over the academic term. Food access resources were utilized an average of 3.1 ± 2.6 times. Criterion-validity between devices found significant associations between measures of plasma carotenoids, skin carotenoids, and dietary carotenoids. Of the biomarkers of interest, skin carotenoids were significantly higher from pre to post, accounting for the interaction of food security status and the frequency of food access resource usage ($Adj\ R^2 = 0.31$; $p = 0.001$). Although this relationship was not observed for plasma carotenoids, or dietary carotenoids, this could be explained by the timing of resource usage. For dietary changes to be reflected in plasma, carotenoid-containing foods must have been consumed within approximately one week of post plasma collection. As only three participants out of the

132 utilized the resources during the final week of the study, changes in plasma carotenoids were not expected to reach significance.

After exploring the physiological impacts of FI on diet-related biomarkers and the potential intervention strategies for improving these health-related biomarkers, further investigation into the cognitive impacts were of interest. The final aim was to explore the interrelationship between executive function (EF), food security status, and diet quality. Participants (n = 230) completed the CANTAB assessment to serve as an objective measure of EF, validated assessments for stress, along with other conditions that may impact cognition, and Diet ID™ to assess diet quality and nutrient intake. There were significant differences in mean scores for various domains of EF by food security status, such that impulsivity, poor decision making, and reduced planning capabilities were more present in individuals experiencing FI compared to their food secure peers ($p < 0.05$).

The findings encompassed in this dissertation provides insight into the severity of physiological and cognitive outcomes associated with FI in the college student population. On-campus food access resources demonstrated promising effects on measurable health outcomes over the brief duration of a single academic term (< 10 weeks). These observed changes to diet and health-related biomarkers warrant the continued development of college and community-based resources aimed at alleviating the burden of food access, prioritizing foods that are nutrient-dense when possible. Future directions should seek to explore the longitudinal impacts of acute and chronic FI in the college student population to determine long-term ramifications to chronic disease risk.

CHAPTER 1

Methods for Assessing Health-Related Outcomes of Food Insecurity in College Students: A Narrative Review

Introduction

College students are disproportionately impacted by food insecurity (FI), with an estimated 40% of college students in the United States reporting a reduced quality or quantity diet, compared to the national average of 10.2%.[1,2] Emerging concerns regarding the inordinate prevalence of FI in college students has provided the impetus to explore the physiological, psychosocial, and cognitive impacts associated with acute and chronic FI in this population.[3,4]

Food insecurity, as defined by the United States Department of Agriculture (USDA), is the inability to maintain or acquire adequate food, which may result in disrupted eating patterns and decreased nutrient intake.[5] Food security status (FSS) is predominately evaluated using the validated USDA Food Security Survey Module (FSSM), which can screen for the risk of FI using a two-item screener or assess FSS at the individual or household level using a 2-item screener, or the 6, 10, or 18 question version.[6] Additional FI assessment methods, often adapted from the USDA FSSM to be utilized in specific populations, such as low- or middle-income countries, are utilized less frequently in the United States.[7,8] An additional emphasis has been placed on the inclusion of nutrition security, to address not only the underconsumption of essential nutrients, but the juxtaposition of overconsuming energy-dense, nutrient-poor foods, leading to an increased risk for nutrient deficiencies in conjunction with chronic disease development.[9]

To date, a majority of studies evaluating the impacts of FI on health-related outcomes in college students utilize subjective measures through self-report, due to the reduced researcher and participant burden; however, self-reported assessments of health outcomes are prone to systematic selection and information biases.[10,11] Additionally, inducing food insecurity for research purposes would be considered unethical, as food is considered a basic human right,

resulting in limited randomized controlled trials to elucidate the cause-and-effect relationship of FI on health outcomes.[12, 13]

Assessing the mechanisms involved in the development of negative health outcomes associated with FI is essential to establish sustainable and effective intervention strategies.[14] The reduction in either the quality or quantity of food in the diet may decrease the intake of macronutrients, vitamins, and minerals that are essential in regulating metabolic processes, subsequently increasing the risk of adverse physical or mental health outcomes.[15] Conversely, a reduced quality diet may also be associated with the increased consumption of ultra-processed and fast foods that contribute to increases in calories, total and saturated fats, added sugars, and sodium, ultimately contributing to weight gain, hypertension, and other chronic disease risk factors.[15,16] Ultra-processed and fast foods are sold in large quantities, often multiple servings in a single meal or package, at low-cost,[17] which is economically favorable for individuals experiencing FI.[18]

This narrative review of research published between January 2017 – March 2023 features an overview of the assessment tools used to evaluate the physical and mental health outcomes associated with the experience of FI in college students. Studies conducted in the United States using the USDA FSSM to determine FSS and validated measurement tools to assess health outcomes were included in this review (**Figure 1**). Findings from this review demonstrate the need for additional research using objective biomarkers to further explore the relationship between FI and health outcomes.

Food Insecurity and General Health

The majority of research studies exploring FI and physiological health outcomes in college students are cross-sectional study designs that implement a variety of questionnaires to assess

various components of health. Self-rated health status is frequently assessed using a Likert scale ranking of excellent, good, fair, or poor.[19-33] Compared to food secure peers, students experiencing FI had significantly poorer overall general health rankings (**Table 1**).[19-33]

Research validating subjective measures of self-reported health status with objective biomarkers of health have reported conflicting results, such that some studies have observed inconsistencies and attenuation biases, with respondents underreporting health or disease burden and others have found convergence between self-report and health outcomes.[34,35] To better understand the mechanisms causing health outcomes in the experience of FI, objective clinical and subclinical biomarkers are needed. The implementation of novel biomarkers may elucidate the role of FI in emerging adult populations on cardiac function,[36] systemic inflammation,[37] and body composition,[38] to promote early detection and treatment of FI-related chronic disease development.[39]

Food Insecurity and Dietary Intake

Changes in dietary intake is the first primary outcome observed in acute or chronic FI. The experience of FI may result in the physiological expression of undernutrition,[40] overnutrition,[41] or a combination of the two (**Table 1**).

Food Insecurity and Undernutrition

Food insecurity may be characterized by the reduction in total calorie intake, as well as specific nutrient deficiencies and inadequacies due to the underconsumption of diverse foods in the diet. The experience of food insecurity is negatively associated with structured daily mealtimes, such as skipping breakfast or consuming only one meal of day.[42] College students who experience food insecurity are more likely to report decreased intake of fruits and vegetables, which may result in inadequate fiber intake, as well as a multitude of vitamins and minerals.[19, 26, 42-45] In the

college student population, research exploring the relationship between FI and dietary intake has only consisted of subjective dietary and beverage assessment methods, such as Food Frequency Questionnaires (FFQ),[26] National Cancer Institute (NCI) Dietary Screener Questionnaire (DSQ),[42-45] Automated Self-Administered 24-hour (ASA24) Dietary Assessment Tool,[46] All Day Fruit and Vegetable Screener,[31] Youth Risk Behavior Surveillance System (YRBS),[47] renditions of a self-reported fruit and vegetable intake questionnaire,[19, 33, 48] and the Beverage Intake Questionnaire (BEV-Q).[31, 45] A recent study assessing dietary intake and FI in the college student population used the Nutrition Data System for Research (NDSR), which is currently referenced as the gold-standard for dietary intake data collection in free-living populations;[49, 50] dietary changes resulting from differences by FSS were not reported.[51]

Specific nutrients can be measured objectively to determine if an individual experiencing FI is nutrient deficient; however, such assessment methods are invasive and require plasma, serum, or tissue samples to extrapolate nutrient concentrations. These methods have been used to quantify nutrients of concern, such as folate, iron (ferritin and transferrin), copper, retinol, and zinc, in children and older adult populations;[52-53] objective measures of nutrient status have yet to be explored in college student populations. Innovative, non-invasive measures of nutrient status may be used to assess nutrient adequacy, including sensor-based technologies to detect sound and movement associated with eating patterns,[54] wearable image-based devices to capture dietary intake,[55] and spectroscopy-based measurements to measure nutrients in the skin and tissue,[56] among other emerging assessments.[51, 57, 58]

Novel objective measures of dietary intake also include relative changes to the microbiome. Exploration into the role of FI on gut microbiome composition has been a topic of interest in the college student population, as the microbiome directly influences nutritional

status.[59, 60] Although differences were observed in the abundance and diversity of microbiota and metabolites, the physiological implication of these microbial changes are unknown.[59] Previous challenges with obtaining fecal samples from college students may be mitigated by the use of improved collection and sequencing technologies, such as ingestible sampling devices or passive monitoring by smart toilets.[60, 61]

Food Insecurity and Overnutrition

Encompassed within the definition of the triple-burden of malnutrition, the experience of overnutrition may still be accompanied by undernutrition or micronutrient deficiencies, as high calorie intake may result in the underconsumption of essential vitamins and minerals.[62-64] Weight status, as it relates to nutritional adequacy is often overlooked, as it is presumed that being overweight or obese corresponds to overnutrition of all nutrients.[63] In the experience of FI, a strategy for satiation is to consume foods that are calorically dense, thus often exceeding nutrient recommendations for added sugars, fat, and sodium, leading to an increased risk for overweight and obesity.[19, 22, 25, 26, 30, 43, 44, 65] To assess the role of FI on body composition in college students, height and weight, along with abdominal and waist circumference, are either self-reported or collected by a trained researcher. Self-reported BMI is habitually underestimated by both biological sexes, although the underestimation of weight occurs to a greater magnitude in females than males; therefore, the impact of FI on weight status may be higher than reported.[66, 67] As college students are generally classified as young, healthy adults, independent of FSS, some research findings did not detect significant differences in BMI between individuals who were food secure (FS) compared to those who were FI.[20, 32, 33, 42, 68-70] It has also been observed that college students tend to not adhere to dietary or physical activity guidelines, leading to an increased risk of universal weight gain, regardless of socioeconomic status.[71, 72] To date, no

research on the influence of FI on body composition in college students have used dual-energy X-ray absorptiometry (DEXA), bioelectrical impedance, bone and total water density, or other more accurate and objective assessment tools.

The overconsumption of total energy and nutrient intake can be assessed through traditional dietary intake measurements, although the recorded stigmatization of the experience of FI for college students may result in additional reporting biases when asked to self-report eating behaviors.[73] Objective measures of overnutrition are generally more financially burdensome and require substantial more resources, such as time and geographic proximity, to collect participant body composition data. To better capture the experience of FI on overnutrition in college students, user-prompted technologies, such as phone camera adiposity measurements,[74] and wearable dietary monitors for assessing blood glucose levels[75] and antioxidant intake.[76] These technologies are historically underutilized in low-income and underrepresented communities and have yet to be implemented in populations experiencing FI.[77]

Food Insecurity and Psychosocial Health Outcomes

With the emergence of the field of nutritional psychology and nutritional psychiatry, the relationship between diet and mental health, as well as the mechanisms driving the psychological response, is advancing (**Table 2**).[78-79]

Food Insecurity and Psychological Distress

Psychological distress includes a multitude of mental health outcomes, such as depression and anxiety, and the associated feelings that can accompany these concerns.[80] In the experience of FI in college students, psychological distress was measured using the Diener Flourishing Scale, which measures various aspects of human functioning, such as positive relationships, life purpose, and feelings of competence.[81] Research using the Diener Flourishing Scale found that

college students with FI had significantly higher psychological distress than their FS counterparts.[27, 82, 83] Psychological well-being was also assessed in select studies through the Kessler-6 Scale[84] or the World Health Organization Five Factor Well-being Index (WHO-5),[85] in which individuals experiencing FI had lower overall psychological well-being than FS peers.[82, 83, 85-87] A more general assessment of mental health through the CDC Healthy Days Core Module found that college students with FI were at an increased risk for poor mental health status.[88-90]

The relationship between depression and FI in college students was primarily assessed using a version of the Patient Health Questionnaire (PHQ), which estimates the risk of depression in accordance with the criteria presented in the Diagnostic and Statistical Manual of Mental Disorders (DSM).[91] The experience of FI resulted in higher rates of depressive symptoms in every study that used the PHQ assessment.[23, 28, 89, 92-96] Other measures of depression and depressive symptoms have also been implemented in the research setting, such as the Depression Anxiety and Stress Scales – 21-item short form (DASS-21),[97] the Center for Epidemiologic Studies Depression Scale (CES-D),[33] the Beck Depression Inventory (BDI-II),[69] with only the BDI-II being the only assessment tool to not report significant differences in depression risk between FI and FS college students.

Similarly with depression, the relationship between anxiety and FI was predominately measured using an adapted version of the Generalized Anxiety Disorder (GAD) questionnaire.[89, 92] Other assessments included the DASS-21[97] and the Anxiety Sensitivity Index (ASI).[94] It was observed that symptoms of anxiety increased with the experience of FI in the college student population, independent of the measurement tool used.[89, 92, 94, 97] Additional psychological outcomes associated with depression and anxiety include feelings of loneliness,[82, 83, 86, 92, 96, 98]

hopelessness,[94] resiliency,[47, 83, 96] and suicidal behaviors,[83] all of which were measured by validated subjective assessments and were negatively impacted by the experience of FI (Table 2).

Reliance on subjective and self-reported measures of mental health is a common practice when assessing psychological outcomes. However, the development of active and passive data collection procedures may provide more consistent mental health monitoring that reduces recall bias.[99, 100] Gamified applications are emerging as a strategy for identifying college individuals experiencing mental health concerns and providing immediate services for support.[101] Research on the development of clinical and subclinical biomarkers for personalized mental health risk factors is on-going.[102]

Food Insecurity and Stress

Feelings of stress are not isolated to the experience of FI in the college student population. Stress in the college environment is ubiquitous, stemming from academic, financial, social, familial, or other life stressors.[103] The experience of stress may serve as a bidirectional mediator to increasing the etiologies of overnutrition with FI, as stress status may result in the consumption of highly palatable foods, which over time can cause excessive weight gain.[104]

Stress levels are commonly measured as a psychological health outcome of FI using the validated Cohen's Perceived Stress Scale (PSS)[42, 70, 96, 105] and the DASS-21.[97] Individual's experiencing FI consistently had higher self-reported stress levels compared to their food secure counterparts. The implementation of the Adverse Childhood Experiences (ACEs) Survey is used as a measure of stress and FI during childhood,[106] although this assessment only provides a glimpse into the severity and duration of stress across the lifespan.[107] Expectedly, the risk of experiencing FI during college years is significantly associated with ACE exposures.[108]

Few objective measures of stress are implemented in the clinical research setting. Due to the variability in diurnal fluctuations in plasma and salivary cortisol, the use of less volatile biomarkers may better discern the relationship between FI and stress.[109] The development of more stable measures of cortisol to be quantified through wearable heat conductance patches,[110] hair cortisol concentration,[111, 112] or telomere length[113] may provide a more in-depth understanding of how stress levels are impacted by the experience of FI in the college student population.

Food Insecurity and Sleep

Whether attributed to elevated stress and cortisol levels, or the underconsumption of nutrients involved in sleep regulation, such as protein (amino acid tryptophan), folic acid, zinc, or vitamin B-12, the experience of FI can influence quality and/or quantity of sleep.[114, 115] Independent of FSS, college students often experience reduced sleep duration and higher sleep disturbances due to increased academic and social responsibilities.[116] Under the experience of FI, these negative sleep outcomes are further exacerbated in college students.

Research on the association between FI and sleep outcomes in college students has been measured using the Pittsburg Sleep Quality Index (PSQI)[31, 70, 88, 117] and the Berlin Sleep Questionnaire.[118] The PSQI is a clinically validated 18-item assessment that estimates sleep quality and disturbances over a one month period,[119] whereas the Berlin Sleep Questionnaire measures is designed to measure obstructive sleep-related disorders, such as sleep apnea.[120] It was observed that the experience of FI resulted in a reduction in a multitude of sleep-related behaviors, including sleep duration, frequency of interruptions, sleep latency, and feelings of fatigue and tiredness,[70, 88, 117, 118] with the exception of one study that did not report significant differences in sleep by FSS.[31]

There are objective measures of sleep that have been measured in relationship to FI, although to date, these assessments have not been implemented in the college student population.[121] Actigraphy continuously measures an individual's movement and heart rate to derive circadian rhythmic parameters to estimate sleep/wake cycles.[122] Such actigraphy devices can be found in commercially available activity-based trackers[123, 124] and sleep rings.[124-126] Assessing the accuracy and validity of sleep tracking technology is on-going, as there are many internal and environmental factors that can affect sleep duration and quality, such as biological sex, body weight, pulse oxygen rates with sleep apnea, among others.[127] Although these discrepancies between sleep monitoring devices have been previously identified, objective measures of sleep may provide more reliable estimates of sleep-related outcomes than self-report.[128, 129]

Food Insecurity and Substance Abuse

In the experience of FI, it has been observed that individuals struggling with addiction may prioritize drug or alcohol intake over food.[130] Validated for use in college student populations, the Alcohol Use Disorders Identification Test (AUDIT) measures the risk for a range of alcohol-related behaviors.[131] Only one study within the last five years assessed alcohol abuse risk using the AUDIT tool and no significant differences were observed by FSS.[95] Additional studies prompt students to report alcohol and drug habits through questions such as "During the past 30 days, on how many days did you drink alcohol or use drugs".[93] Standardized assessments specifically for use in the college student setting include nine tools to determine alcohol-related problems.[132] Novel assessments, including digital communication strategies for identifying, diagnosing, and treating drug- and alcohol-related behaviors is on-going in the college student

population;[133] further, implementing the USDA FSSM would provide additional insight into the role of FI on drug and alcohol intake.

Food Insecurity and Disordered Eating Behaviors

Due to disrupted eating patterns under the experience of FI, the risk of disordered eating behaviors may subsequently increase.[134] In the college student population, disordered eating behaviors include binge eating, body dysmorphia, and concerns about weight.[135] Using the Eating Attitudes Test,[70] 5-item Sick, Control, One stone, Fat, Food Questionnaire (SCOFF-5),[135] the Eating Disorder Examination Questionnaire (EDE-Q),[136] and the Emotional Eating Survey (EES),[68] positive associations between FI and risk of disordered eating behaviors were observed in all assessments. The continued screening and detection of disordered eating behaviors in the college student population is warranted to explore the efficacy of pharmacological and community-based interventions aimed reducing the compounded risk of FI with disordered eating behaviors.[137, 138]

Food Insecurity and Cognitive Outcomes

Cognitive function refers to the ability to acquire, store, and process information through six domains: memory and learning, language, executive function, complex attention, social cognition, and perceptual and motor functions.[139] The experience of FI may affect one or more domains of cognitive function due to the impact of nutritional deficiencies, elevated stress levels, lack of sleep, or a combination of the various health outcomes previously discussed.[140] Inverse relationships between neural connectivity, cognitive function, and FI have been observed across the lifespan,[141, 142] with a considerable focus on the older adult population;[140, 143-145] however, there is limited research addressing this relationship in college students.[106]

Cognitive function can be assessed both objectively and subjectively. Objective measures used to determine the relationship between FI and cognitive outcomes include magnetic resonance imaging (MRI) to determine the anatomical structure of the brain and using functional magnetic resonance imaging (fMRI) to assess neural network activity.[106, 146] Cognitive function may also be determined using objective assessment tools, such as the Cambridge Neuropsychological Test Automated Battery (CANTAB);[147] however, this type of objective assessment battery has yet to be used to explore the impact of FI on executive function outcomes in the college student population. Subjective measures of cognition used in the college student population include the Behavior Rating Inventory of Executive Function-2 (BRIEF-2) assessment, a self-reported questionnaire that has participants rate the frequency of difficulty with tasks, the inability to problem solve, and other intrapersonal cognitive qualities.[106] In college students, individuals experiencing FI had higher BRIEF-2 scores, which reflects poorer cognitive function. Although the BRIEF-2 assessment is subjective and relies on participant perception, BRIEF-2 scores were correlated with the objective outcome of neural connectivity from fMRI output in various regions of the brain.[106]

Cognitive restraint with regard to food-making decisions has also been assessed as it relates to the FI in the college student population.[117] Using the Three Factor Eating Questionnaire (TFEQ-18), a self-reported questionnaire that calculates a score for emotional eating, uncontrolled eating, and cognitive restraint, it was observed that only uncontrolled eating scores were significantly associated with the experience of FI.[117] Further research into the role of FI on cognitive function is warranted as many college students are still experiencing brain maturation that impacts cognitive development up through the age of 24 years.[148] Concerns regarding the high prevalence of FI impacting cognitive processes in college students may also

be recognized through decreased academic performance [23, 27, 83, 94, 149-156] and other cognitively derived outcomes.

The use of objective measures of the domains of cognitive function are critical to further understand the driving mechanisms behind the experience of FI on neural development and cognitive function. Research findings have elucidated to the potential antioxidant role of carotenoids,[157] flavonoids,[158] and other phytochemicals[159] on cognitive health. As diet quality is impacted with the experience of FI, health-promoting phytonutrients may not be consumed as frequently and neuroinflammatory foods, such as saturated fats and added sugars, may be consumed more regularly, ultimately resulting in increased intestinal permeability, causing gut-brain dysbiosis to compromise the blood brain barrier.[160] Future research to explore the potential for cognitive flexibility and reversibility of cognitive decline with the experience of acute and/or chronic FI on brain development is needed.

Limitations

It is apparent from reviewing the recent literature that the field of FI research in college students consists predominately of subjective assessments for health outcomes. For this reason, only studies that used previously validated assessment tools were included, resulting in the omission of qualitative research studies that relied on participant verbal responses. The focus of this review on the college student population was selected due to the disproportionate rates of FI compared to the U.S. household prevalence; however, research in other populations, such as children or older adults, may yield additional assessment tools not yet implemented in the college setting.

Conclusions

This narrative review explores the recent literature on assessment methods used to determine physiological, psychosocial, and cognitive outcomes that are impacted by the experience of FI in college students. Findings from this review highlight the myriad of negative health outcomes associated with the experience of FI in college students. The dearth of clinical and subclinical biomarkers makes it challenging to assess other determinants of FI, such as the severity of acute and chronic FI on physical and mental well-being.[161] It is essential to identify the underlying mechanisms and pathways between FI and health outcomes to develop targeted interventions aimed at improving FSS in the college student population.

Future Directions

The disproportionate rates of FI in the college student population, along with the implicated health outcomes, are imperative to address. Future directions are twofold: the incorporation of objective biomarkers to address the clinical research gaps and the development of effective interventions at the public health and policy level to improve FSS. The incorporation of food, nutrition, and financial literacy programs into the college setting have served as effective interventions to reduce health disparities from the experience of FI.[162-163] Geographic Information Systems (GIS) may also be a novel approach to geocoding dietary behaviors by socioeconomic status to better identify at risk college student populations.[164] As FI is an interdisciplinary and multifaceted public health concern, effective interventions to alleviate the health impacts require collaboration in areas of nutrition, medicine, agriculture, sustainability, and technology, among many others.

This manuscript was submitted for publication and is currently under review:

Radtke MD, Steinberg FM, Scherr RS. Methods for Assessing Health-Related Outcomes of Food Insecurity in College Students: A Narrative Review. Under review at *Advances for Nutrition* as of April 5, 2023.

References

1. Nikolaus CJ, An R, Ellison B, Nickols-Richardson SM. *Food Insecurity among College Students in the United States: A Scoping Review.* Adv Nutr 2020;11(2):327-348. doi:10.1093/advances/nmz111
2. United States Department of Agriculture Economic Research Service. Key Statistics & Graphics - Food Security Status of U.S. Households in 2021 [Accessed March 25, 2023]. Available at https://www.ers.usda.gov/topics/food-nutrition-assistance/food-security-in-the-u-s/key-statistics-graphics/#foodsecure
3. Zenk SN, Tabak TA, Perez-Stable EJ. Research Opportunities to Address Nutrition Insecurity and Disparities. JAMA 2022;327(20):1953-1954. doi:10.1001/jama.2022.7159
4. Butrum B, Mitchum DM, Hunter FA, Littlejohn L, Young M, Rutberg D, Wilson S, Jarosik T, Grant P, Wertz R. Notice of Special Interest (NOSI): Stimulating Research to Understand and Address Hunger, Food and Nutrition Insecurity [Accessed on March 25, 2023]. Available at https://grants.nih.gov/grants/guide/notice-files/not-od-22-135.html.
5. United States Department of Agriculture. Definitions of Food Security 2021 [Accessed March 25, 2023]. Available at https://www.ers.usda.gov/topics/food-nutrition-assistance/food-security-in-the-us/definitions-of-food-security/.
6. United States Department of Agriculture Economic Research Service. Food security in the U.S.: measurement [Accessed on March 25, 2023]. Available at https://www.ers.usda.gov/topics/food-nutrition-assistance/food-security-in-the-u-s/measurement/#measurement.
7. Keenan DP, Olson C, Hersey JC, Parmer SM. Measures of food insecurity/security. J Nutr Educ 2001;33(1):S49-58. doi: 10.1016/s1499-4046(06)60069-9

8. Jones AD, Ngure FM, Pelto G, Young SL. What are we assessing when we measure food security? A compendium and review of current metrics. Adv Nutr 2013;4(5):481-505. doi:10.3945/an.113.004119

9. Ingram J. Nutrition security is more than food security. Nature Food 2020;1(1):2. doi:10.1038/s43016-019-0002-4

10. Kuhnle GGC. Nutritional biomarkers for objective dietary assessment. J Sci Food Agric 2012;92(6):1145-1149. doi:10.1002/jsfa.5631

11. Althubaiti A. Information bias in health research: definition, pitfalls, and adjustment methods. J Multidiscip Healthc 2016;9:211-217. doi:10.2147/JMDH.S104807

12. Murrell AJ, Jones R, Rose S, Firestine A, Bute J. Food Security as Ethics and Social Responsibility: An Application of the Food Abundance Index in an Urban Setting. Int J Environ Res Public Health 2022;19(16):10042. doi:10.3390/ijerph191610042

13. United Nations Human Rights Office of the High Commisioner. About the Universal Declaration of Human Rights Translation Project [Accessed March 25, 2023]. Available at https://www.ohchr.org/en/human-rights/universal-declaration/universal-declaration-human-rights/about-universal-declaration-human-rights-translation-project.

14. Loopstra R. Interventions to address household food insecurity in high-income countries. Proc Nutr Soc 2018;77(3):270-281. doi:10.1017/S002966511800006X

15. Gundersen C, Ziliak JP. Food Insecurity And Health Outcomes. Health Aff (Millwood) 2015;34(11):1830-1839. doi:10.1377/hlthaff.2015.0645

16. Leung CW, Fulay AP, Parnarouskis L, Martinez-Steele E, Gearhardt AN, Wolfson JA. Food insecurity and ultra-processed food consumption: the modifying role of

participation in the Supplemental Nutrition Assistance Program (SNAP). Am J Clin Nutr 2022;116(1):197-205. doi:10.1093/ajcn/nqac049

17. Young LR, Nestle M. Portion Sizes of Ultra-Processed Foods in the United States, 2002 to 2021. Am J Public Health 2021;111(12):2223-2226. doi:10.2105/AJPH.2021.306513

18. Lee K, Hyun J, Lee Y. Why do and why Don't people consume fast Food?: An application of the consumption value model. Food Quality and Preference 2022;99:104550. doi:10.1016/j.foodqual.2022.104550

19. Martinez SM, Grandner MA, Nazmi A, Ruben Caedo E, Ritchie LD. Pathways from Food Insecurity to Health Outcomes among California University Students. Nutrients 2019;11(6):1419. doi:10.3390/nu11061419

20. Knol LL, Robb CA, McKinley EM, Wood M. Food Insecurity, Self-rated Health, and Obesity among College Students. American Journal of Health Education 2017;48(4):248-255. doi:10.1080/19325037.2017.1316689

21. McArthur LH, Ball L, Danek AC, Holbert D. A High Prevalence of Food Insecurity Among University Students in Appalachia Reflects a Need for Educational Interventions and Policy Advocacy. J Nutr Educ Behav 2018;50(6):564-572. doi:10.1016/j.jneb.2017.10.011

22. McArthur LH, Fasczewski KS, Wartinger E, Miller J. Freshmen at a University in Appalachia Experience a Higher Rate of Campus than Family Food Insecurity. J Community Health 2018;43(5):969-976. doi:10.1007/s10900-018-0513-1

23. Payne-Sturges DC, Tjaden A, Caldeira KM, Vincent KB, Arria AM. Student Hunger on Campus: Food Insecurity Among College Students and Implications for Academic Institutions. Am J Health Promot 2018;32(2):349-354. doi:10.1177/0890117117719620

24. Soldavini J, Berner M. The importance of precision: differences in characteristics associated with levels of food security among college students. Public Health Nutr 2020;23(9):1473-1483. doi:10.1017/S1368980019004026

25. Soldavini J, Berner M, Da Silva J. Rates of and characteristics associated with food insecurity differ among undergraduate and graduate students at a large public university in the Southeast United States. Prev Med Rep 2019;14:100836. doi:10.1016/j.pmedr.2019.100836

26. Davitt ED, Heer MM, Winham DM, Knoblauch ST, Shelley MC. Effects of COVID-19 on University Student Food Security. Nutrients 2021;13(6):1932. doi:10.3390/nu13061932

27. Ahmed T, Ilieva R, Shane J, Reader S, Aleong C, Wong HY, et al. A Developing Crisis in Hunger: Food Insecurity within 3 Public Colleges before and during the COVID-19 Pandemic. Journal of Hunger & Environmental Nutrition 2022;18(1):1-20. doi:10.1080/19320248.2022.2026853

28. Leung CW, Farooqui S, Wolfson JA, Cohen AJ. Understanding the Cumulative Burden of Basic Needs Insecurities: Associations With Health and Academic Achievement Among College Students. Am J Health Promot 2021;35(2):275-278. doi:10.1177/0890117120946210

29. Hiller MB, Winham DM, Knoblauch ST, Shelley MC. Food Security Characteristics Vary for Undergraduate and Graduate Students at a Midwest University. Int J Environ Res Public Health 2021;18(11):5730. doi:10.3390/ijerph18115730

30. Olfert MD, Hagedorn-Hatfield RL, Houghtaling B, Esquivel MK, Hood LB, MacNell L, et al. Struggling with the basics: food and housing insecurity among college students

across twenty-two colleges and universities. J Am Coll Health 2021:1-12. doi:10.1080/07448481.2021.1978456

31. Ryan RA, Murphy B, Deierlein AL, Lal S, Parekh N, Bihuniak JD. Food Insecurity, Associated Health Behaviors, and Academic Performance Among Urban University Undergraduate Students. J Nutr Educ Behav 2022;54(3):269-275. doi:10.1016/j.jneb.2021.08.008

32. Soldavini J, Andrew H, Berner M. Campus-Based Food Insecurity: The Case of International Students at a Southeastern University. Journal of Student Affairs Research and Practice 2022;59(3):338-351. doi:10.1080/19496591.2021.1997755

33. Willis DE. Feeding inequality: food insecurity, social status and college student health. Sociol Health Illn 2021;43(1):220-237. doi:10.1111/1467-9566.13212

34. Onur I, Velamuri M. The gap between self-reported and objective measures of disease status in India. PLoS One 2018;13(8):e0202786. doi:10.1371/journal.pone.0202786

35. Krueger AB, Stone AA. Psychology and economics. Progress in measuring subjective well-being. Science 2014;346(6205):42-3. doi: 10.1126/science.1256392

36. Shemisa K, Bhatt A, Cheeran D, Neeland IJ. Novel Biomarkers of Subclinical Cardiac Dysfunction in the General Population. Curr Heart Fail Rep 2017;14(4):301-310. doi:10.1007/s11897-017-0342-z

37. Valacchi G, Virgili F, Cervellati C, Pecorelli A. OxInflammation: From Subclinical Condition to Pathological Biomarker. Front Physiol 2018;9:858. doi:10.3389/fphys.2018.00858

38. Aleksandrova K, Rodrigues CE, Floegel A, Ahrens W. Omics Biomarkers in Obesity: Novel Etiological Insights and Targets for Precision Prevention. Curr Obes Rep 2020;9(3):219-230. doi:10.1007/s13679-020-00393-y

39. Seligman HK, Laraia BA, Kushel MB. Food insecurity is associated with chronic disease among low-income NHANES participants. J Nutr 2010;140(2):304-10. doi:10.3945/jn.109.112573

40. Moradi S, Mirzababaei A, Mohammadi H, Moosavian SP, Arab A, Jannat B, Mirzaei K. Food insecurity and the risk of undernutrition complications among children and adolescents: A systematic review and meta-analysis. Nutrition 2019;62:52-60. doi:10.1016/j.nut.2018.11.029

41. Franklin B, Jones A, Love D, Puckett S, Macklin J, White-Means S. Exploring mediators of food insecurity and obesity: a review of recent literature. J Community Health 2012;37(1):253-64. doi:10.1007/s10900-011-9420-4

42. Bruening M, van Woerden I, Todd M, Laska MN. Hungry to learn: the prevalence and effects of food insecurity on health behaviors and outcomes over time among a diverse sample of university freshmen. Int J Behav Nutr Phys Act 2018;15(1):9. doi:10.1186/s12966-018-0647-7

43. El Zein A, Colby SE, Zhou W, Shelnutt KP, Greene GW, Horacek TM, Olfert MD, Matthews AE. Food Insecurity Is Associated with Increased Risk of Obesity in US College Students. Curr Dev Nutr 2020;4(8):nzaa120. doi:10.1093/cdn/nzaa120

44. Leung CW, Wolfson JA, Lahne J, Barry MR, Kasper N, Cohen AJ. Associations between Food Security Status and Diet-Related Outcomes among Students at a Large, Public

Midwestern University. J Acad Nutr Diet 2019;119(10):1623-1631. doi:10.1016/j.jand.2019.06.251

45. Mei J, Fulay AP, Wolfson JA, Leung CW. Food Insecurity and Dietary Intake among College Students with Unlimited Meal Plans at a Large, Midwestern University. J Acad Nutr Diet 2021;121(11):2267-2274. doi:10.1016/j.jand.2021.04.009

46. Silva FB, Osborn DE, Owens MR, Kirkland T, Moore CE, Patterson MA, et al. Influence of COVID-19 Pandemic Restrictions on College Students' Dietary Quality and Experience of the Food Environment. Nutrients 2021;13(8):2790. doi:10.3390/nu13082790

47. Laska MN, Lenk K, Lust K, McGuire CM, Porta CM, Stebleton M. Sociodemographic and health disparities among students screening positive for food insecurity: Findings from a large college health surveillance system. Prev Med Rep 2021;21:101297. doi:10.1016/j.pmedr.2020.101297

48. Sackey JD, Pike K, Rothpletz-Puglia P, Brody R, Touger-Decker R. Food Insecurity Among Health Sciences Graduate Students at a Large Northeastern University. J Nutr Educ Behav 2021;53(5):428-433. doi:10.1016/j.jneb.2020.11.003

49. Steinfeldt L, Anand J, Murayi T. Food Reporting Patterns in the USDA Automated Multiple-Pass Method. Procedia Food Science 2013;2:145-156. doi:10.1016/j.profoo.2013.04.022

50. Conway JM, Ingwersen LA, Moshfegh AJ. Accuracy of dietary recall using the USDA five-step multiple-pass method in men: an observational validation study. J Am Diet Assoc 2004;104(4):595-603. doi:10.1016/j.jada.2004.01.007

51. Radtke MD, Chodur GC, Bissell MCS, Kemp LC, Medici V, Steinberg FM, Scherr RE. Validation of Diet ID™ in Predicting Nutrient Intake Compared to Dietary Recalls, Skin Carotenoid Scores, and Plasma Carotenoids in University Students. Nutrients 2023;15(2):409. doi: 10.3390/nu15020409

52. Jun S, Cowan AE, Dodd KW, Tooze JA, Gahche JJ, Eicher-Miller HA, et al. Association of food insecurity with dietary intakes and nutritional biomarkers among US children, National Health and Nutrition Examination Survey (NHANES) 2011-2016. Am J Clin Nutr 2021;114(3):1059-1069. doi:10.1093/ajcn/nqab113

53. Eicher-Miller HA, Mason AC, Weaver CM, McCabe GP, Boushey CJ. Food insecurity is associated with iron deficiency anemia in US adolescents. Am J Clin Nutr 2009;90(5):1358-1371. doi:10.3945/ajcn.2009.27886

54. Rantala E, Balatsas-Lekkas A, Sozer N, Pennanen K. Overview of objective measurement technologies for nutrition research, food-related consumer and marketing research. Trends in Food Science & Technology 2022;125:100-113. doi:10.1016/j.tifs.2022.05.006

55. Jobarteh ML, McCrory MA, Lo B, Sun M, Sazonov E, Anderson AK, et al. Development and Validation of an Objective, Passive Dietary Assessment Method for Estimating Food and Nutrient Intake in Households in Low- and Middle-Income Countries: A Study Protocol. Curr Dev Nutr 2020;4(2):nzaa020. doi:10.1093/cdn/nzaa020

56. Radtke MD, Jilcott Pitts S, Jahns L, Firnhaber GC, Loofbourrow BM, Zeng A, Scherr RE. Criterion-Related Validity of Spectroscopy-Based Skin Carotenoid Measurements as a Proxy for Fruit and Vegetable Intake: A Systematic Review. Adv Nutr 2020;11(5):1282-1299. doi:10.1093/advances/nmaa054

57. Kong NA, Moy FM, Ong SH, Tahir GA, Loo CK. MyDietCam: Development and usability study of a food recognition integrated dietary monitoring smartphone application. Digit Health 2023;9:20552076221149320. doi:10.1177/20552076221149320

58. Mortazavi BJ, Gutierrez-Osuna R. A Review of Digital Innovations for Diet Monitoring and Precision Nutrition. J Diabetes Sci Technol 2023;17(1):217-223. doi:10.1177/19322968211041356

59. Mohr AE, Jasbi P, Vander Wyst KB, van Woerden I, Shi X, Gu H, Whisner CM, Bruening M. Association of food insecurity on gut microbiome and metabolome profiles in a diverse college-based sample. Sci Rep 2022;12(1):14358. doi:10.1038/s41598-022-18515-y

60. Tang Q, Jin G, Wang G, Liu T, Liu X, Wang B, Cao H. Current Sampling Methods for Gut Microbiota: A Call for More Precise Devices. Front Cell Infect Microbiol 2020;10:151. doi:10.3389/fcimb.2020.00151

61. Ge TJ, Rahimzadeh VN, Mintz K, Park WG, Martinez-Martin N, Liao JC, Park SM. Passive monitoring by smart toilets for precision health. Sci Transl Med 2023;15(681):eabk3489. doi:10.1126/scitranslmed.abk3489

62. Tan PY, Moore JB, Bai L, Tang G, Gong YY. In the context of the triple burden of malnutrition: A systematic review of gene-diet interactions and nutritional status. Crit Rev Food Sci Nutr 2022:1-29. doi:10.1080/10408398.2022.2131727

63. Astrup A, Bugel S. Overfed but undernourished: recognizing nutritional inadequacies/deficiencies in patients with overweight or obesity. Int J Obes (Lond) 2019;43(2):219-232. doi:10.1038/s41366-018-0143-9

64. Darmon N, Ferguson EL, Briend A. A cost constraint alone has adverse effects on food selection and nutrient density: an analysis of human diets by linear programming. J Nutr 2002;132(12):3764-3771. doi:10.1093/jn/132.12.3764

65. Huelskamp A, Waity J, Russell J. Effects of campus food insecurity on obesogenic behaviors in college students. J Am Coll Health 2021;69(5):572-575. doi:10.1080/07448481.2019.1684298

66. Gosse MA. How accurate is self-reported BMI? Nutrition Bulletin 2014;39(1):105-114. doi:10.1111/nbu.12075

67. Lyons AA, Park J, Nelson CH. Food insecurity and obesity: a comparison of self-reported and measured height and weight. Am J Public Health 2008;98(4):751-757. doi:10.2105/AJPH.2006.093211

68. Frank ML, Sprada GB, Hultstand KV, West CE, Livingston JA, Sato AF. Toward a deeper understanding of food insecurity among college students: examining associations with emotional eating and biological sex. J Am Coll Health 2021:1-9. doi:10.1080/07448481.2021.1936536

69. Umeda M, Ullevig S, Ching E, Kim Y, Escobedo TJ, Zeitz CJ. Depression Mediates the Relationship between Food Insecurity and Pain Interference in College Students. Int J Environ Res Public Health 2020;18(1):78. doi:10.3390/ijerph18010078

70. El Zein A, Shelnutt KP, Colby S, Vilaro MJ, Zhou W, Greene G, et al. Prevalence and correlates of food insecurity among U.S. college students: a multi-institutional study. BMC Public Health 2019;19(1):660. Doi:10.1186/s12889-019-6943-6

71. Bailey CP, Sharma S, Economos CD, Hennessy E, Simon S, Hatfield DP. College campuses' influence on student weight and related behaviours: A review of observational and intervention research. Obes Sci Pract 2020;6(6):694-707. doi:10.1002/osp4.445

72. United States Department of Agriculture and the United States Department of Health and Human Services. 2020 - 2025 Dietary Guidelines for Americans. 9th Edition. December 2020 [Accessed March 25, 2023]. Available at https://www.dietaryguidelines.gov/sites/default/files/2020-12/Dietary_Guidelines_for_Americans_2020-2025.pdf

73. Meza A, Altman E, Martinez S, Leung CW. "It's a Feeling That One Is Not Worth Food": A Qualitative Study Exploring the Psychosocial Experience and Academic Consequences of Food Insecurity Among College Students. J Acad Nutr Diet 2019;119(10):1713-1721 e1. doi:10.1016/j.jand.2018.09.006

74. Majmudar MD, Chandra S, Yakkala K, Kennedy S, Agrawal A, Sippel M, et al. Smartphone camera based assessment of adiposity: a validation study. NPJ Digit Med 2022;5(1):79. doi:10.1038/s41746-022-00628-3

75. Kumar Das S, Nayak KK, Krishnaswamy PR, Kumar V, Bhat N. Review—Electrochemistry and Other Emerging Technologies for Continuous Glucose Monitoring Devices. ECS Sensors Plus 2022;1(3). doi:10.1149/2754-2726/ac7abb

76. Xu Z, Liu G, Wu J. A novel hydrogel with glucose-responsive hyperglycemia regulation and antioxidant activity for enhanced diabetic wound repair. Nano Research 2022;15(6):5305-5315. doi:10.1007/s12274-022-4192-y

77. Huhn S, Axt M, Gunga HC, Maggioni MA, Munga S, Obor D, et al. The Impact of Wearable Technologies in Health Research: Scoping Review. JMIR Mhealth Uhealth, 2022;10(1):e34384. doi:10.2196/34384

78. Adan RAH, van der Beek EM, Buitelaar JK, Cryan JF, Hebebrand J, Higgs S, Schellekens H, Dickson SL. Nutritional psychiatry: Towards improving mental health by what you eat. Eur Neuropsychopharmacol 2019;29(12):1321-1332. doi:10.1016/j.euroneuro.2019.10.011

79. Godos J, Currenti W, Angelino D, Mena P, Castellano S, Caraci F, Galvano F, Del Rio D, Ferri R, Grosso G. Diet and Mental Health: Review of the Recent Updates on Molecular Mechanisms. Antioxidants (Basel) 2020;9(4):346. doi:10.3390/antiox9040346

80. Myers CA. Food Insecurity and Psychological Distress: a Review of the Recent Literature. Curr Nutr Rep 2020;9(2):107-118. doi:10.1007/s13668-020-00309-1

81. Diener E, Wirtz D, Tov W, Kim-Prieto C, Choi DW, Oishi S, Biswas-Diener R. New Well-being Measures: Short Scales to Assess Flourishing and Positive and Negative Feelings. Social Indicators Research 2009;97(2):143-156. doi:10.1007/s11205-009-9493-y

82. Marmolejo C, Banta JE, Siapco G, Djara MB. Examining the association of student mental health and food security with college GPA. J Am Coll Health 2022:1-7. doi:10.1080/07448481.2022.2058327

83. DeBate R, Himmelgreen D, Gupton J, Heuer JN. Food Insecurity, Well-being, and Academic Success among College Students: Implications for Post COVID-19 Pandemic Programming. Ecol Food Nutr 2021;60(5):564-579. doi:10.1080/03670244.2021.1954511

84. Sunderland M, Hobbs MJ, Anderson TM, Andrews G. Psychological distress across the lifespan: examining age-related item bias in the Kessler 6 Psychological Distress Scale. Int Psychogeriatr 2012;24(2):231-42. doi:10.1017/S1041610211001852

85. Haskett ME, Majumder S, Kotter-Grühn D, Gutierrez I. The role of university students' wellness in links between homelessness, food insecurity, and academic success. Journal of Social Distress and Homelessness 2020;30(1):59-65. doi:10.1080/10530789.2020.1733815

86. Guzman PG, Lange JE, McClain AC. The Association between Food Security Status and Psychological Distress and Loneliness among Full-Time Undergraduate Students at a Minority-Serving Institution. Int J Environ Res Public Health 2022;19(22):15245. doi:10.3390/ijerph192215245

87. Becerra MB, Becerra BJ. Psychological Distress among College Students: Role of Food Insecurity and Other Social Determinants of Mental Health. Int J Environ Res Public Health 2020;17(11):4118. doi:10.3390/ijerph17114118

88. Hagedorn RL, Olfert MD, MacNell L, Houghtaling B, Hood LB, Savoie Roskos MR, Goetz JR, et al. College student sleep quality and mental and physical health are associated with food insecurity in a multi-campus study. Public Health Nutr 2021;24(13):4305-4312. doi:10.1017/S1368980021001191

89. Coakley KE, Cargas S, Walsh-Dilley M, Mechler H. Basic Needs Insecurities Are Associated with Anxiety, Depression, and Poor Health Among University Students in the State of New Mexico. J Community Health 2022;47(3):454-463. doi:10.1007/s10900-022-01073-9

90. Wattick RA, Hagedorn RL, Olfert MD. Relationship between Diet and Mental Health in a Young Adult Appalachian College Population. Nutrients 2018;10(8):957. doi:10.3390/nu10080957

91. Kroenke K, Spitzer RL, Williams JB. The PHQ-9: validity of a brief depression severity measure. J Gen Intern Med 2001;16(9):606-13. doi:10.1046/j.1525-1497.2001.016009606.x

92. Oh H, Snith L, Jacob L, Du J, Shin J, Zhou S, Koyanagi A. Food insecurity and mental health among young adult college students in the United States. J Affect Disord 2022;303:359-363. doi:10.1016/j.jad.2022.02.009

93. Neal L, Zigmont VA. Undergraduate food insecurity, mental health, and substance use behaviors. Nutr Health 2022:2601060221142669. doi:10.1177/02601060221142669

94. Raskind IG, Haardorfer R, Berg CJ. Food insecurity, psychosocial health and academic performance among college and university students in Georgia, USA. Public Health Nutr 2019;22(3):476-485. doi:10.1017/S1368980018003439

95. Reeder N, Tapanee P, Persell A, Tolar-Paterson T. Food Insecurity, Depression, and Race: Correlations Observed Among College Students at a University in the Southeastern United States. Int J Environ Res Public Health 2020;17(21):8268. doi:10.3390/ijerph17218268

96. Diamond KK, Stebleton MJ, delMas RC. Exploring the Relationship Between Food Insecurity and Mental Health in an Undergraduate Student Population. Journal of Student Affairs Research and Practice 2019;57(5):546-560. doi:10.1080/19496591.2019.1679158

97. Coffino JA, Spoor SP, Drach RD, Hormes JM. Food insecurity among graduate students: prevalence and association with depression, anxiety and stress. Public Health Nutr 2021;24(7):1889-1894. doi:10.1017/S1368980020002001

98. Cockerham M, Camel S, James L, Neill D. *Food insecurity in baccalaureate nursing students: A cross-sectional survey.* J Prof Nurs 2021;37(2):249-254. doi:10.1016/j.profnurs.2020.12.015

99. Boukhechba M, Caros AR, Fua K, Chow P, Teachman B, Barnes L. DemonicSalmon: Monitoring mental health and social interactions of college students using smartphones. Smart Health 2018;9(10):192-203. Doi:10.1016/j.smhl.2018.07.005

100. Ng MM, Firth J, Minen M, Torous J. User Engagement in Mental Health Apps: A Review of Measurement, Reporting, and Validity. Psychiatr Serv 2019;70(7):538-544. doi:10.1176/appi.ps.201800519

101. Nicolaidou I, Aristeidis L, Lambrinos L. A gamified app for supporting undergraduate students' mental health: A feasibility and usability study. Digit Health 2022;8:20552076221109059. doi:10.1177/20552076221109059

102. Hickie IB, Scott EM, Cross SP, Iorfino F, Davenport TA, Guastella AJ, et al. Right care, first time: a highly personalised and measurement-based care model to manage youth mental health. Med J Aust 2019;211(9):S3-S46. doi:10.5694/mja2.50383

103. Yikealo D, Yemane B, Karvinen I. The Level of Academic and Environmental Stress among College Students: A Case in the College of Education. Open Journal of Social Sciences 2018;6(11):40-57. doi:10.4236/jss.2018.611004

104. Sominsky L, Spencer SJ. Eating behavior and stress: a pathway to obesity. Front Psychol 2014;5:434. doi:10.3389/fpsyg.2014.00434

105. Richards AL, Specker B. Evaluating hours of sleep and perceived stress on dietary cognitive restraint in a survey of college students. J Am Coll Health 2019;68(8):824-831. doi:10.1080/07448481.2019.1618312

106. Guerithault N, McClure SM, Ojinnaka CO, Blair Braden B, Bruening M. Resting-State Functional Connectivity Differences in College Students with and without Food Insecurity. Nutrients 2022;14(10):2064. doi:10.3390/nu14102064

107. Crosswell AD, Lockwood KG. Best practices for stress measurement: How to measure psychological stress in health research. Health Psychol Open 2020;7(2):2055102920933072. doi:10.1177/2055102920933072

108. Testa A, Jackson DB. Adverse Childhood Experiences and Food Insecurity in Adulthood: Evidence From the National Longitudinal Study of Adolescent to Adult Health. J Adolesc Health 2020;67(2):218-224. doi:10.1016/j.jadohealth.2020.02.002

109. Adam EK, Quinn ME, Tavernier R, McQuillan MT, Dahlke KA, Gilbert KE. Diurnal cortisol slopes and mental and physical health outcomes: A systematic review and meta-analysis. Psychoneuroendocrinology 2017;83:25-41. doi:10.1016/j.psyneuen.2017.05.018

110. Nyein HYY, Bariya M, Tran B, Ahn CH, Brown BJ, Ji W, Davis N, Javey A. A wearable patch for continuous analysis of thermoregulatory sweat at rest. Nat Commun 2021;12(1):1823. doi:10.1038/s41467-021-22109-z

111. Li Y, Jia W, Yan N, Hua Y, Han T, Yang J, Ma L, Ma L. Associations between chronic stress and hair cortisol in children: A systematic review and meta-analysis. J Affect Disord 2023;329:438-447. doi:10.1016/j.jad.2023.02.123

112. Ling J, Robbins LB, Xu D. Food Security Status and Hair Cortisol among Low-income Mother-Child Dyads. West J Nurs Res 2019;41(12):1813-1828. doi:10.1177/0193945919867112

113. Montoya M, Uchino BN. Social support and telomere length: a meta-analysis. J Behav Med 2023. doi:10.1007/s10865-022-00389-0

114. Ding M, Keiley MK, Garza KB, Duffy PA, Zizza CA. Food insecurity is associated with poor sleep outcomes among US adults. J Nutr 2015;145(3):615-21. doi:10.3945/jn.114.199919

115. Binks H, Vincent GE, Gupta C, Irwin C, Khalesi S. Effects of Diet on Sleep: A Narrative Review. Nutrients 2020;12(4):936. doi:10.3390/nu12040936

116. Wang F, Biro E. Determinants of sleep quality in college students: A literature review. Explore (NY) 2021;17(2):170-177. doi:10.1016/j.explore.2020.11.003

117. Richards AL, Specker B. Evaluating hours of sleep and perceived stress on dietary cognitive restraint in a survey of college students. J Am Coll Health 2020;68(8):824-831. doi:10.1080/07448481.2019.1618312

118. Becerra MB, Bol BS, Granados R, Hassija C. Sleepless in school: The role of social determinants of sleep health among college students. J Am Coll Health 2020;68(2):185-191. doi:10.1080/07448481.2018.1538148

119. Buysse DJ, Reynolds III CF, Monk TH, Berman SR< Kupfer DJ. The Pittsburgh Sleep Quality Index: a new instrument for psychiatric practice and research. Psychiatry Res 1989;28(2):193-213. doi:10.1016/0165-1781(89)90047-4

120. Senaratna CV, Perret JL, Matheson MC, Lodge CJ, Lowe A, Cassim R, et al. Validity of the Berlin questionnaire in detecting obstructive sleep apnea: A systematic review and meta-analysis. Sleep Med Rev 2017;36:116-124. doi:10.1016/j.smrv.2017.04.001

121. Troxel WM, Haas A, Ghosh-Dastidar B, Richardson AS, Hale L, Buysse DJ, Buman MP, Kurka J, Dubowitz T. Food Insecurity is Associated with Objectively Measured Sleep Problems. Behav Sleep Med 2020;18(6):719-729. doi:10.1080/15402002.2019.1669605

122. Ancoli-Israel S, Cole R, Alessi C, Chambers M, Moorcroft W, Pollak CP. The Role of Antigraphy in the Study of Sleep and Circadian Rhythms American Academy of Sleep Medicine 2003;23(3):342-392. doi:10.1093/sleep/26.3.342

123. Weiss AR, Johnson NL, Berger NA, RedlineS. Validity of Activity-Based Devices to Estimate Sleep. Journal of Clinical Sleep Medicine 2010;6(4):336-342.

124. Asgari Mehrabadi M, Azimi I, Sarhaddi F, Axelin A, Niela-Vilén H, Myllyntausta S, et al. Sleep Tracking of a Commercially Available Smart Ring and Smartwatch Against Medical-Grade Actigraphy in Everyday Settings: Instrument Validation Study. JMIR Mhealth Uhealth 2020;8(10):e20465. doi:10.2196/20465

125. de Zambotti M, Rosas L, Colrain IM, Baker FC. The Sleep of the Ring: Comparison of the OURA Sleep Tracker Against Polysomnography. Behav Sleep Med 2019;17(2):124-136. doi:10.1080/15402002.2017.1300587

126. Altini M, Kinnunen H. The Promise of Sleep: A Multi-Sensor Approach for Accurate Sleep Stage Detection Using the Oura Ring. Sensors (Basel) 2021;21(13):4302. doi:10.3390/s21134302

127. Danzig R, Wang M, Shah A, Trotti LM. The wrist is not the brain: Estimation of sleep by clinical and consumer wearable actigraphy devices is impacted by multiple patient- and device-specific factors. J Sleep Res 2020;29(1):e12926. doi:10.1111/jsr.12926

128. Bianchi MT, Williams KL, McKinney S, Ellenbogen JM. The subjective-objective mismatch in sleep perception among those with insomnia and sleep apnea. J Sleep Res 2013;22(5):557-568. doi:10.1111/jsr.12046

129. O'Donnell D, Silva EJ, Münch M, Ronda JM, Wang W, Duffy JF. Comparison of subjective and objective assessments of sleep in healthy older subjects without sleep complaints. J Sleep Res 2009;18(2):254-63. doi:10.1111/j.1365-2869.2008.00719.x

130. Nagata JM, Palar K, Gooding HC, Garber AK, Tabler JL, Whittle HJ, et al. Food Insecurity, Sexual Risk, and Substance Use in Young Adults. J Adolesc Health 2021;68(1):169-177. doi:10.1016/j.jadohealth.2020.05.038

131. Demartini KS, Carey KB. Optimizing the use of the AUDIT for alcohol screening in college students. Psychol Assess 2012;24(4):954-963. doi:10.1037/a0028519

132. Devos-Comby L, Lange JE. Standardized measures of alcohol-related problems: a review of their use among college students. Psychol Addict Behav 2008;22(3):349-361. doi:10.1037/0893-164X.22.3.349

133. Jensen M, Hussong A. Text message content as a window into college student drinking: Development and initial validation of a dictionary of "alcohol talk". Int J Behav Dev 2021;45(1):3-10. doi:10.1177/0165025419889175

134. Hazzard VM, Loth KA, Hooper L, Black Becker C. Food Insecurity and Eating Disorders: a Review of Emerging Evidence. Curr Psychiatry Rep 2020;22(12):74. doi:10.1007/s11920-020-01200-0

135. Barry MR, Sonneville KR, Leung CW. Students with Food Insecurity Are More Likely to Screen Positive for an Eating Disorder at a Large, Public University in the Midwest. J Acad Nutr Diet 2021;121(6):1115-1124. doi:10.1016/j.jand.2021.01.025

136. Royer MF, Ojinnaka CO, Bruening M. Food Insecurity is Related to Disordered Eating Behaviors Among College Students. J Nutr Educ Behav 2021;53(11):951-956. doi:10.1016/j.jneb.2021.08.005

137. Grammer AC, Fitzsimmons-Craft EE, Laing O, De Pietro B, Wilfley DE. Eating Disorders on College Campuses in the United States: Current Insight on Screening, Prevention, and Treatment. Curr Psychopharmacol 2020;9(2):91-102. doi:10.2174/2211556009999200416153022

138. Stice E, Shaw H. Eating disorders: Insights from imaging and behavioral approaches to treatment. J Psychopharmacol 2017;31(11):1485-1495. doi:10.1177/0269881117722999

139. Prickett C, Brennan L, Stolwyk R. Examining the relationship between obesity and cognitive function: a systematic literature review. Obes Res Clin Pract 2015;9(2):93-113. doi:10.1016/j.orcp.2014.05.001

140. Na M, Dou N, Ji N, Xie D, Huang J, Tucker KL, Gao X. Food Insecurity and Cognitive Function in Middle to Older Adulthood: A Systematic Review. Adv Nutr 2020;11(3):667-676. doi:10.1093/advances/nmz122

141. Royer MF, Guerihault N, Blair Braden B, Laska MN, Bruening M. Food Insecurity Is Associated with Cognitive Function: A Systematic Review of Findings across the Life Course. International Journal of Translational Medicine 2021;1(3):205-222. doi:10.3390/ijtm1030015

142. Gallegos D, Eivers A, Sondergeld P, Pattinson C. Food Insecurity and Child Development: A State-of-the-Art Review. Int J Environ Res Public Health 2021;18(17):8990. doi: 10.3390/ijerph18178990

143. Gao X, Scott T, Flacon LM, Wilde PE, Tucker KL. Food insecurity and cognitive function in Puerto Rican adults. Am J Clin Nutr 2009;89(4):1197-1203. doi:10.3945/ajcn.2008.26941

144. Frith E, Loprinzi PD. Food insecurity and cognitive function in older adults: Brief report. Clin Nutr 2018;37(5):1765-1768. doi:10.1016/j.clnu.2017.07.001

145. Portela-Parra ET, Leung CW. Food Insecurity Is Associated with Lower Cognitive Functioning in a National Sample of Older Adults. J Nutr 2019;149(10):1812-1817. doi:10.1093/jn/nxz120

146. Holdsworth SJ, Bammer R. Magnetic resonance imaging techniques: fMRI, DWI, and PWI. Semin Neurol 2008;28(4):395-406. doi:10.1055/s-0028-1083697

147. Wilens TE, Carrellas NW, Martelon M, Yule AM, Fried R, Anselmo R, McCabe SE. Neuropsychological functioning in college students who misuse prescription stimulants. Am J Addict 2017;26(4):379-387. doi:10.1111/ajad.12551

148. Arain M, Haque M, Johal L, Mathur P, Nel W, Rais A, Sandhu R, Sharma S. Maturation of the adolescent brain. Neuropsychiatr Dis Treat 2013;9:449-461. doi:10.2147/NDT.S39776

149. Weaver RR, Vaughn NA, Hendricks SP, McPherson-Myers PE, Jia Q, Willis SL, Rescigno KP. University student food insecurity and academic performance. J Am Coll Health 2020;68(7):727-733. doi:10.1080/07448481.2019.1600522

150. Camelo K, Elliott M. Food Insecurity and Academic Achievement Among College Students at a Public University in the United States. Journal of College Student Development 2019;60(3):307-318. doi:10.1353/csd.2019.0028.

151. Martinez SM, Frongillo EA, Leung CW, Ritchie L. No food for thought: Food insecurity is related to poor mental health and lower academic performance among students in California's public university system. J Health Psychol 2020;25(12):1930-1939. doi:10.1177/1359105318783028

152. Silva MR, Kleinert WL, Sheppard AV, Cantrell KA, Freeman-Coppadge DJ, Tsoy E, RIBERTS T, PEARROW M. The Relationship Between Food Security, Housing Stability, and School Performance Among College Students in an Urban University. Journal of College Student Retention: Research, Theory & Practice 2015;19(3):284-299. doi:10.1177/1521025115621918

153. Hege A, Stephenson T, Pennell M, Revlett B, VanMeter C, Stahl D, Oo K, Bressler J, Crosby C. College Food Insecurity: Implications on Student Success and Applications for Future Practice. Journal of Student Affairs Research and Practice 2020;58(1):44-61. Doi:10.1080/19496591.2020.1726359

154. Phillips E, McDaniel A, Croft A. Food Insecurity and Academic Disruption Among College Students. Journal of Student Affairs Research and Practice 2018;55(4):353-372. doi:10.1080/19496591.2018.1470003

155. van Woerden I, Hruschka D, Bruening M. Food insecurity negatively impacts academic performance. Journal of Public Affairs 2019;19(3). doi:10.1002/pa.1864

156. Zigmont V, Linsmeier A, Gallup P. Understanding the Why of College Student Food Insecurity. Journal of Hunger & Environmental Nutrition 2019;16(5):595-610. doi:10.1080/19320248.2019.1701600

157. Davinelli S, Ali S, Solfrizzi V, Scapagnini G, Corbi G. Carotenoids and Cognitive Outcomes: A Meta-Analysis of Randomized Intervention Trials. Antioxidants (Basel) 2021;10(2):223. doi:10.3390/antiox10020223

158. Macready AL, Kennedy OB, Ellis JA, Williams CM, Spencer JPE, Butler LT. Flavonoids and cognitive function: a review of human randomized controlled trial studies and recommendations for future studies. Genes Nutr 2009;4(4):227-242. doi:10.1007/s12263-009-0135-4

159. Probst YC, Guan VX, Kent K. Dietary phytochemical intake from foods and health outcomes: a systematic review protocol and preliminary scoping. BMJ Open 2017;7(2):e013337. doi:10.1136/bmjopen-2016-013337

160. Noble EE, Hsu TM, Kanoski SE. Gut to Brain Dysbiosis: Mechanisms Linking Western Diet Consumption, the Microbiome, and Cognitive Impairment. Front Behav Neurosci 2017;11:9. doi:10.3389/fnbeh.2017.00009

161. Brown AGM, Esposito LE, Fisher RA, Nicastro HL, Tabor DC, Walker JR. Food insecurity and obesity: research gaps, opportunities, and challenges. Transl Behav Med 2019;9(5):980-987. doi:10.1093/tbm/ibz117

162. Jomaa L, Diab-El-Harake M, Kharroubi S, Mattar L. Associations Between Food Literacy and Food Insecurity Among College Students in Lebanon: A Cross-Sectional Study. Current Developments in Nutrition 2022;6(1):120. doi:10.1093/cdn/nzac051.036

163. Henry LR. Understanding Food Insecurity Among College Students: Experience, motivation, and local solutions. Annals of Anthropological Practice 2017;41(1):6-19. doi:10.1111/napa.12108

164. McGuirt JT, Jilcott Pitts SB, Labban JD, Anderson Steeves ET, Haynes-Maslow L, Henry S, Gustafson A. Evidence of Geospatial and Socioeconomic Disparities in Access to Online Grocery Shopping for Fresh and Frozen Produce in North Carolina. J Acad Nutr Diet 2022;122(11):2106-2114. doi:10.1016/j.jand.2022.05.008

Tables and Figures

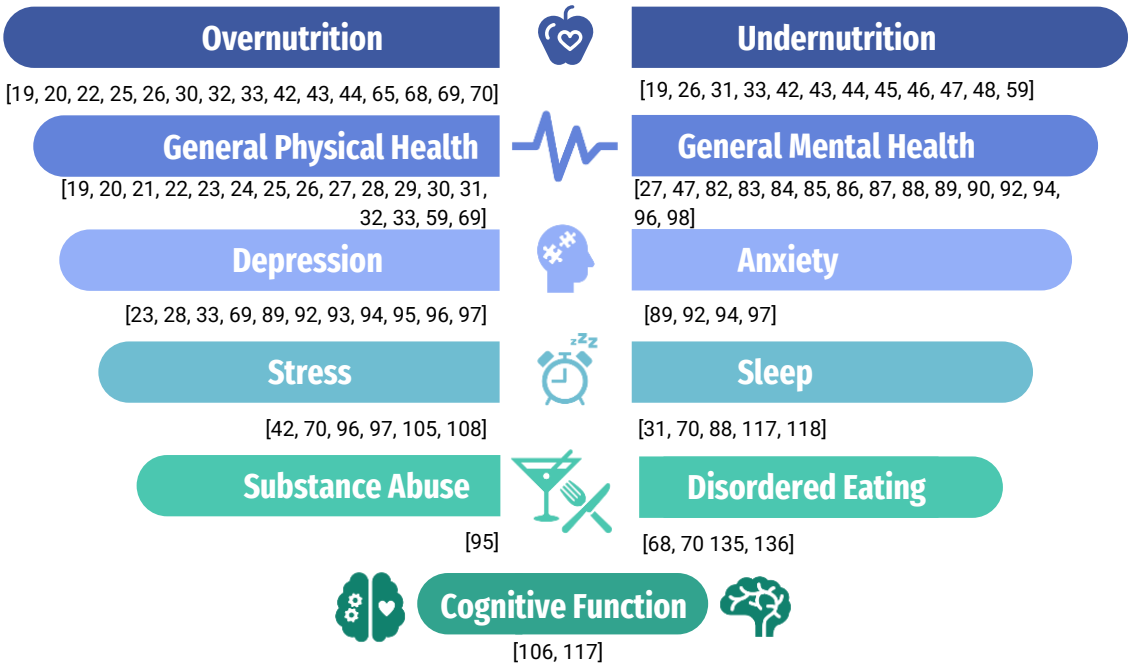


Figure 1. Physiological, psychosocial, and cognitive outcomes associated with the experience of food insecurity in the college student population. The bracketed numbers correspond with the literature published from 2017 – 2023 that included the corresponding health outcomes.

Table 1. Included studies published within the years 2017 – 2023 examining the relationship between food insecurity and physiological health outcomes in college students in the United States.

Citation	Sample Size	USDA Tool	Health Outcome	Assessment Tool	Outcomes Associated with FI
Knol et.al., 2017[20]	n = 351	10-item	1. Body composition 2. General health	1. Anthropometrics: self-reported BMI 2. Self-reported description of general health	1. No Δ in BMI 2. ↓ General health
Bruening et.al., 2018[42]	n = 1138	6-item	1. Body composition 2. Eating behaviors	1. Anthropometrics: BMI collected by trained researcher 2. NCI 26-item DSQ	1. No Δ in BMI 2. ↓ Diet quality
McArthur et. al., 2018[21]	n = 1,093	10-item	1. Body composition 2. General health	1. Anthropometrics: self-reported BMI 2. Self-reported description of general health	1. ↑ BMI 2. ↓ General health
McArthur et. al., 2018[22]	n = 456	10-item	1. General health	1. Self-reported description of general health	1. ↓ General health
Payne-Sturges et. al., 2018[23]	n = 237	18-item	1. Body composition 2. General health	1. Anthropometrics: self-reported height and weight 2. Self-reported description of general health	1. No results reported 2. ↓ General health
El Zein et. al., 2019[70]	n = 855	10-item	1. Body composition	1. Anthropometrics: BMI and waist circumference collected by trained researcher	1. No Δ in BMI or waist circumference
Leung et.al, 2019[44]	n = 851	10-item	1. Body composition 2. Dietary intake	1. Anthropometrics: self-reported BMI 2. NCI 26-item DSQ	1. ↑ BMI in very low FSS only 2. ↓ F/V; ↓ Whole-grains; ↓ Fiber; ↑ Added sugars; ↑ SSB
Martinez et.al., 2019[19]	n = 8,705	6-item	1. F/V intake 2. Body composition 3. General health	1. Self-reported daily servings 2. Anthropometrics: self-reported BMI 3. Self-reported description of general health	1. ↓ Daily servings of F/V 2. ↑ BMI 3. ↓ General health
Soldavini et. al., 2019[25]	n = 4,819	10-item	1. Body composition 2. General health	1. Anthropometrics: self-reported BMI	1. ↑ BMI 2. ↓ General health

				2. Self-reported description of general health	
El Zein et.al., 2020[43]	n = 683	10-item	1. Obesity 2. Dietary intake	1. Anthropometrics: BMI, waist, hip, and neck circumference collected by trained researcher 2. NCI 26-item DSQ	1. ↑ BMI; ↑ Waist and hip circumference in females only 2. ↓ F/V; ↑ Added sugars; ↑ SSB
Olfert et. al., 2020[30]	n = 22,153	10-item	1. Body composition 2. General health	1. Anthropometrics: self-reported BMI 2. Self-reported description of general health	1. ↑ BMI 2. ↓ General health
Soldavini et. al., 2020[24]	n = 4,829	10-item	1. General Health	1. Self-reported description of general health	1. ↓ General health
Umeda et. al., 2020[69]	n = 176	10-item	1. Recent pain Experience 2. Body composition	1. BDS 2. Anthropometrics: self-reported BMI	1. ↑ Pain interference 2. No Δ in BMI
Davitt et. al., 2021[26]	n = 1,434	6-item	1. Dietary intake 2. Body composition 3. General health	1. Validated food frequency screener for fruit, vegetables, and fiber 2. Anthropometrics: self-reported BMI 3. Self-reported description of general health	1. No Δ in total F/V intake, ↓ whole fruit and "other vegetables" 2. ↑ BMI 3. ↓ General health
Frank et. al., 2021[68]	n = 232	6-item	1. Body composition	1. Anthropometrics: self-reported BMI	1. No Δ in BMI
Hiller et. al., 2021[29]	n = 675	10-item	1. General health	1. Self-reported description of general health	1. ↓ General health
Huelskamp et. al., 2021[65]	n = 547	10-item	1. Body composition	1. Anthropometrics: self-reported BMI	1. ↑ BMI
Laska et. al., 2021[47]	n = 13,720	2-item screener	1. Body composition 2. Dietary intake	1. Anthropometrics: self-reported BMI 2. YRBS	1. ↑ BMI 2. ↑ SSB; ↑ Fast food; No Δ in F/V intake
Leung et. al., 2021[28]	n = 793	10-item	1. General health	1. Self-reported description of general health	1. ↓ General health
Mei et. al., 2021[45]	n = 1,033	6-item	1. Dietary intake 2. Beverage intake	1. NCI 26-item DSQ 2. BEV-Q	1. ↓ F/V intake, ↓ Fiber, ↑ Added sugars 2. ↑ SSB
Ryan et. al., 2021[31]	n = 257	6-item	1. General health 2. Dietary intake	1. Self-reported description of general health 2. All Day F/V Screener 3. BEV-Q	1. ↓ General health 2. No Δ in F/V intake

			3. Beverage intake		3. ↑ SSB (> 100kcals per day)
Sackey et. al., 2021[48]	n = 302	6-item	1. Diet quality	1. Self-reported rating of diet quality	1. ↓ Diet quality
Silva et. al., 2021[46]	n = 502	2-item screener and 6-item	1. Dietary intake 2. Body composition	1. ASA24, HEI 2. Anthropometrics: self-reported BMI	1. ↓ HEI 2. No results reported
Willis, 2021[33]	n = 300	6-item	1. Body composition 2. General health 3. F/V intake	1. Anthropometrics: self-reported BMI 2. Self-reported description of general health 3. Self-reported weekly servings	1. No Δ BMI 2. ↓ General health 3. Δ Between groups not reported
Ahmed et. al., 2022[27]	n = 1,989	10-item	1. General health	1. Self-reported description of general health	1. ↓ General health
Mohr et.al., 2022[59]	n = 60	2-item screener	1. Gut microbiome	1. 16S rRNA amplicon sequencing using fecal samples	1. Δ in microbial and metabolite composition
Soldavini et. al., 2022[32]	n = 263	10-item	1. Body composition 2. General health	1. Anthropometrics: self-reported BMI 2. Self-reported description of general health	1. No Δ BMI 2. ↓ General health

Abbreviations

Automated Self-Administered 24-hour Dietary Assessment Tool: ASA24; Beverage Intake Questionnaire-15; BEV-Q; Bodily Pain Scale: BDS; Body mass index: BMI; Dietary Screener Questionnaire: DSQ; Fruit and Vegetables: F/V; Healthy Eating Index: HEI; National Cancer Institute: NCI; Sugar sweetened beverage: SSB; Youth Risk Behavior Surveillance System: YRBS.

Table 2. Included studies published within the years 2017 – 2023 examining the relationship between food insecurity, mental health, and cognitive outcomes in college students in the United States.

Citation	Sample Size	USDA Tool	Health Outcome	Assessment Tool	Outcomes Associated with FI
Bruening et. al., 2018[42]	n = 1,138	6-item	1. Stress 2. Depression	1. Cohen's PSS 2. National College Health AssessmentIIsurvey	1. ↑ Stress 2. ↑ Depression
Payne-Sturges et. al., 2018[23]	n = 237	18-item	1. Depression	1. PHQ-9	1. ↑ Depressive symptoms
Wattick et. al., 2018[90]	n = 1,956	Used the USDA tool – version not specified	1. Depression 2. Anxiety	1. & 2. CDC Healthy Days Core Module	1. ↑ Depression 2. ↑ Anxiety
Diamond et. al., 2019[96]	n = 1,229	6-item	1. Depression 2. Stress 3. Social isolation 4. Resiliency	1. PHQ-9 2. Cohen's PSS 3. 3-item Loneliness Scale 4. 6-item Brief Resiliency Scale	1. ↑ Depression 2. ↑ Stress 3. ↑ Social isolation 4. ↓ Perceived resiliency
El Zein et. al., 2019[70]	n = 855	10-item	1. Stress 2. Sleep 3. Disordered eating behaviors	1. Cohen's PSS 2. PSQI 3. Eating Attitudes Test-26	1. ↑ Stress 2. ↓ Sleep 3. ↑ Risk of disordered eating behaviors
Raskind et. al., 2019[94]	n = 2,377	6-item	1. Depression 2. Anxiety 3. Hope	1. PHQ-9 2. ASI-3 3. 6-item Adult State Hope Scale	1. ↑ Depression 2. ↑ Anxiety 3. ↓ Hope
Becerra et. al., 2020[87]	n = 302	6-item	1. Psychological distress	1. Kessler-6 Scale	1. ↑ Psychological distress
Becerra et. al., 2020[118]	n = 282	6-item	1. Sleep	1. Berlin Sleep Questionnaire	1. ↑ Tiredness, sleepiness, fatigue; ↓ Sleep duration
Haskett et. al., 2020[85]	n = 1,330	10-item	1. Well-being	1. WHO-5	1. ↓ Psychological well-being

Martinez et. al., 2020[151]	n = 8,765	6-item	1. Mental health	1. Nine items from the National College Health AssessmentIIsurvey	1. ↑ Poor mental health
Reeder et. al., 2020[95]	n = 131	6-item	1. Depression 2. Substance abuse	1. PHQ-9 2. AUDIT	1. ↑ Depression 2. No Δ in AUDIT scores
Richard et. al., 2020[117]	n = 153	6-item	1. Sleep 2. Stress 3. Dietary cognitive restraint	1. PSQI 2. PSS 3. TFEQ-R18V2	1. ↓ Sleep 2. ↑ Stress 3. ↑ Uncontrolled eating
Umeda et. al., 2020[69]	n = 176	10-item	1. Depression	1. BDI-II	1. No Δ in depressive symptoms
Barry et. al., 2021[135]	n = 851	10-item	1. Disordered eating behaviors	1. SCOFF-5	1. ↑ Positive SCOFF-5 screens
Cockerham et. al., 2021[98]	n = 55	6-item	1. Social support	1. MSPSS	1. ↓ Social support
Coffino et. al., 2021[97]	n = 263	6-item	1. Depression 2. Anxiety 3. Stress	1.- 3. DASS-21	1. ↑ Depression 2. ↑ Anxiety 3. ↑ Stress
DeBate et. al., 2021[83]	n = 1,743	6-item	1. Psychological well-being 2. Psychological distress 3. Loneliness 4. Resilience 5. Suicidal behaviors	1. Diener Flourishing Scale 2. Kessler-6 Scale 3. UCLA 3-item Loneliness Scale 4. CD-RISC 5. SBQ-R	1. ↓ Psychological well-being 2. ↑ Psychological distress 3. ↑ Loneliness 4. ↓ Resiliency 5. ↑ Suicidal behaviors
Frank et. al., 2021[68]	n = 232	6-item	1. Disordered eating behaviors	1. EES	1. ↑ Emotional eating
Hagedorn et. al., 2021[88]	n = 17,686	10-item	1. Sleep quality 2. Mental well-being	1. PSQI 2. CDC Healthy Days Core Module	1. ↓ Sleep 2. ↑ Days with poor mental health
Laska et. al., 2021[47]	n = 13,720	2-item screener	1. Resiliency	1. 6-item Brief Resiliency Scale	1. ↓ Perceived resiliency
Leung et. al., 2021[28]	n = 793	10-item	1. Depression	1. PHQ-4	1. ↑ Depression

Royer et. al., 2021[136]	n = 533	10-item	1. Disordered Eating Behaviors	1. EDE-Q	1. ↑ Global DEBS, eating concern, shape concern, weight concern, no Δ in restraint
Ryan et. al., 2021[31]	n = 257	6-item	1. Sleep	1. PSQI	1. No Δ in sleep patterns
Willis, 2021[33]	n = 300	6-item	1. Depression	1. CES-D	1. ↑ Depressive symptoms
Ahmed et. al., 2022[27]	n = 1,989	10-item	1. Psychological well-being	1. Diener Flourishing Scale	1. ↓ Psychological well-being
Coakley et. al, 2022[89]	n = 833	10-item	1. Depression 2. Anxiety 3. Mental well-being	1. PHQ-2 2. GAD-2 3. CDC Healthy Days Core Module	1. ↑ Depression 2. ↑ Anxiety 3. ↑ Risk of fair/poor mental health
Guerithault et. al., 2022[106]	n = 40	10-item	1. Brain Activity 2. Executive function 3. Childhood trauma	1. Structural and Functional MRI 2. BRIEF-2 3. ACEs	1. Δ Functional connectivity between key cognitive networks 2. ↓ Executive function 3. ↑ ACEs
Guzman et. al., 2022[86]	n = 441	6-item	1. Psychological distress 2. Loneliness	1. Kessler-6 Scale 2. UCLA 3-item Loneliness Scale	1. ↑ Psychological distress 2. ↑ Loneliness
Marmolejo et. al., 2022[82]	n = 48,103	6-item	1. Psychological well-being 2. Loneliness 3. Psychological distress	1. Diener Flourishing Scale 2. UCLA 3-item Loneliness Scale 3. Kessler-6 Scale	1. ↑ Psychological distress 2. ↑ Loneliness 3. ↓ Psychological well-being
Neal et. al., 2022[93]	n = 589	6-item	1. Depression	1. PHQ-9	1. ↑ Depression
Oh et. al., 2022[92]	n = 96,379	2-item screener	1. Depression 2. Anxiety 3. Loneliness	1. PHQ-9 2. GAD-7 3. UCLA 3-item Loneliness Scale	1. ↑ Depression 2. ↑ Anxiety 3. ↑ Loneliness

Abbreviations

Alcohol Use Disorders Identification Test: AUDIT; Anxiety Sensitivity Index: ASI-3; Beck Depression Inventory II: BDI-II; Behavior Rating Inventory of Executive Function-2: BRIEF-2; Center for Epidemiologic Studies Depression Scale: CES-D; Cohen's Perceived Stress Scale: Cohen's PSS; Connor-Davison Resiliency Scale: CD-RISC; Depression Anxiety and Stress Scales – 21-item short form: DASS-21; Eating Disorder Examination Questionnaire: EDE-Q; Emotional Eating Survey: EES; Everyday Discrimination Scale: EDS; Generalized Anxiety Disorder Screener: GAD-2; General Anxiety Disorder – 7: GAD-7; Multidimensional Scale of Perceived Social Support: MSPSS; Patient Health Questionnaire-2: PHQ-2; Patient Health Questionnaire – 9: PHQ-9;Pittsberg Sleep Quality Index: PSQI; Sick, Control, One stone, Fat, Food: SCOFF-5; Suicidal Behaviors Questionnaire: SBQ-R; Three Factor Eating Questionnaire Revised: TFEQ-R18V2; World Health Organization Five Factor Well-Being Index: WHO-5.

STATEMENT OF PURPOSE

The understanding of food insecurity (FI) as it relates to acute and chronic health-outcomes is an emerging field of interest. In the United States, college students experience food FI at a prevalence that is four times higher than the national average. This increased risk of inadequate quality and/or quantity of food in the diet demonstrates extensive vulnerability to the multitude of health-consequences associated with FI. Therefore, the purpose of this dissertation was to explore the physiological and cognitive impacts of food insecurity using novel, innovative, and objective biomarkers, with the goal of providing an impetus for addressing effective interventions aimed at improving nutrient-dense food availability and accessibility in the college student population.

The approach of this dissertation was to identify if these outcomes are generalizable to the college student population and determine interventions for improving food security status and subsequent physiological and cognitive outcomes. Chapter 2 of this dissertation is a systematic review of spectroscopy-based measurements as a proxy for fruit and vegetable intake, including the criterion validity of the Veggie Meter® against plasma carotenoids and validated dietary assessments. Chapter 3 includes a validation study of Diet ID™, a novel technology that implements a patented Diet Quality Photo Navigation (DQPN) algorithm to estimate dietary patterns, diet quality, and nutrient intake. Chapter 4 details the use of the Veggie Meter®, Diet ID™, and plasma carotenoids to assess the efficacy of food access resources on improvements to diet-related biomarkers. Chapter 5 explores how additional health-outcomes may be impacted by the experience of FI, particularly cognitive processes in the various domains of executive function.

CHAPTER 2

Criterion-related Validity of Spectroscopy-based Skin Carotenoid Measurements as a Proxy for Fruit and Vegetable Intake: A Systematic Review

Introduction

Adequate fruit and vegetable intake is associated with positive health outcomes and reduced risk of developing chronic diseases.[1] Fruits and vegetables contain a variety of health-promoting bioactive components, such as phytochemicals, vitamins, and minerals.[2] Carotenoids are a class of compounds in fruits, vegetables, grains, nuts, legumes and some animal products that have demonstrated protective properties against macular degeneration, cardiovascular disease, sarcopenia, skin damage from ultraviolet radiation (UV) exposure, and protection against oxidative damage.[3, 4]

With over 700 identified carotenoids, the most prevalent and highly-researched that correlate with fruit and vegetable intake are α-carotene, β-carotene, lycopene, lutein, zeaxanthin, and β-cryptoxanthin.[5] Carotenoids are found in a variety of yellow, red, pink, orange, and green pigmented plant products and some animal products, such as eggs and pink or red fleshed seafood.[5] Carotenoids are exogenous compounds that must be acquired through dietary sources and cannot be synthesized *de novo*; Thus, due to the array of foods that contain high amounts of carotenoids, circulating blood carotenoids are considered the gold-standard biomarker of fruit and vegetable intake.[6,7]

For research, evaluation, and surveillance purposes, fruit and vegetable intake is objectively assessed using carotenoids measured in serum or plasma samples, or estimated subjectively using dietary recall methods. Serum is derived from coagulated whole blood and plasma from anticoagulated blood. Both media can be extracted and analyzed for carotenoid concentrations using High Performance Liquid Chromatography (HPLC) or Liquid Chromatography-Tandem Mass Spectroscopy (LC-MS).[8-11] Although blood biomarkers are a relatively accurate measure and considered the standard for assessing fruit and vegetable intake,

the process of collecting blood samples is mildly invasive and may not be reflective of long-term dietary intake due to the short half-lives of circulating carotenoids.[12, 13] Thus, plasma or serum carotenoids are reflective of recent dietary intake and are detectable in the blood for approximately two weeks after intake.[14, 15]

In addition to serum and plasma carotenoids, FV intake is also measured using subjective dietary recall methods or dietary observations.[16] Commonly used methods include observer-recorded food records, 24-hour dietary recall, dietary record, or food frequency questionnaires (FFQs). However, all of these dietary recall methods are prone to time burden, subjective biases, and intervention-related biases that may result in inaccurate representation of true dietary intake.[17, 18] Because the existing methods for recalling dietary intake contain error and bias, such measures may negatively impact the validity of quantifying fruit and vegetable intake, demonstrating the need for an objective indicator of dietary intake.[19]

Spectroscopy has emerged as a non-invasive, objective approach to measuring dietary intake of fruits and vegetables.[20] Spectroscopy measures the absorption and emission of light waves at a specific wavelength to identify the type and density of molecular compounds in the skin.[14] Carotenoids are easily identifiable bioactive compounds that can be quantified using optical spectroscopy as they are deposited and primarily visible in the stratum corneum of the skin at a UV range of 400nm-500nm.[21] To account for intra-individual variability, studies using spectroscopy to determine carotenoid status often assess multiple locations on the human body and use duplicate or triplicate measurements.[22-30] The index finger, palm, inner arm, and heel are frequently used as sites for spectroscopy-based carotenoid measurement because the thickness of the skin prevents other skin chromophores, like melanin, from obstructing the detection of carotenoid compounds.[21] Additionally, the index finger, palm, inner arm, and heel locations do

not experience excessive sun exposure, a major factor in altering the molecular structure of carotenoids, which decreases the amount of identifiable carotenoid molecules.[4]

There are various types of spectroscopy technology used to identify and quantify carotenoids in the skin, including resonance Raman spectroscopy (RRS), pressure-mediated reflection spectroscopy (RS), and spectrophotometers. Reflection spectroscopy uses a broad-band light source (460nm – 500nm) to measure the density of skin carotenoids with minimal interference from other compounds.[14] Subsequently, the carotenoids are superimposed on a reflection-based absorption spectrum for reference.[14] Supradermal pressure is added during the measurement to temporarily limit blood flow to the assessment location to reduce the presence of confounding molecules, specifically oxygenated or deoxygenated hemoglobin, which can interfere with carotenoid absorption, thus minimizing the misidentification of carotenoid compounds.[14] Resonance Raman spectroscopy utilizes light photons to manipulate the conjugated bonds of the carotenoid molecules to generate excitatory Raman signals.[31, 32] The excitatory signals initiate vibrational state changes that alter the bond strength of carotenoids, resulting in a distinct signal on the Raman spectra.[31] Resonance Raman spectroscopy detects combined concentrations of skin carotenoids with efficiency and precision; however, this method requires expensive instrumentation and analysis software.[14] Reflection spectroscopy and RRS detect the major blood concentrations of carotenoids, but do not detect colorless carotenoids, such as phytoene and phytofluene due to differences in spectral regions.[33] Spectrophotometers are measurement devices that evaluate and analyze the color of dermatological pigments yellowness and redness, which are reflective of skin carotenoid concentration.[34] Spectrophotometers measure the intensity of light transmitted through a solvent and identify carotenoid compounds due to the absorption or reflection in a specific wavelength within the

color spectrum (~350nm-500nm).[34, 35] These advancements in technology have resulted in non-invasive, convenient, and efficient methods of measuring carotenoid status as a proxy for fruit and vegetable intake.[21]

The development and validation of a non-invasive, objective measurement to assess fruit and vegetable intake has the potential to change the standard for collecting accurate dietary intake data. The aim of this systematic review was to examine criterion-related validity of three methods of spectroscopy as a proxy for fruit and vegetable intake by evaluating studies that examined associations between skin carotenoid status measured via spectroscopy and (1) serum or plasma carotenoid concentration or (2) self-reported dietary intake or (3) both serum or plasma carotenoid concentration and self-reported dietary intake.

Methods

In accordance with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) statement for improved reporting of systematic reviews, the protocol for this systematic review was prospectively registered with PROSPERO (registration number 114605).[36]

Literature Search Strategy

To identify relevant studies, literature searches of PubMed, Excerpta Medica Database (EMBASE), Cumulative Index of Nursing and Allied Health Literature (CINAHL), ProQuest Search, Cochrane Database of Systematic Reviews (CDSR), and Cochrane Central Register of Controlled Trials (CENTRAL) were performed in December 2018 to identify studies addressing criterion-related validity of spectroscopy for assessing carotenoid levels of human skin as a measure of dietary fruit and vegetable intake. Primary search terms for spectroscopy included spectrum analysis, Veggie Meter®, and skin reflectance. Primary search terms for carotenoids

included carotenoids, α-carotene, and β-carotene. Additional carotenoids, such as lutein and lycopene were added to expand the search (**Table 1**). Attempts to include human skin and diet as search concepts consistently led to inadequate retrieval of studies, so these concepts were not included in the final search strategies. Exact search terms and the PubMed search strategy are available in **Supplemental Table 1**. All articles identified through literature searching were loaded into Endnote 9.1 (Clarivate Analytics, Philadelphia, PA), which was used to identify and remove publications prior to 1990, newspaper articles, and duplicate citations. Remaining items were then loaded into Rayyan QCRI for the initial review of eligibility criteria.[37]

Three authors independently screened titles and abstracts with disagreements resolved by additional authors. The article abstracts were reviewed using strict inclusion criteria. If the abstract did not report a correlation or validation between spectroscopy and dietary intake and/or serum or plasma carotenoids, the abstract was excluded prior to the full-text review phase. Full-text review was then independently performed by two authors to verify eligibility based on study protocol and inclusion criteria. A third author was consulted and assisted with conflict resolution. References from the included articles were hand-searched by two authors to ensure no relevant articles were missed in the initial database searches. Seven additional manuscripts eligible for inclusion were identified during this process.[22, 29, 30, 38-41]

Inclusion and Exclusion Criteria

Peer-reviewed publications assessing criterion-related validity of spectroscopy using human skin against dietary intake and/or plasma or serum carotenoids were the focus of this review. Animal models and *in vitro* studies were excluded. Studies using whole fruits and vegetables with naturally occurring carotenoids were included. Studies using supplementation were excluded due to unrealistic concentrations of carotenoids, which would not be present in whole fruits and

vegetables, and limited data regarding the metabolism and bioavailability of dietary supplements and extracts. It should be acknowledged that multiple studies that used high-dose supplementation strategies compared spectroscopy-based skin carotenoid measurements to serum or plasma carotenoids and/or dietary intake at baseline prior to supplementation.[42-46] However, due to our previously defined criteria regarding the exclusion of supplementation studies, these studies were not included in the full analysis. Additionally, studies comparing spectroscopy to dermal biopsies were excluded from this review. Although dermal biopsies can confirm the accuracy of spectroscopic measurements, such biopsies would not corroborate a relationship between fruit or vegetable intake, and therefore would not support the objective of this review. There were no exclusion criteria for study design, statistical methods/tools, or population characteristics. Explicit eligibility criteria are displayed in **Table 1**.

Data Extraction

Data extraction was performed independently by two authors. A third author reviewed the information and compiled the two extraction data sets into a single entry to ensure a comprehensive analysis. The following information was extracted: sample characteristics (mean age, race/ethnicity, sex, and Body Mass Index (BMI; kg/m^2) if provided by the author), study design, type of spectroscopy used, intervention details when applicable, statistical interpretation of the correlation or validation of spectroscopy against blood and/or dietary intake, and primary findings.

Risk of Bias and Quality Evaluation

Quality of the studies was assessed independently by two authors using a modified and combined version of the Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) Checklist[47] and the National Institutes of Health (NIH) Quality Assessment Tool for

Observational Cohort and Cross-Sectional Studies.[48] Authors graded the publications based on the quality assessment and risk of bias criteria. If the two reviewing authors did not agree on overall quality of the publication, discrepancies were mediated by a third author independently reviewing the discrepancy without prior knowledge of the conflicting ratings. Obvious study limitations such as small sample sizes, population homogeneity, study design and lack of control group were considered; however, studies were graded holistically.

For the purposes of this manuscript, the data were considered as reported by the author in each study. The correlation strength was also interpreted according to how the authors analyzed the data within the individual papers. However, if the manuscripts did not verbally describe the strength of the correlation coefficients, the following interpretations were used: very strong (0.90-1.0), strong (0.70-0.89), moderate (0.50-0.69), low (0.30-0.49), weak (0.20-0.29), or negligible ($\leq$ 0.19).[49]

Results

Overview of Search Results

The comprehensive literature search resulted in 16,134 potentially relevant articles based on the initial database literature search. Removal of duplicate articles, publications prior to 1990, and non-peer reviewed manuscripts resulted in 7,931 articles. The initial title and abstract screening for eligibility criteria resulted in 54 articles selected for the full-text review. Following the completion of full-text review and hand search reference screening, 29 studies satisfied the inclusion criteria and were included in the present review. The comprehensive article selection process is depicted in **Figure 1**.

Characteristics of Included Studies

The included articles varied in study design and comprised of cross-sectional studies (n=21), prospective cohort studies (n=5), randomized crossover trial (n=1), single-arm experimental trial (n=1), and randomized control trial (n=1). The quality of the studies was classified as very good (n=5), good (n=20), fair (n=4), and poor (n=0).

A majority of the studies involved adult participants; however, four studies examined infants[38, 41, 50, 51] and six studies evaluated carotenoid status in children.[52-57] The number of participants included in the studies ranged from 29[15, 58] to 497.[59] The race/ethnicity of the participants was variable amongst the studies. Six studies included racially and ethnically diverse populations,[53-57, 59] thirteen studies focused on predominantly Caucasian subjects,[15, 22, 24-30, 38, 60-62] two studies only had participants from Thailand,[52, 58] and eight studies did not report the race or ethnicity of the participants.[23, 39-41, 50, 51, 63, 64]

Type of Spectroscopy Used

Three methods of skin carotenoid detection were used in the included articles: RRS, RS, and spectrophotometers. A majority of the studies (n=20) used RRS to measure skin carotenoids, a technology developed by Werner Gellermann et al.[31, 65] Seven studies used the NuSkin BioPhotonic Scanner® developed by Pharmanex and thirteen studies used unspecified custom-built RRS devices. Seven of the studies used spectrophotometers to measure skin carotenoids; four studies used the CM700D spectrophotometer by Konica Minolta,[23, 25, 27, 28] two studies used CM2600D spectrophotometer by Konica Minolta,[29, 30] and one study used the Spectro-Guide 450 Gloss 6801 spectrophotometer.[24] Two of the studies used RS to measure skin carotenoids using the Veggie Meter® device developed by Igor V. Ermakov and Werner Gellermann.[59, 62] Information about the types of spectroscopy devices is presented in **Table 2**.

Results of Included Studies

The included studies can be differentiated by the method of comparison against spectroscopy technique. The efficacy of spectroscopy as an objective biomarker for dietary intake was compared against serum or plasma carotenoid concentrations, self-reported dietary intake data, or both serum or plasma carotenoid concentrations and dietary intake data.

Spectroscopy and Plasma or Serum Carotenoids

Of the included studies, n=11 articles compared spectroscopy to plasma (n=3) or serum (n=8) carotenoid concentration using HPLC (**Table 3**). Multiple studies correlated spectroscopy to the combined total carotenoids in the blood[39-41, 50, 51, 64] and other studies analyzed both total blood carotenoid in conjunction with individual carotenoid analysis, such as α-carotene, β-carotene, lycopene, lutein, β-cryptoxanthin, and zeaxanthin and correlated spectroscopy data with each specific carotenoid compound.[15, 28, 61-63] A majority of the studies analyzing serum or plasma carotenoids used RRS with the exception of two studies, one study using RS[62] and another using the CM700D spectrophotometer.[28]

The correlation coefficients between blood carotenoids and spectroscopy ranged from strong positive correlations to weak positive correlations. Of the studies evaluating blood carotenoids, most studies reported combined total carotenoid concentration when compared to spectroscopy-based skin carotenoids, unless specified. Two of the studies using blood carotenoids as the comparison variable reported very strong correlation coefficients of $r = 0.81$; $p < 0.001$[62, 64] and four of the studies found strong correlation coefficients of $r = 0.78$; $p < 0.001$,[40] $r = 0.72$; $p < 0.001$,[15] $r = 0.72$; $p < 0.01$[61] and a linear regression correlation $R = 0.75$.[51] Four of the studies found moderate correlations of $r = 0.63$; $p < 0.001$ for mothers (and a low correlation with infants $r = 0.39$; $p = 0.02$),[41] $r = 0.47$; $p = 0.001$,[39] $r = 0.44$; $p = 0.01$,[50] and pre ($r=0.450$, p

<0.0001) to post (r=0.56; p <0.0001), specifically against lycopene.[63] Only one study reported a relatively weak correlation of r = 0.27; p <0.05[28] when comparing the CD700M spectrophotometer to plasma carotenoids. There were no discernible differences in correlation coefficient strength between plasma and serum samples.

Spectroscopy and Dietary Intake

Of the included studies, n=12 articles correlated spectroscopy to dietary intake data using various subjective dietary collection methods (**Table 4**). Food Frequency Questionnaires were used in n=7,[22, 24, 29, 30, 52, 54, 57] FFQ in conjunction with the Automated Self-Administered Dietary Assessment Tool (ASA-24®) was used in n=1,[55] the Australian Eating Score (AES) was used in n=2,[25, 27] the Australian Recommended Food Score (ARFS) and Fruit and Vegetable Variety Index (FAVVA) were used in n=1,[23] and a USDA resource quantifying fruit and vegetable servings per day was used in n=1.[58] After recording dietary intake data carotenoid concentration was estimated using the USDA National Nutrient Database for Standard Reference,[24, 52, 55, 58, 56] AusFoods and FoodWorks,[25, 27] NutritionQuest,[54, 57] the Fred Hutchinson Cancer Research Center for quantification,[22] the Australian Guide to Healthy Eating and the Australian Dietary Guidelines,[23] the Canadian Nutrient File v2007b[29] or simply by the servings of fruit and vegetables.[30]

The correlation coefficients between self-reported dietary intake and spectroscopy were lower than spectroscopy and serum or plasma carotenoid concentrations. The correlation between spectroscopy and dietary intake of fruits and vegetables were analyzed against multiple variables depending on the study design; dietary intake of fruits and vegetables, total dietary carotenoid intake, or individual dietary carotenoids were used to determine a correlation between spectroscopy-based skin carotenoid measurements and dietary intake as displayed in the

Correlation Outcomes in **Table 4**. In terms of the strength of association between the studies comparing spectroscopy-based skin carotenoid measurements to dietary intake, the studies differed in statistical methodologies, and therefore may not be directly comparable. Studies using Pearson's Correlations found weak to moderate correlation coefficients, with total dietary carotenoids having the strongest correlation ($r = 0.599$; $p < 0.001$[24] and $r = 0.52$; $p = 0.001$[54]) and weaker associations with individual carotenoids, such as lutein ($r = 0.197$; $p = 0.01$) and lycopene ($r = 0.287$; $p = 0.01$).[52] Studies using Spearman's Correlations established weak to low correlations ranging from $\rho = 0.224$; $p = 0.045$[30] to $\rho = 0.47$; $p < 0.001$.[29] Additional methods of linear regression models (β-coefficient $\pm$ SE) were used to determine the relationship between spectroscopy-based skin carotenoid measurements and dietary intake.[22, 25, 27, 57] The studies comparing spectroscopy to dietary intake used RRS[22, 52, 54, 55, 57, 58] or spectrophotometers[23-25, 27, 29, 30] to measure skin carotenoid status or skin yellowness/redness.

Spectroscopy and Both Dietary Intake and Plasma or Serum Carotenoids

Of the included studies, n=6 analyzed both dietary intake and plasma or serum carotenoids to assess the criterion-related validity of spectroscopy (**Table 5**). Of the studies analyzing blood carotenoids in conjunction with dietary intake, a majority of the studies used RRS with the exception of one study using RS.[59] Of the five studies evaluating both blood carotenoids and dietary intake, three studies analyzed plasma carotenoids[26, 53, 59] and three studies evaluated serum carotenoids.[38, 56, 60] All of the studies analyzing blood samples along with dietary intake used HPLC to quantify the carotenoids in the blood except for one study that used the LC-MS extraction method.[59] To record dietary intake, a variety of data collection tools were used, including FFQs,[26, 53, 59] FFQ in conjunction with ASA-24®,[56] multiple day food recall diaries[38] and Fruit and Vegetable Intake Scores.[60] The nutrient analysis of the dietary intake data was

performed using the USDA Food database,[38, 56] the National Cancer Institute (NCI) standard algorithm and the Fred Hutchinson Cancer Research Center for quantification,[59] NutritionQuest,[53] University of Minnesota Nutrition Coding Center Nutrient Data System,[26] or the National Institute of Health (NIH) prescribed algorithm for fruit and vegetable consumption.[60]

The correlation pattern mirrored the corresponding studies that evaluated either blood carotenoids or dietary intake, such that the correlation coefficients comparing total serum or plasma carotenoids were all considered moderate to strong correlations and ranged from $r=0.62$; $p<0.006$[26] to $r=0.78$; $p<0.0001$.[38] Although dietary intake correlation coefficients were lower than blood carotenoids, there were weak to moderate correlations with the skin carotenoids varying from $r=0.38$; $p=0.016$[60] to $r=0.69$; $p<0.0001$.[59]

Discussion

Summary of Results

This systematic review examined current literature that validated spectroscopy against blood, reported dietary intake, or both to investigate whether spectroscopy-based skin carotenoid measurements are an objective, valid biomarker of fruit and vegetable intake. All 29 included studies found statistically significant correlations between skin carotenoids measured via spectroscopy and plasma or serum carotenoids and/or dietary intake. Although the included studies differed in study design, population size, age, and participant demographics, the evidence provided in all 29 studies supports the use of spectroscopy as a proxy for fruit and vegetable intake when compared to blood carotenoids and/or self-reported dietary intake. Overall, the strongest correlations existed between spectroscopy and blood carotenoids; however, data

supports statistically significant associations between spectroscopy and blood carotenoids and/or self-reported dietary intake in all of the included studies.

Although the data support the use of spectroscopy as a non-invasive, objective biomarker of dietary fruit and vegetable intake, additional research is warranted before spectroscopy-based skin carotenoid measurements are considered an equally valid biomarker of fruit and vegetable intake as plasma or serum carotenoids or validated dietary intake tools. Understanding the metabolism, absorption, and storage of carotenoids among all age groups, and under differing genetic and environmental conditions is essential to their accurate detection *in vivo*. Increasing the methodological strength through experimental study designs, such as randomized controlled or crossover trials and dose-response studies are required to understand the efficacy of spectroscopy-based skin carotenoid measurements as an approximation of fruit and vegetable intake in individuals or populations. In addition, expanding future research to encompass more diverse populations will improve the generalizability of this technique and the acceptance of spectroscopy-based skin carotenoid measurements as a predictive biomarker of fruit and vegetable intake.

Differences Between Spectroscopy Devices

The three methods of spectroscopy all produced significant correlations between dietary intake and/or blood carotenoids. Of the manuscripts included in this review, a majority used RRS to quantify skin carotenoids. Raman Resonance spectroscopy has previously demonstrated increased accuracy and precision in detecting skin carotenoids in comparison to RS and spectrophotometers; however, RS produced moderate to strong correlations, while spectrophotometers produced weak to moderate correlations. Previous research between the efficacy of spectroscopy devices found spectrophotometer devices to be more prone to error and

chromophore interference compared to RRS;[44] to date, no such research has compared RS to spectrophotometry. However, as reported in **Tables 3, 4, 5**, there were no observable differences in the correlational strength of the relationships between method of detection and blood or dietary intake. It is important to continue future research on the sensitivity and specificity of the three methods to assess fruit and vegetable intake.

The three spectroscopy devices explored in this review have limitations that may determine what device is most appropriate for specific study purposes. Resonance Raman spectroscopy technically has the capacity to detect different carotenoid molecules based on the absorption detection spectrum; however, RRS is unable to produce individualized scores for each carotenoid compound.[67] Different wavelengths have varying affinities depending on the carotenoid compound; for example, at 514.5nm, lycopene exerts an excitation signal six times that of β-carotene.[68] Therefore, to examine individual carotenoid molecules, the excitation wavelength must be predetermined depending on the length of the conjugated carbon backbone.[68] Thus, the individual carotenoid isomers are measured collectively with RRS, to avoid constantly recalibrating the wavelength of the device. Reflection spectroscopy is dependent on the skin matrix and the potentially confounding chromophores, such as melanin and hemoglobin, that may affect the RS measurement.[14] Reflection spectroscopy is unable to differentiate between the carotenoid isomers due to a more simplified spectral detection methodology, and therefore presents only total dermal carotenoids as the output.[14] Spectrophotometers measure skin carotenoids through the dermatological pigmentation of the skin, and therefore are limited by the concentration of skin pigment interference. For accurate evaluation of skin carotenoids using a spectrophotometer, the participants must have a relatively fair complexion in order for the device to measure the carotenoid compounds within the color

spectrum.[69] These limitations should be considered when selecting a device for spectroscopy-based skin carotenoid measurements, acknowledging that many factors, including participant demographics and environmental conditions, may contribute inaccuracies to spectroscopic detection.

Spectroscopy and Blood Carotenoids

Blood carotenoid concentrations were positively associated with skin carotenoid status measured via spectroscopy. Carotenoids are detectable in plasma or serum for approximately 2 weeks following initial consumption of carotenoid-containing foods.[15] In comparison, the deposition of carotenoids into adipose cells increases the longevity of carotenoids in the skin to approximately 4 weeks after dietary intake.[15, 53] Carotenoids in the plasma or serum may be analyzed as total combined carotenoids or individual carotenoid compounds, while individual carotenoids in the skin cannot be easily detected. As previously confirmed by HPLC analysis of skin tissue biopsies, human skin is relatively enriched in β-carotene and lycopene compared to other carotenoids, and are found in increased concentrations in the blood, indicating that spectroscopy may be more sensitive to sources of these carotenoids than is blood.[68, 70] Contrarily, carotenoid compounds such as lutein and zeaxanthin are more concentrated in the macula of the eye, and therefore are not often associated with skin carotenoid concentrations.[20] Among studies that assessed individual carotenoids in plasma or serum, the reported data confirmed stronger correlations between skin carotenoid scores and blood-derived α-carotene and β-carotene.[60] Further research into the relationship of plasma or serum carotenoids and skin carotenoids to assess fruit and vegetable intake and the types of foods that are reflected in the skin is warranted.

Spectroscopy and Dietary Carotenoids

Studies comparing spectroscopy to dietary intake found positive and statistically significant associations between skin carotenoids and fruit and vegetable consumption or dietary carotenoids. Among the studies that used self-reporting dietary recalls, reporting bias was a major critique in recall accuracy and was mentioned as a potential limitation in multiple studies.[54, 57] The variety of databases used to analyze dietary intake may contribute to inconsistencies in nutrient composition of food items, as does the ability of food composition databases to reflect actual carotenoid content of foods, consequently affecting carotenoid estimation. For instance, processing and holding of fruits and vegetables affects the carotenoid content of that food source. Thus, the use of an objective spectroscopy-based skin carotenoid assessment is very appealing due to the decreased likelihood of subjective biases and lack of reliance on nutrient databases. Additional cross-validation studies of skin carotenoids compared to various measures of subjective recall, particularly if skin carotenoid status can be used as a covariate to strengthen dietary intake analysis, would be valuable to the study of dietary intake of fruits and vegetables.

Spectroscopy in Diverse Populations

Despite the studies in this review generating statistically significant correlations between spectroscopy, blood carotenoids, and/or reported dietary intake, it is imperative to acknowledge potential confounding variables, such as age, sex, BMI, and race/ethnicity.[71] The papers analyzed in this review indicated that spectroscopy may be an effective measure of carotenoid status in most ages, including infants, children, and adults. Assessing infant carotenoid status using spectroscopy is challenging due to the thin, delicate skin of newborns and infants, resulting in subdermal laser penetration beyond the epidermis, reducing the accuracy of carotenoid detection.

However, RRS scores in infants were strongly correlated with serum carotenoids (R = 0.75) in healthy infants and relatively weak to moderate correlations in premature infants (r = 0.44; p = 0.01 and r = 0.52; p = 0.01), respectively.[50, 51] Scarmo et al. detected high skin carotenoid scores along with a positive association between age and skin carotenoid status in a large population of preschool-aged children.[57] The age of participants is also considered as a potential confounding variable due to the lack of knowledge regarding carotenoid metabolism and aging.[53, 71, 72] In addition to the limited understanding of carotenoid metabolism, few studies addressed the physiological changes that accompany aging, which may result in difficulties detecting carotenoids using spectroscopy.[26, 27] Although only one of the studies was conducted primarily in older adults,[39] Bernstein et al. found a positive and significant correlation between serum carotenoid and spectroscopy-based skin carotenoids measurements. Mayne et al. included the lack of older adults as a study limitation and acknowledged the potential differences with skin quality, skin thinning due to collagen loss, and decreased energy intake that may affect the accuracy of spectroscopy in this population.[26] Although the included studies reflect the ages across the lifespan, the sample sizes in some of the studies are relatively small, and therefore may not be generalizable to the broader population in that specific age range.

Studies controlling for individual variability resulted in differences in carotenoid status based on BMI classification.[25, 27, 53-59, 63] Nguyen et al. found incongruencies between reported dietary intake, skin carotenoids, and plasma carotenoids and attributed this to the increased BMI within a subgroup of participants due to the storage of circulating carotenoids into adipose cells.[53] To limit the effect of BMI potentially altering skin carotenoid status, some studies controlled for weight by only including non-obese, adult participants with a BMI of <30kg/m^2.[15, 24, 60] Additional methods of stratifying analyses by weight or BMI percentile were used to minimize

the potential effect of adiposity on skin carotenoid detection.[51, 53, 55-57] It should be noted that studies using BMI percentiles for stratifying data analyses were conducted in study populations primarily consisting of child participants, and therefore extrapolating these results to adult populations may result in inaccurate assumptions.

Race and ethnicity may impact skin carotenoid measurements due to the interference of confounding compounds, such as melanin.[14, 23, 24, 51] Melanin is detected within a similar absorption spectrum as carotenoids, ranging from 360-560nm.[73] To minimize the effect of skin pigmentation, many studies used the palm or the heel to measure skin carotenoids, as there is minimal melanin interference and an increased thickness of the stratum corneum to prevent the laser from penetrating beyond the storage location of carotenoids.[26, 51, 53, 57, 62] Reflection spectroscopy accounts for the potential melanin obstruction through an automatic de-convolution algorithm to correct for residual melanin and other biochrome compounds that may interfere in the tissue site.[74] Therefore, RS has a lower specificity for the exclusive detection of carotenoid molecules due to the potential error of this algorithmic computation.[14] To the authors' knowledge, there have been no studies evaluating the algorithmic correction in individuals with high melanin concentrations, though it may be assumed that larger margin of error is associated with higher skin chromophores. With regards to RRS, it has been observed that melanin interference may be easily corrected by spectrophotometrically measuring the melanin content of the skin and correcting for individual differences in skin pigmentation; however, these methods for correcting for melanin interference prompt RRS to underestimate skin carotenoids whereas RS overestimates skin carotenoid status.[14, 62, 75, 76] Ermakov et al. investigated the effects of melanin interference in carotenoid detection using both RS and RRS and found that both methods had very low correlation coefficients when compared to melanin indices, indicating no

significant association between melanin and RS or RRS.[62] Spectrophotometers specifically measure the melanin in the skin and provide a melanin index, which may be used to adjust for differences in melanin concentration.[62, 77] However, all seven of the studies that used spectrophotometers included in this review adjusted for potential melanin interference by selecting predominately homogeneous Caucasian sample populations.[23-25, 27-30]

Genetic factors may also influence carotenoid metabolism and detection via spectroscopy.[78] Jilcott Pitts et al. attempted to determine the effectiveness of spectroscopy to measure skin carotenoids in a diverse population and found that the association between self-reported total fruit and vegetable intake and RS-assessed skin carotenoids was not significant among African-American participants, although the association was significant in a Caucasian subsample within the same study.[59] As discussed above, melanin may not be a significant confounder, suggesting a potential genetic difference between race/ethnicities.[59, 78] There are many regulatory proteins involved in the uptake, transport, and cleavage of carotenoids that may be susceptible to genetic modifications by protein transcription.[78] These modifications, including single nucleotide polymorphisms (SNPs), may affect the utilization or storage of carotenoids, thus resulting in the inaccurate reflection of dietary intake of fruits and vegetables.[78, 79] It has been demonstrated through genomic sequencing that different ethnic groups display varying efficiencies of carotenoid metabolism, and that ethnic origin should be considered as a covariate when assessing skin carotenoids using spectroscopy.[79]

Spectroscopy and Seasonality

Seasonality may affect skin carotenoid concentrations by either reflecting differences in dietary intake of carotenoid-rich foods or skin carotenoid oxidation by UV exposure.[22] Dietary data collected by Beccarelli et al. showed seasonal variations due to increased consumption of

carotenoid-rich autumn vegetables, such as sweet potatoes, compared to springtime vegetables lower in carotenoids, such as cucumbers.[54] Other researchers have controlled for potential seasonal effects of sun exposure on skin carotenoid status by conducting studies during only one season.[56] Mayne et al. evaluated skin carotenoid scores over a six-month period in a climate with notable seasonal differences and found no differences by season, with intraclass correlation coefficients over the six time points that ranged from 0.85-0.89.[26] In agreement with Mayne et al., a one-year study by Jahns et al. found no differences in skin carotenoid scores based on season; however, blood carotenoids were lower in the summer, lending credence to the potential of seasonality to affect skin carotenoid scores.[80] Additional studies are warranted to determine the effect of seasonality on skin carotenoid status, and researchers should collect data on season of measurement to test as a potential confounder in statistical models.

Spectroscopy in Non-Clinical Settings

A sub-aim of multiple studies was to determine the feasibility of using spectroscopy in atypical, non-clinical settings. Community environments, such as day-care centers,[62] elementary schools,[53, 54] small food (corner) stores,[59] and outdoor community parks[62] were all locations assessed for field feasibility in both child and adult participants. Jilcott Pitts et al. and Scarmo et al. reported the average time it took to complete triplicate measurements and found on average it took approximately 94 seconds per participant to record triplicate measures using RS[59] or an average of 30 seconds per measurement using RRS.[57]

Conclusions

The reviewed literature suggests that all three spectroscopy methods are valid tools for quantifying skin carotenoids as an approximation of fruit and vegetable intake. The data collected from spectroscopy-based skin carotenoid measurements were positively and

significantly correlated to blood carotenoids and/or reported dietary intake, supporting the use of spectroscopy as a valid biomarker of dietary intake of fruit and vegetable intake.

Application of the Findings

The data provided in this review support the use of spectroscopy as a reflective measure of fruit and vegetable intake in diverse ages and racial/ethnic groups; however, more research is required for these results to be extrapolated to the general population. Many of the studies included in this review conducted on-site data collection in various community settings as spectroscopy provides a rapid and painless measure of dietary intake in a matter of seconds. This technology has the potential to enhance the health field by providing information on dietary patterns and tracking dietary behaviors to support preventative health services.[26, 51, 61, 63] The consistent monitoring of carotenoid status may increase early detection or track the progression of various chronic diseases;[51, 52, 61, 63, 81] however, current methods of spectroscopy are unable to diagnose acute or chronic diseases exclusively based on carotenoid status.

As a result of the successful implementation of spectroscopy techniques in the community setting, this method may be used to assess the outcomes of nutrition intervention programs in large, diverse populations.[82] Spectroscopy has the capability of providing an objective reflection of fruit and vegetable intake in children in the school setting.[53, 54] This rapid and quantitative assessment of skin carotenoids may be an impactful method for evaluating nutrition-based interventions as the need for effective strategies to support obesity and chronic disease prevention in both children and adults are public health priorities.[82]

Limitations of the Review

It is imperative to acknowledge that the current review presents several limitations. Although the findings were established in diverse populations in different ages and ethnic groups, a majority

of the studies were cross-sectional, or prospective cohort study designs, and therefore do not provide the degree of evidence to prove causation that result from randomized controlled trials. In addition, non-whole food supplementation was used as an exclusion criterion in order to complete the objective of this review – that spectroscopy is a valid biomarker of fruit and vegetable intake. High-dose supplementation likely results in substantial increases in both spectroscopy-based skin carotenoid measurements and plasma or serum carotenoids; however, this is not indicative of normal dietary intake as would be seen in nutrition surveillance or intervention evaluation studies. Thus, we limited our review to papers that did not include high-dose supplementation to investigate the sensitivity of the spectroscopy-based skin measurement devices to detect changes in carotenoid concentrations that may be found naturally in fruit and vegetables. Additionally, the use of dietary supplements were also excluded to account for differences in bioavailability and gastrointestinal absorption compared to dietary consumption of fruits and vegetables.[83] However, due to this exclusion criterion, it was noted that multiple studies were excluded that used natural food concentrates, such as kale extract or high carotenoid additives. Nonetheless, the elevated concentration of carotenoids likely exceeded the typical daily intake and therefore studies using any type of non-whole food supplementation were not included in the review. Finally, it should also be recognized that due to the high volume of potential articles retrieved during the comprehensive literature search, only articles published in English were considered for this review.

This manuscript was published:

Radtke MD, Jilcott Pitts SB, Jahns L, Firnhaber GC, Loofbourrow BM, Zeng A, Scherr RE. Criterion-related Validity of Spectroscopy-based Skin Carotenoid Measurements as a Proxy for Fruit and Vegetable Intake: A Systematic Review. *Advances in Nutrition* 2020; 11(5):1282-1299. doi: https://doi.org/10.1093/advances/nmaa054.

References

1. Slavin JL, Lloyd B. Health benefits of fruits and vegetables. Adv Nutr 2012;3(4):506-16.
2. Liu RH. Health-promoting components of fruits and vegetables in the diet. Adv Nutr 2013;4(3):384-392.
3. Eggersdorfer M, Wyss A. Carotenoids in human nutrition and health. Arch Biochem Biophys 2018;652:18-26.
4. Stahl W, Sies H. Antioxidant activity of carotenoids. Mol Aspects Med 2003;24(6):345-351.
5. Britton G, Liaaen-Jensen S, Pfander H. Carotenoids Handbook Vol. 1. Birkhäuser Verlag, Basel, Boston Berline 2004.
6. Olson JA. Benefits and Liabilities of Vitamin A and Carotenoids. J Nutr 1996;126(4):1208-1212.
7. Rahiman R, Ali MAM, Ab-Rahman MS. Carotenoids Concentration Detection Investigation: A Review of Current Status and Future Trend. Int J Biosci Biochem Bioinforma 2013;3(5):466-472.
8. Yu Z, Kastenmüller G, He Y, Belcredi P, Möller G, Prehn C, Mendes J, Wahl S, Roemisch-Margl W, Ceglarek U, et al., Differences between human plasma and serum metabolite profiles. PLoS One 2011;6(7):e21230.
9. Barua AB, Furr HC. Extraction and Analysis by High-Performance Liquid Chromatography of Carotenoids in Human Serum. Methods Enzymol 1992;213:273-281.
10. Hrvolova B, Martínez-Huélamo M, Colmán-Martínez M, Hurtado-Barroso S, Lamuela-Raventós RM, Kalina J. Development of an Advanced HPLC-MS/MS Method for the

Determination of Carotenoids and Fat-Soluble Vitamins in Human Plasma. Int J Mol Sci 2016;17(10):1719.

11. Baldrick FR, Woodside JV, Elborn JS, Young IS, McKinley MC. Biomarkers of fruit and vegetable intake in human intervention studies: a systematic review. Crit Rev Food Sci Nutr 2011;51(9):795-815.

12. Block G, Norkus E, Hudes M, Mandel S, Helzlsouer K. Which Plasma Antioxidants Are Most Related to Fruit and Vegetable Consumption? Am J Epidemiol 2001;154(12):1113-1118.

13. Michels KB, Welch AA, Luben R, Bingham SA, Day NE. Measurement of Fruit and Vegetable Consumption with Diet Questionnaires and Implications for Analyses and Interpretation. Am J of Epidemiol 2005;161(10):987-994.

14. Ermakov IV, Gellermann W. Dermal carotenoid measurements via pressure mediated reflection spectroscopy. J Biophotonics 2012;5(7):559-70.

15. Jahns L, Johnson LK, Mayne ST, Cartmel B, Picklo MJ Sr, Ermakov IV, Gellermann W, Whigham LD. Skin and plasma carotenoid response to a provided intervention diet high in vegetables and fruit: uptake and depletion kinetics. Am J Clin Nutr 2014;100(3):930-937.

16. Shim JS, Oh K, Kim KC. Dietary assessment methods in epidemiologic studies. Epidemiol Health 2014;36:e2014009.

17. Kroes R, Müller D, Lambe J, Löwik MR, van Klaveren J, Kleiner J, Massey R, Mayer S, Urieta I, Verger P, Visconti A. Assessment of intake from the Diet. Food and Chem Toxicol 2002;40(2-3):327-385.

18. Harnack L, Himes JH, Aniliker J, Clay T, Gittelsohn J, Jobe JB, Ring K, Snyder P, Thompson J, Weber JL. Intervention-related bias in reporting of food intake by fifth-grade children participating in an obesity prevention study. Am J Epidemiol 2004;160(11):1117-1121.

19. Natarajan L, Flatt SW, Sun X, Gamst AC, Major JM, Rock CL, Al-Delaimy W, Thomson CA, Newman VA, Pierce JP, Women's Healthy Eating and Living Study Group. Validity and systematic error in measuring carotenoid consumption with dietary self-report instruments. Am J Epidemiol 2006;163(8): 770-778.

20. Ermakov IV, Sharifzadeh M, Ermakova M, Gellermann W. Resonance Raman detection of carotenoid antioxidants in living human tissue. J Biomed Opt 2005;10(6):064028.

21. Darvin ME, Sterry W, Lademann J, Vergou T. The Role of Carotenoids in Human Skin. Molecules 2011;16(12):10491-10506.

22. Scarmo S, Cartmel B, Lin H, Leffell DJ, Ermakov IV, Gellermann W, Bernstein PS, Mayne ST. Single v. multiple measures of skin carotenoids by resonance Raman spectroscopy as a biomarker of usual carotenoid status. Br J Nutr 2013;110(5):911-917.

23. Ashton LM, Pezdirc KB, Hutchesson MJ, Rollo ME, Collins CE. Is Skin Coloration Measured by Reflectance Spectroscopy Related to Intake of Nutrient-Dense Foods? A Cross-Sectional Evaluation in Australian Young Adults. Nutrients 2018;10(1):11.

24. Bixley GS, Clark KM, James AP. Skin colour predicts fruit and vegetable intake in young Caucasian men: A cross-sectional study. J Nutr Intermed Metab 2018;12:20-27.

25. Coyle DH, Pezdirc K, Hutchesson MJ, Collins CE. Intake of specific types of fruit and vegetables is associated with higher levels of skin yellowness in young women: A cross-sectional study. Nutr Res 2018;56:23-31.

26. Mayne ST, Cartmel B, Scarmo S, Lin H, Leffell DJ, Welch E, Ermakov I, Bhosale P, Berstein PS, Gellermann W. Noninvasive assessment of dermal carotenoids as a biomarker of fruit and vegetable intake. Am J Clin Nutr 2010;92(4):794-800.

27. Pezdirc K, Hutchesson MJ, Whitehead R, Ozakinci G, Perrett D, Collins CE. Fruit, Vegetable and Dietary Carotenoid Intakes Explain Variation in Skin-Color in Young Caucasian Women: A Cross-Sectional Study. Nutrients 2015;7(7):5800-5815.

28. Pezdirc K, Hutchesson MJ, Williams RL, Rollo ME, Burrows TL, Wood LG, Oldmeadow C, Collins CE. Consuming High-Carotenoid Fruit and Vegetables Influences Skin Yellowness and Plasma Carotenoids in Young Women: A Single-Blind Randomized Crossover Trial. J Acad Nutr Diet 2016;116(8):1257-65.

29. Stephen ID, Coetzee V, Perrett DI. Carotenoid and melanin pigment coloration affect perceived human health. Evol Hum Behav 2011;32(3):216-227.

30. Whitehead RD, Re D, Xiao D, Ozakinci G, Perrett DI. You are what you eat: within-subject increases in fruit and vegetable consumption confer beneficial skin-color changes. PLoS One 2012;7(3):e32988.

31. Ermakov IV, Ermakova MR, McClane RW, Gellermann W. Resonance Raman detection of carotenoid antioxidants in living human tissues. Opt Lett 2001;26(15):1179-1181.

32. Hata TR, Scholz TA, Ermakov IV, McClane RW, Khachik F, Gellermann W, Pershing LK. Non-invasive raman spectroscopic detection of carotenoids in human skin. J Invest Dermatol 2000;115(3):441-448.

33. Meléndez-Martinez AJ, Mapelli-Brahm P, Benítez-González A, Stinco CM. A comprehensive review on the colorless carotenoids phytoene and phytofluene. Arch Biochem Biophys 2015;572:188-200.

34. Wang Y, Luo MR, Wang M, Xiao K, Pointer M. Spectrophotometric measurement of human skin colour. Color Res Appl 2017;42(6):764-774.

35. Wang M, Xiao K, Luo MR, Pointer M, Cheung V, Wuerger S. An investigation into the variability of skin colour measurements. Color Res Appl 2018;43(4):458-470.

36. Liberati A, Altman DG, Tetzlaff J, Mulrow C, Gøtzsche PC, Ioannidis JPA, Clarke M, Devereaux PJ, Kleijnen J, Moher D. The PRISMA statement for reporting systematic reviews and meta-analyses of studies that evaluate health care interventions: explanation and elaboration. J Clin Epidemiol 2009;62(10):e1-34.

37. Ouzzani M, Hammady H, Fedorowicz Z, Elmagarmid A. Rayyan-a web and mobile app for systematic reviews. Syst Rev 2016;5(1):210.

38. Bernstein PS, Sharifzadeh M, Liu A, Ermakov IV, Nelson K, Sheng X, Panish C, Carlstrom B, Hoffman RO, Gellermann W. Blue-Light Reflectance imaging of Macular Pigment in Infants and Children. Invest Opthalmol Vis Sci 2013;54(6):4034-4040.

39. Bernstein PS, Ahmed F, Liu A, Allman S, Sheng X, Sharifzadeh M, Ermakov IV, Gellermann W. Macular pigment imaging in AREDS2 participants: an ancillary study of AREDS2 subjects enrolled at the Moran Eye Center. Invest Ophthalmol Vis Sci 2012;53(10):6178-6186.

40. Gellermann W, Zidichouski JA, Smidt CR, Bernstein, PS. Raman detection of carotenoids in human tissue. In: Packer L, Krämer K, Obermüller-Jervic U, Sies H., editors. Carotenoids and Retinoids: Molecular Aspects and Health Issues. Vol. Chapter 6. AOCS Press; Champaign Illinois 2005:86-114.

41. Henriksen BS, Chan G, Hoffman RO, Sharifzadeh M, Ermakov IV, Gellermann W, Bernstein PS. Interrelationships between maternal carotenoid status and newborn infant

macular pigment optical density and carotenoid status. Invest Ophthalmol Vis Sci 2013;54(8):5568-78.

42. Stahl W, Heinrich U, Jungmann H, von Laar J, Schietzel M, Sies H, Tronnier H. Increased Dermal Carotenoid Levels Assessed by Noninvasive Reflection Spectrophotometry Correlate with Serum Levels in Women Ingesting Betatene. J Nutr 1998;128(5):903-907.

43. Meinke MC, Darvin ME, Vollert H, Lademann J. Bioavailability of natural carotenoids in human skin compared to blood. Eur J Pharm Biopharm 2010;76(2):269-274.

44. Meinke MC, Schanzer S, Lohan SB, Shchatsinin I, Darvin ME, Vollert H, Magnussen B, Köcher W, Helfmann J, Lademann J. Comparison of different cutaneous carotenoid sensors and influence of age, skin type, and kinetic changes subsequent to intake of a vegetable extract. J Biomed Opt 2016;21(10):107002.

45. Blume-Peytavi U, Rolland A, Darvin ME, Constable A, Pineau I, Voit C, Zappel K, Schäfer-Hesterberg G, Meinke M, Clavez RL, Sterry W, Lademann J. Cutaneous lycopene and beta-carotene levels measured by resonance Raman spectroscopy: high reliability and sensitivity to oral lactolycopene deprivation and supplementation. Eur J Pharm Biopharm 2009;73(1):187-194.

46. Meinke MC, Friedrich A, Tscherch K, Haag SF, Darvin ME, Vollert H, Groth N, Lademann J, Rohn S. Influence of dietary carotenoids on radical scavenging capacity of the skin and skin lipids. Eur J Pharm Biopharm 2013;84(2):365-373.

47. von Elm E, Altman DG, Egger M, Pocock SJ, Gøtzsche PC, Vandenbroucke JP; STROBE Initiative. The Strengthening the Reporting of Observational Studies in

Epidemiology (STROBE) statement: guidelines for reporting observational studies. J Clin Epidemiol 2008;61(4):344-349.

48. National Heart, Lung, and Blood Institute. Development and use of quality assessment tools. Available at: nhlbi.nih.gov/health-pro/guidelines/in-develop/cardiovascular-risk-reduction/tools.

49. Mukaka MM. A guide to appropriate use of Correlation coefficient in medical research. Malawi Med J 2012;24(3):69-71.

50. Chan GM, Chan MM, Gellermann W, Ermakov I, Ermakova M, Bhosale P, Bernstein P, Rau C. Resonance Raman spectroscopy and the preterm infant carotenoid status. J Pediatr Gastroenterol Nutr 2013;56(5):556-559.

51. Ermakov IV, Ermakova MR, Berstein PS, Chan GM, Gellermann W. Resonance Raman based skin carotenoid measurements in newborns and infants. J Biophotonics 2013;6(10):793-802.

52. Rerksuppaphol S, Rerksuppaphol L. Carotenoid intake and asthma prevalence in Thai children. Pediatr Rep 2012;4(1):e12.

53. Nguyen LM, Scherr RE, Linnell JD, Ermakov IV, Gellermann W, Jahns L, Keen CL, Miyamoto S, Steinberg FM, Young HM, Zidenberg-Cherr S. Evaluating the relationship between plasma and skin carotenoids and reported dietary intake in elementary school children to assess fruit and vegetable intake. Arch Biochem Biophys 2015;572:73-80.

54. Beccarelli LM, Scherr RE, Dharmar M, Ermakov IV, Gellermann W, Jahns L, Linnell JD, Keen CL, Steinberg FM, Young HM, Zidenberg-Cherr S. Using Skin Carotenoids to Assess Dietary Changes in Students After 1 Academic Year of Participating in the Shaping Healthy Choices Program. J Nutr Educ Behav 2017;49(1):73-78

55. Aguilar SS, Wengreen HJ, Dew J. Skin Carotenoid Response to a High-Carotenoid Juice in Children: A Randomized Clinical Trial. J Acad Nutr Diet 2015;115(11):1771-8.

56. Aguilar SS, Wengreen HJ, Lefevre M, Madden GJ, Gast J. Skin carotenoids: a biomarker of fruit and vegetable intake in children. J Acad Nutr Diet 2014;114(8):1174-80.

57. Scarmo S, Henebery K, Peracchio H, Cartmel B, Lin H, Ermakov IV, Gellermann W, Berstein PS, Duffy VB, Mayne ST. Skin carotenoid status measured by resonance Raman spectroscopy as a biomarker of fruit and vegetable intake in preschool children. Eur J Clin Nutr 2012;66(5):555-60.

58. Rerksuppaphol S, Rerksuppaphol L. Effect of Fruit and Vegetable Intake on Skin Carotenoid Detected by Non-Invasive Raman Spectroscopy. J Med Assoc Thai 2006;89(8):1206-1212.

59. Jilcott Pitts SB, Jahns L, Wu Q, Moran NE, Bell RA, Truesdale KP, Laska MN. A non-invasive assessment of skin carotenoid status through reflection spectroscopy is a feasible, reliable and potentially valid measure of fruit and vegetable consumption in a diverse community sample. Public Health Nutr 2018;21(9):1664-1670.

60. Van Rensburg JA, Wenhold F. Validity and Reliability of Field Resonance Raman Spectroscopy for Assessing Carotenoid Status. J Nutr Sci Vitaminol 2016;62(5):317-321.

61. Conrady CD, Bell JP, Besch BM, Gorusupudi A, Farnsworth K, Ermakov I, Sharifzadeh M, Ermakova M, Gellermann W, Berstain PS. Correlations Between Macular, Skin, and Serum Carotenoids. Invest Ophthalmol Vis Sci 2017;58(9):3616-3627.

62. Ermakov IV, Ermakova M, Sharifzadeh M, Gorusupudi A, Farnsworth K, Berstein PS, Stookey J, Evans J, Arana T, Tao-Lew L, et al., Optical assessment of skin carotenoid

status as a biomarker of vegetable and fruit intake. Arch Biochem Biophys 2018;646:46-54.

63. Perrone A, Pintaudi AM, Traina A, Carruba G, Attanzio A, Gentile C, Tesoriere L, Livrea MA. Raman Spectroscopic Measurements of Dermal Carotenoids in Breast Cancer Operated Patients Provide Evidence for the Positive Impact of a Dietary Regimen Rich in Fruit and Vegetables on Body Oxidative Stress and BC Prognostic Anthropometric Parameters: A Five-Year Study. Oxid Med Cell Longev 2016;2016:2727403.

64. Zidichouski JA, Mastaloudis A, Poole SJ, Reading JC, Smidt CR. Clinical validation of a noninvasive, Raman spectroscopic method to assess carotenoid nutritional status in humans. J Am Coll Nutr 2009;28(6):687-93.

65. Gellermann W, Ermakov IV, Ermakova MR, McClane RW. Noninvasive laser Raman detection of carotenoid antioxidants in living human skin. Lasers in Surgery: Advanced Characterization, Therapeutics, and Systems XI 2001;4244.

66. United States Department of Agriculture, Agricultural Research Service. Nutrient Data Laboratory. USDA National Nutrient Database for Standard Reference. Available at: http://www.ars.usda.gov/nea/bhnrc/mafcl.

67. Morgan EH, Graham ML, Marshall GA, Hanson KL, Seguin-Fowler RA. Serum carotenoids are strongly associated with dermal carotenoids but not self-reported fruit and vegetable intake among overweight and obese women. Int J Behav Nutr Phys Act 2019;16(1):104.

68. Ermakov IV, Ermakova MR, Gellermann W, Lademann J. Noninvasive selective detection of lycopene and beta-carotene in human skin using Raman spectroscopy. J Biomed Opt 2004;9(2):332-8.

69. Weatherall IL, Coombs BD. Skin color measurements in terms of CIELAB color space values. J Invest Dermatol 1992;99(4):468-73.

70. Scarmo S, Cartmel B, Lin H, Leffell DJ, Welch E, Bhosale P, Bernstein PS, Mayne ST. Significant correlations of dermal total carotenoids and dermal lycopene with their respective plasma levels in healthy adults. Arch Biochem Biophys 2010;504(1):34-9.

71. Moran NE, Mohn ES, Hason N, Erdman JW Jr, Johnson EJ. Intrinsic and Extrinsic Factors Impacting Absorption, Metabolism, and Health Effects of Dietary Carotenoids. Adv Nutr 2018;9(4):465-492.

72. Eroglu A, Harrison EH. Carotenoid metabolism in mammals, including man: formation, occurrence, and function of apocarotenoids. J Lipid Res 2013;54(7):1719-30.

73. Gallas JM, Eisner M. Fluorescence of Melanin-Dependence Upon Excitation Wavelength and Concentration. Photochem Photobiol 1987;45(5):595-600.

74. Longevity Link Corporation. The VEGGIE METER - skin carotenoids as indicator of fruit and vegetable intake and biomarker associated with better health, 2016. Available at: longevitylinkcorporation.com/home.html

75. Darvin ME, Sandhagen C, Koecher W, Sterry W, Lademann J, Meinke MC. Comparison of two methods for noninvasive determination of carotenoids in human and animal skin: Raman spectroscopy versus reflection spectroscopy. J Biophotonics 2012;5(7):550-8.

76. Mayne ST, Cartmel B, Scarmo S, Jahns L, Ermakov IV, Gellermann W. Resonance Raman spectroscopic evaluation of skin carotenoids as a biomarker of carotenoid status for human studies. Arch Biochem Biophys 2013;539(2):163-70.

77. Dwyer T, Muller HK, Blizzard L, Ashbolt R, Phillips G. The Use of Spectroscopy to Estimate Melanin Density in Caucasians. Cancer Epidemiol Biomarkers Prev 1998;7(3):203-206.

78. Borel P. Genetic variations involved in interindividual variability in carotenoid status. Mol Nutr Food Res 2012;56(2):228-40.

79. Lietz G, Oxley A, Leung W, Hesketh J. Single nucleotide polymorphisms upstream from the beta-carotene 15,15'-monoxygenase gene influence provitamin A conversion efficiency in female volunteers. J Nutr 2012;142(1):161S-5S.

80. Jahns L, Johnson LK, Conrad Z, Bukowski M, Raatz SK, Jilcott Pitts, S, Wang Y, Ermakov IV, Gellermann W. Concurrent validity of skin carotenoid status as a concentration biomarker of vegetable and fruit intake compared to multiple 24-h recalls and plasma carotenoid concentrations across one year: a cohort study. Nutr J 2019;18(1):78.

81. Guimarães H, Guedes MB, Rocha G, Tomé T, Albino-Teixeira A. Vitamin A in Prevention of Bronchopulmonary Dysplasia. Curr Pharm Des 2012;18(21):3101-3113.

82. Scherr RE, Laugero KD, Graham DJ, Cunningham BT, Jahns L, Lora KR, Reicks M, Mobley AR. Innovative Techniques for Evaluating Behavioral Nutrition Interventions. Adv Nutr 2017;8(1):113-125.

83. Khachik F. Distribution and metabolism of dietary carotenoids in humans as a criterion for development of nutritional supplements. Pure Appl Chem 2006;78(8):1551-1557.

Tables and Figures

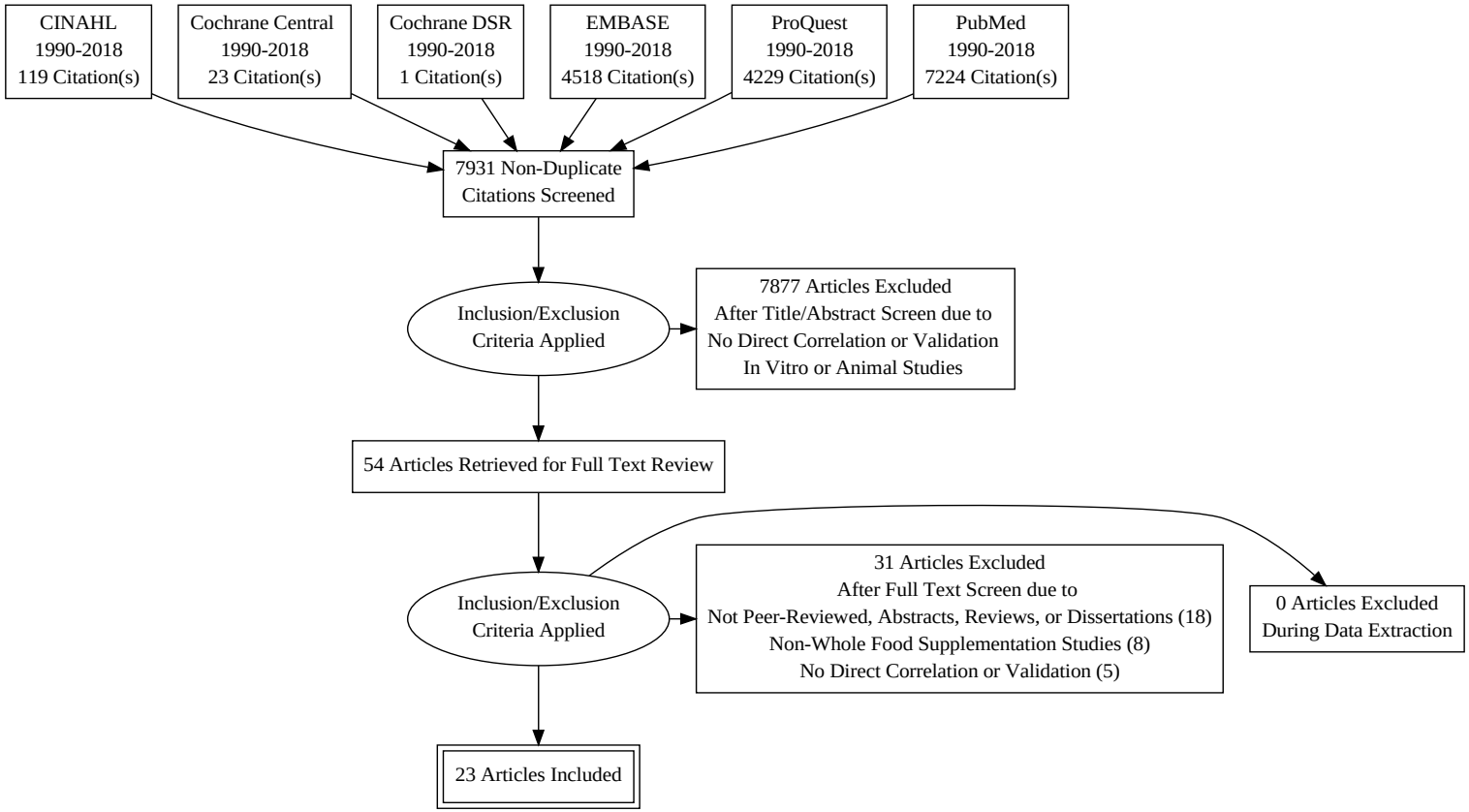


Figure 1: PRISMA flow diagram of detailed search strategy and article selection process

Table 1: Search terms and predefined exclusion criteria applied to article selection process

Search Terms	***Exclusion Criteria***
Spectroscopy Primary search terms: spectroscopy, spectrum analysis, Veggie Meter®, skin reflectance Expanded search terms: BioPhotonic scanner, carotenoid sensor(s), optical assessment, optical detection, Raman microscopy, reflectance spectrophotometer, spectroscopic method(s), spectrophotometry	1. No direct correlation or validation against dietary intake or serum or plasma carotenoids
	2. Any dietary intervention using non-whole food supplementation (including fruit or vegetable extracts) - this is due to differences in dose-response and unrealistic carotenoid concentrations not found in normal dietary amounts.
Carotenoids Primary search terms: carotenoids, α-carotene, β-carotene Expanded search terms: astacene, β-cryptoxanthin, canthaxathin, fucoxanthin, lutein, lycopene, zeaxanthin	3. Not a peer-reviewed publication, abstract only, review articles, or dissertations
	4. Non-human subjects (including *in vitro* studies using human cell lines)
	5. Non-validated methods of recording dietary intake
	6. Review articles

Table 2: Brief overview of spectroscopy-based skin carotenoid devices

Spectroscopy method	Type of spectroscopy device used	Number of validation studies meeting the eligibility criteria of review	Number of validation studies with multiple racial/ethnic groups included	Number of validation studies with infants and/or children	Number of validation studies using plasma or serum carotenoids	Number of validation studies using dietary intake	Number of validation studies using both plasma or serum carotenoids and dietary intake
Resonance Raman Spectroscopy (RRS)	NuSkin BioPhotonic Scanner®	7[52, 55, 56, 58, 60, 63, 64]	2[55, 56]	3[52, 55, 56]	2[63, 64]	3[52, 55, 58]	2[56, 60]
	Custom-built Scanner	13[15, 22, 26, 38, 39, 40, 41, 50, 51, 53, 54, 57, 61]	4 [38, 54, 57, 61]	6[38, 41, 51, 53, 54 57]	7[15, 39, 40, 41, 50, 51, 61]	3[22, 54, 57]	3[26, 38, 53]
Pressure-Mediated Reflection Spectroscopy (RS)	Veggie-Meter®	2[59, 62]	1[59]	0	1[62]	0	1[59]
Spectrophotometer	CM700D Konica Minolta	4[23, 25, 27, 28]	0	0	1[28]	3[23, 25, 27]	0
	Spectro-Guide 450 Gloss 6801	1[24]	0	0	0	1[24]	0
	CM2600D Konica Minolta	2[29, 30]	0	0	0	2[29, 30]	0

Table 3: Summary of studies evaluating the relationship between spectroscopy-based skin carotenoid measurements and serum or plasma carotenoids in diverse populations (n=11)[1]

Reference	Study Design	Population Characteristics (n, sex, age)	BMI and Race/Ethnicity	Type of Spectroscopy	Criterion Measure	Statistical Test	Correlation Outcomes	Quality Assessment
Bernstein et al. (2012)[39]	Cross sectional	n=53 24M, 29F 77.4 ± 7.7 yrs	N/A	RRS – custom built scanner	Serum; HPLC	Pearson's Correlations	SCS and total serum carotenoids: r = 0.47; p = 0.001 SCS and (lutein + zeaxanthin): r = 0.18; p = 0.226	Good
Chan et al. (2014)[50]	Prospective Cohort	n=40 Age: <33 wks. gestation	Birth weight 500-1500 grams	RRS – custom built scanner	Serum; HPLC	Pearson's Correlations	SCS and total serum carotenoids: r = 0.44; p = 0.01	Good
Conrady et al. (2017)[61]	Cross Sectional	n=88 39 M, 49 F 59 ± 17 yrs	Caucasian (n=74) African (n=1) Asian (n=1) Hispanic (n=1) Multinational (n=1) Not recorded (n=10)	RRS – custom built scanner	Serum; HPLC	Linear Regression (exact method not stated)	SCS and total serum carotenoids: r = 0.722; p < 0.01 SCS and serum lutein: r = 0.655; p < 0.01 SCS and serum zeaxanthin: r = 0.656; p < 0.01	Good
Ermakov et al. (2013)[51]	Cross Sectional	n=32 Age: 1 day - 6 years	N/A	RRS – custom built scanner	Serum; HPLC	Linear fit with correlation coefficient	R = 0.75 No p-value listed in paper	Good
Ermakov et al. (2018)[62]	Cross Sectional	n=54 24 M, 30 F 54 ± 19yrs	Caucasian (n=53) African American (n=1)	RS – Veggie Meter®	Serum; HPLC	Linear Regression	r = 0.81; p < 0.001	Good

Gellermann et al. (2005)[40]	Cross Sectional	n=104	N/A	RRS – custom built scanner	Serum; HPLC	Pearson's Correlations	SCS and total serum carotenoids: r=0.78; p < 0.001	Good
Henriksen et al. (2013)[41]	Cross Sectional	n=30 Age: 48-72 hours	N/A	RRS – custom built scanner	Serum; HPLC	Pearson's Correlations	SCS and infant total serum carotenoids: r=0.39; p = 0.02 SCS and maternal total serum carotenoids: r=0.63; p < 0.001	Good
Jahns et al. (2014)[15]	Single Arm Experimental	n=29 9 M, 20 F 32.1 ± 2.5 yrs	BMI = 23.6 ± 0.6 Caucasian (n=28) Other (n=1)	RRS – custom built scanner	Plasma; HPLC	Pearson's Correlations	SCS and total plasma carotenoids: r = 0.72; p < 0.001.	Good
Perrone et al. (2016)[63]	Prospective Cohort	n=71 All Female Age: 38-70yrs	BMI = 24.5 ± 3.0	RRS – NuSkin BioPhotonic Scanner®	Plasma; HPLC	Pearson's Correlations	SCS and plasma lycopene: pre (r=0.450, p <0.0001) post (r=0.559; p <0.0001) five-year dietary intervention period.	Good
Pezdirc et al. (2016)[28]	Randomized Crossover Trial	n = 30 All Female 22.0 ± 4.2yrs	BMI = 23.4 ± 9.7 Caucasian (n=25) Asian (n=4) Other (n=1)	CM700D Spectrophotometer (Konica, Minolta)	Plasma; HPLC	Spearman's Correlations	SCS and total plasma carotenoids: r = 0.27; p < 0.05 SCS and plasma α-carotene: r= 0.29; p < 0.05. SCS and plasma β-carotene: r = 0.35; p < 0.001.	Very Good
Zidichouski et al. (2009)[64]	Cross Sectional	n=372 199 M, 173 F 33.4 ±10.0 yrs	N/A	RRS – NuSkin BioPhotonic Scanner®	Serum; HPLC	Pearson's Correlations	SCS and total serum carotenoids: r=0.81; p <0.001)	Very Good

[1]BMI, body mass index; F, female; HPLC, high performance liquid chromatography; M, male; RRS, resonance Raman spectroscopy; RS, reflection spectroscopy; SCS, skin carotenoid score; yrs, years

Table 4: Summary of studies evaluating the relationship between spectroscopy-based skin carotenoid measurements and dietary intake in diverse populations (n=12)[2]

Reference	Study Design	Population Characteristics (n, gender, mean age)	BMI and Race/Ethnicity	Type of Spectroscopy	Criterion Measure	Statistical Test	Correlation Outcomes	Quality Assessment
Aguilar et al. (2015)[55]	Randomized Control Trial	n=58 27 M, 31F 10.8 ± 3.6 yrs	BMI percentiles: < 5 (n = 4) 5th-85th (n = 41) >85th (n = 13) Caucasian (n=46) Hispanic (n=10) Asian (n=1) Polynesian (n=1)	RRS – NuSkin BioPhotonic Scanner®	3 24-hour dietary recalls and FFQ	Multivariate Linear Regression	SCS and FFQ F/V: R^2=0.22 (r =0.47); p < 0.01 SCS and 24HDR F/V: R^2=0.16 (r = 0.40); p < 0.01 SCS and 24HDR total carotenoids: R^2=0.17 (r = 0.41); p < 0.01 SCS and 24HDR lycopene: R^2=0.16 (r = 0.40); p < 0.01 SCS and 24HDR α-carotene: R^2=0.14 (r = 0.37); p < 0.05 SCS and 24HDR β-carotene: R^2=0.14 (r = 0.37); p < 0.01 SCS and 24HDR (lutein + zeaxanthin): R^2=0.13 (r = 0.36); p < 0.01	Very Good
Ashton et al. (2018)[23]	Cross Sectional	n=148 66 M, 82 F 21.7 ± 2.2 yrs	BMI = 23.9 ± 4.1	CM700D Spectrophotometer (Konica, Minolta)	ARFS and FAVVA	Spearman's Correlations	SCS (yellowness) and ARFS total F/V intake: ρ = 0.30; p < 0.001 SCS (yellowness) and FAVVA total F/V	Fair

							intake: ρ= 0.39; p < 0.001	
Beccarelli et al. (2017)[54]	Prospective Cohort	n=30 9 M, 21 F 9.9 ± 0.6 yrs	BMI percentiles: < 5 (n = 0) 5th-85th (n= 13) 85th-95th (n = 12) >95th (n = 5) Non-Hispanic Black (n=3) Asian/Pacific Islander (n=11) Caucasian (n=8) Hispanic/Latino (n=1) Other (n=3) Multiethnic (n=1) Not Reported (n=3)	RRS – custom built scanner	2004 Block Kids FFQ	Pearson's Correlations	SCS and total dietary carotenoids: pre r = 0.46; p = 0.001 post r=0.52; p = 0.001	Good
Bixley et al. (2018)[24]	Cross Sectional	n=30 All Male 21.7 ± 2.6 yrs	BMI = 23.6 ± 3.4 All Caucasian	Spectro-Guide 450 Gloss 6801 spectrometer	FFQ	Pearson's Correlations	SCS (yellowness) and carotenoid intake: r = 0.599; p < 0.001. SCS (yellowness) and F/V intake: r=0.422; p = 0.02	Fair
Coyle et al. (2018)[25]	Cross Sectional	n=118 All Female Age: 24.7	Median BMI = 23.3 All Caucasian	CM700D Spectrophotometer (Konica, Minolta)	AES 2010	Linear Regression (β-coefficient ± SE)	SCS (yellowness) and F/V intake: β= 0.29 ± 0.03; p = 0.0004	Fair
Pezdirc et al. (2015)[27]	Cross Sectional	n=91 All Female Median age: 22.1 yrs	Median BMI = 22.9 All Caucasian	CM700D Spectrophotometer (Konica, Minolta)	AES 2010	Linear Regression (β-coefficient ± SE)	SCS (yellowness) and F/V intake β= 0.80 ± 0.3; p = 0.017	Very Good

Rerksuppaphol et al. (2006)[58]	Cross Sectional	n=29 2 M, 27 F 31.9 ± 8.3 yrs	BMI = 21.2 ± 3.2 All born in Thailand	RRS – NuSkin BioPhotonic Scanner®	USDA servings per day	Univariate Linear Regression	SCS and F/V intake (p = 0.01)*	Fair
Rerksuppaphol et al. (2012)[52]	Cross Sectional	Asthma: n=73 40 M, 33 F 9.2 ±3.4 yrs No Asthma: n = 350 185 M, 165 F 10.3 ± 3.2yrs	Asthma: BMI = 17.9 ± 4.0 No Asthma: BMI =18.0 ± 3.9 All children born in Thailand	RRS – NuSkin BioPhotonic Scanner®	FFQ	Pearson's Correlations	SCS and α-carotene: r = 0.355; p = 0.01. SCS and β-carotene: r = 0.347; p = 0.01. SCS and β-cryptoxanthin: r = 0.418; p = 0.01. SCS and Lycopene: r = 0.287; p = 0.01. SCS and Lutein: r = 0.197; p = 0.01.	Good
Scarmo et al. (2012)[57]	Cross Sectional	n=381 193 M, 188 F 3.80 yrs	BMI percentiles: < 5 (n = 15) 5th-85th (n = 235) 85th-95th (n = 61) >95th (n = 51) Non-Hispanic White (n=22) Non-Hispanic Black (n=98) Hispanic/Latino (n=228) Biracial (n=22) Other (n=11)	RRS – custom built scanner	FFQ	Univariate Linear Regression	SCS and F/V intake: β = 0.87; p = 0.02	Good
Scarmo et al. (2013)[22]	Prospective Cohort	n=74 28M, 46F 36.6 yrs	BMI Percentiles: underweight (n = 4) healthy (n = 45) overweight (n = 20) obese (n = 5) White (n=62)	RRS – custom built scanner	FFQ	Multivariate Linear Regression	SCS and intake of total carotenoids Baseline: β = 0.28; p < 0.01 Over 6 months: β = 0.23; p < 0.01	Good

			Non-white (n=12)					
Stephen et al. (2011)[29]	Cross Sectional	n=82 34M, 48F Ages: 18-26	All Caucasian	CM2600D Spectrophotometer (Konica, Minolta)	FFQ	Spearman's Correlation	SCS (yellowness) and F/V intake: $\rho = 0.45$; $p < 0.001$ SCS (yellowness) and β-carotene: $\rho = 0.47$; $p < 0.001$	Good
Whitehead et al. (2012)[30]	Cross Sectional	n=35 14M, 21F 20.74 yrs	Caucasian (n=34) East Asian (n=1)	CM2600D Spectrophotometer (Konica, Minolta)	FFQ	Spearman's Correlation	SCS (redness) and F/V intake: $\rho = 0.224$; $p = 0.045$ SCS (yellowness) and F/V intake: $\rho = 0.251$; $p=0.038$	Good

*Correlation value not listed in the manuscript, only p-value indicating statistical significance. Author did not respond to inquiry.

[2]24HDR, 24-hour dietary recall; AES, Australian Eating Survey; ARFS, Australian Recommended Food Score; BMI, body mass index; F, female; FAVVA, Fruit and Vegetable Variety Index; FFQ, food frequency questionnaire; F/V, fruit and vegetable; M, male; RRS, resonance Raman spectroscopy; RS, reflection spectroscopy; SCS, skin carotenoid score; USDA, United States Department of Agriculture; yrs, years

Table 5: Summary of studies evaluating the relationship between spectroscopy-based skin carotenoid measurements and both plasma or serum carotenoids and dietary intake in diverse populations (n=6)[3]

Reference	Study Design	Population Characteristics (n, gender, mean age)	BMI and Race/Ethnicity	Type of Spectroscopy	Criterion Measure	Statistical Test	Correlation Outcomes	Quality Assessment
Aguilar et al. (2014)[56]	Cross Sectional	n=45 20 M, 25 F Mean Age = 10.5 yrs	BMI Percentiles: < 5th (n = 4) 5th-85th (n = 34) >85th (n = 20) Caucasian (n=34) Hispanic (n=7) Asian (n = 3) Pacific Islander (n = 1)	RRS – NuSkin BioPhotonic Scanner®	Dietary intake and serum carotenoids; FFQ and ASA-24 and HPLC	Multivariable Linear Regression	SCS and FFQ dietary intake: $R^2 = 0.32$ ($r = 0.57$); $p < 0.001$ SCS and 24HDR dietary intake: $R^2 = 0.31$ ($r = 0.56$); $p < 0.001$ SCS and total serum carotenoids: $R^2 = 0.62$ ($r = 0.79$); $p < 0.001$	Good
Bernstein et al. (2013)[38]	Cross Sectional	n=51 24M, 27F Age: 1 day – 7 years	Caucasian (n=43) Hispanic (n=4) Multi-racial (n= 4)	RRS – custom built scanner	Dietary intake and serum carotenoids; 3-day food diaries and HPLC	Pearson's Correlations	SCS and total dietary intake: $r = 0.40$; $p = 0.046$. SCS and dietary (lutein + zeaxanthin): $r = 0.57$; $p = 0.0032$ SCS and total serum carotenoids: $r = 0.78$; $p < 0.0001$ SCS and serum (lutein + zeaxanthin): $r = 0.50$; $p = 0.0015$	Good

Janse van Rensburg et al. (2016)[60]	Cross Sectional	n=81 19 M, 62 F M: 40.6 ± 12.2 yrs F: 42.8 ± 12.0 yrs	BMI Males = 25 ± 2.2 BMI Females= 23.7 ± 2.7 Caucasian (n=78) Indian (n=1) African (n=2)	RRS – NuSkin BioPhotonic Scanner®	Dietary intake and serum carotenoids; Fruit and Vegetable intake score and HPLC	Pearson's Correlations	SCS and dietary intake: Season one: r = 0.38, p = 0.016 Season two: r = 0.42, p < 0.001 SCS and total serum carotenoids: r = 0.72; p < 0.001 SCS and serum β-carotene: r = 0.78; p < 0.001 SCS and serum lycopene: r = 0.45; p < 0.001 SCS and serum (lutein + zeaxanthin): r = 0.50; p < 0.001	Good
Jilcott Pitts et al. (2018)[59]	Cross sectional	Part 2: n=30 No gender listed 32.9 ± 11.8 yrs	Part 2: BMI 25.1 ± 2.7 African American (n=17) Caucasian (n=13)	RS – Veggie Meter®	Dietary intake and plasma carotenoids; NCI Fruit and Vegetable Screener and FFQ, and LC-MS	Pearson's Correlations	Part 2: SCS and dietary intake: r = 0.69; p < 0.0001. SCS and total plasma carotenoids: r = 0.71; p < 0.0001	Good
Mayne et al. (2010)[26]	Prospective Cohort	n=74 28 M, 46 F Mean Age = 37 yrs	BMI Percentiles: underweight (n = 4) healthy (n = 45) overweight (n = 20) obese (n = 5) White (n=62) Non-white (n=12)	RRS – custom built scanner	Dietary intake and plasma carotenoids; FFQ and HPLC	Pearson's Correlations	SCS and dietary intake: r = 0.52; p < 0.001. SCS and total plasma carotenoids: r = 0.62; p = 0.006	Very Good

Nguyen et al. (2015)[53]	Cross Sectional	RRS and FFQ: n=128 51 M, 77 F 11.10 ± 0.6 yrs Blood, RRS, & FFQ; n=38 11M, 27 F 11.2 ± 0.5 yrs	BMI distribution: RRS and FFQ under (n = 0) normal (n = 65) over (n = 33) obese (n = 29) no data (n= 1) Blood, RRS and FFQ under (n = 0) normal (n = 19) over (n = 9) obese (n = 10)	RRS – custom built scanner	Dietary intake and plasma carotenoids; 2004 Block Kids FFQ and HPLC	Pearson's Correlations	SCS and dietary intake: r = 0.40; p<0.0001 SCS and total plasma carotenoids: r=0.62; p <0.001	Good

[3]24HDR, 24-hour dietary recall; AES, Australian Eating Survey; ARFS, Australian Recommended Food Score; ASA 24, Automated Self-Administers 24-hour Dietary Assessment Tool; BMI, body mass index; F, female; FAVVA, Fruit and Vegetable Variety Index; FFQ, food frequency questionnaire; F/V, fruit and vegetable; HPLC, high performance liquid chromatography; LC-MS, liquid chromatography – mass spectroscopy; M, male; NCI, National Cancer Institute; RRS, resonance Raman spectroscopy; RS, reflection spectroscopy; SCS, skin carotenoid score; USDA, United States Department of Agriculture

CHAPTER 3

Validation of Diet ID™ in Predicting Nutrient Intake Compared to Dietary Recalls, Skin Carotenoid Scores, and Plasma Carotenoids in University Students

Introduction

Collecting accurate information on dietary intake is an essential component of understanding the physiological relationship between food and health.[1, 2] In particular, the habitual consumption of fruits and vegetables is associated with improved biomarkers for health and the reduction of chronic disease risk across the lifespan due to the vitamins, minerals, phytonutrients, fiber, and other bioactive compounds.[3, 4] Commonly used measures of fruit and vegetable intake include 24 h dietary recalls, food frequency questionnaires (FFQs), and food records.[5, 6] These subjective assessment tools often introduce unintended reporting errors or response biases that may impact the accuracy of dietary data.[7] Objective measures may also be implemented to determine nutrient consumption, such as blood or urinary biomarkers, and tissue or dermal biopsies.[6] However, such assessments are inherently resource-intensive and subject to participant and researcher burden.[8] Therefore, innovative techniques for rigorously assessing dietary intake, emphasizing fruit and vegetable consumption, are warranted in the research setting.[9]

Carotenoids are a class of phytochemicals found in many fruits and vegetables and therefore are a useful marker for dietary assessment. Carotenoids are fat-soluble compounds that are transported in lipoproteins, making them detectable and quantifiable in the blood and skin.[10] In addition to identifying dietary carotenoids through traditional dietary assessments, carotenoid levels may also be identified through innovative techniques, such as spectroscopy-based skin carotenoid measurements and technology-based or image-based dietary assessment methods.[11-13] The Veggie Meter® is a device that utilizes pressure-mediated reflection spectroscopy to quantify the density of carotenoids in the skin.[14] Skin carotenoid scores (SCS) are reflective of long-term dietary changes, approximately one month of intake, due to the longer half-life and slower degradation of carotenoids in the skin compared to plasma or serum, which is evident of

approximately two weeks of dietary intake.[15, 16] Technology-based or image-based dietary assessment methods may have the capacity to evaluate both short- and long-term dietary intake of carotenoid compounds.

Photo navigation technology is an emerging approach used to estimate dietary patterns and nutrient intake in the research setting. The transition from static images of dietary intake using cameras or handheld devices to dynamic, real-time image-assisted or image-based dietary technologies provides additional improvements for mitigating common errors and biases in traditional dietary assessments.[17] Validation studies comparing image-based technologies to other forms of dietary assessments, including 24 h dietary recalls, weighed food records, and double-labeled water, found inconsistencies between the methods of reporting dietary intake, further highlighting the need for the development of more accurate and reliable image-based dietary assessment tools.[18] In addition to the limited number of validated image-based dietary assessment techniques, most studies have yet to include micronutrients, phytonutrients, or other bioactive compounds, making it challenging to definitively quantify the prominent components of fruit and vegetable intake using such methods.[18]

Diet ID™ is a novel application that assesses dietary patterns through Diet Quality Photo Navigation (DQPN®), a patented image-based algorithm that provides estimates of nutrient intake, based on a series of food images.[19, 20] Diet ID™ was developed using dietary data extracted from the National Health and Nutrition Examination Survey (NHANES), as well as a comprehensive review of food intake surveys and epidemiological research to determine estimates of dietary patterns, portion sizes, and eating frequencies of adults in the United States (US).[19, 20] Diet ID™ provides nutrient estimations for energy intake, macronutrients, and micronutrients, including phytonutrients and other bioactive compounds, such as carotenoids

based on the NDSR food database. Diet ID™ not only estimates total carotenoid intake but quantifies the nutrient output for the following carotenoid compounds: α-carotene, β-carotene, lycopene, lutein, and zeaxanthin. Research exploring the relationship between individual carotenoid compounds and total carotenoid intake estimated by Diet ID™ with other measures of fruit and vegetable intake has yet to be conducted.

The present analysis aimed to explore the criterion validity of Diet ID™ against other methods of dietary assessments, including plasma carotenoid concentrations, skin carotenoid scores, and 24 h dietary recalls in a population of university students. This validation study was derived from a larger study that seeks to investigate various biomarkers found in blood and skin, and to measure dietary intake through repeated 24 h NDSR recalls and Diet ID™ to determine if food access programs at the University of California, Davis improve biomarkers for health and fruit and vegetable consumption among students who use these services.

Materials and Methods

The protocol and procedures for this study were approved by the University of California, Davis Institutional Review Board. Participants provided informed written consent prior to study commencement (1476178-4).

Study Design

A prospective cohort (n = 42) consisting of college students from the University of California, Davis was recruited in January 2020 to participate in an effectiveness evaluation of campus food access programs. The study timeline was selected to minimize excessive sun exposure, reduce the variation from seasonal, high carotenoid-containing foods, such as squash, tomatoes, and berries,[21, 22] and for winter break to serve as a washout period for students who had used campus

food access programs prior to enrolling in the study. The study duration was conducted in accordance with the 10-week academic quarter (January–March 2020), with the first data collection period occurring during weeks 1–3 and the second data collection period occurring in weeks 8–10 of the term. Specific to the larger evaluation study, an eight-week duration between timepoints was allotted to ensure biomarkers of interest had an adequate acclimation period to respond to changes in dietary intake.

Participants were recruited prospectively through fliers, social media, and other means of communication, such as verbal or email contact. Participants were healthy, biological males and females above the age of 18 currently enrolled as undergraduate or graduate students at the University of California, Davis, and within a BMI range of 18.5–34.9 kg/m^2.[23, 24] Exclusion criteria included smoking or living in a household with an indoor smoker (including cigarettes, electronic cigarettes, vaping, marijuana), consuming edible products containing tetrahydrocannabinol (THC), the psychoactive component in marijuana, and excessive drinking (consuming >5 alcoholic drinks per week), as the metabolism and absorption of carotenoid compounds under these conditions is unknown.[10] Additionally, individuals participating in artificial tanning methods, such as UV light exposure, or consuming oral or topical high-dose Vitamin A medication (i.e., Accutane, retinol cream) were ineligible to participate due to the potential for elevated carotenoid detection in the blood or skin from non-dietary sources.[25] Prospective study subjects completed a short screening by telephone and those who met the inclusion criteria were invited to schedule an in-person study visit. Study visits were conducted at the Ragle Human Nutrition Research Center at the University of California, Davis.

Anthropometric Data

Anthropometric data were collected at each timepoint to capture any changes during the study period. Height and weight were measured twice to ensure values were within 0.3 cm and 0.1 kg, respectively, and the mean value was reported. Height was measured using a stadiometer and weight was measured using a digital scale; subsequently, BMI was calculated as weight in kilograms divided by height in meters squared (kg/m^2). Blood pressure was measured twice with a sphygmomanometer for an average reading, to ensure participants were normotensive.

Sociodemographic Data

Sociodemographic information including age, sex, race/ethnicity, food security status, and physical activity was acquired for inclusion as potential covariates. Participants self-reported use of food access resources. Food security status was measured at both study timepoints using the United States Department of Agriculture (USDA) 10-item Adult Food Security Survey Module.[26] The following classifications were used in accordance with the USDA to indicate food security status over the last 30 days: 0: high food security; 1–2: marginal food security; 3–5: low food security; and 6–10: very low food security.[27]

Dietary Intake Data

Diet ID™

Participants completed the Diet ID™ assessment in person at each clinic visit. As the application is designed to measure habitual dietary patterns over the last 30 days of intake, only one assessment per timepoint was required. Participants received detailed instructions provided by the manufacturer for standardization among users.

Diet ID™ initially provided a set of screening questions to identify select food group consumption, such as a vegan, vegetarian, gluten-free, or alcohol-free dietary patterns. The application then displayed two images containing a variety of food items to identify the general types of foods that may be consumed. As the users selected the food items most similar to those they consume regularly, the algorithm provided more specific images by incorporating varying types of the same foods, such as low-fat versus full-fat dairy products, and asked individuals to choose the food images that may be present in their eating pattern on a day-to-day basis. Once the application identified an individual's typical eating pattern, foods from the final image were quantified for nutrient analysis by the Diet ID™ algorithm in accordance with the Nutrition Data System for Research (NDSR) database (Version 2017). In addition to specific nutrient output, diet quality was computed by Diet ID™ software using criteria from the Healthy Eating Index 2015 (HEI-2015).[28] When participants completed multiple Diet ID™ assessments at the same study visit, the nutrient values from first assessment were used.

NDSR 24 h Dietary Recalls

Three 24 h dietary recalls using NDSR Software (Version 2019) were conducted by phone within one week of each in-person clinic visit, for a total of six recalls per participant. Each recall (n = 252) was unannounced and consisted of two non-consecutive weekdays (n = 180) and one weekend day (n = 72), when possible, to capture potential variations in dietary intake and to minimize observer bias. Participants who did not respond to researcher inquiries over the weekend had all recalls recorded on weekdays to ensure three days of intake were collected within a week of the in-person clinic visit. Dietary recalls were conducted by trained researchers under the guidance of a registered dietitian. Participants were asked to report all intake starting from midnight the previous day, inclusive of food, beverages, and supplements. As quality

control, the supervising registered dietitian compared the intake as entered in the initial "Quick List" to the "Food Record." Due to the racial and ethnic diversity of the sample population, some of the culturally diverse foods consumed were not matched to records in the NDSR database. Missing food items were reviewed independently by two researchers for consistency with other records in the database. Examples of food classified as "missing" from the NDSR database included boba or bubble tea, international snacks (i.e., shrimp chips, fish jerky) and brand-specific items (i.e., Kirkland protein bars, Dave's Killer Bread). Food labels were reviewed for nutrient analysis if no best fit in the NDSR system was identified. Diet quality, measured using the HEI-2015, was calculated based on the nine components of nutrient adequacy and the four components of nutrient moderation from the foods consumed in the NDSR dietary recalls.[29] Total carotenoids were calculated through summation of individual carotenoid output from NDSR in micrograms (mcg).

Skin Carotenoid Scores

Skin carotenoid scores were measured using the Veggie Meter®. The Veggie Meter® is a validated, research-grade instrument that utilizes pressure-mediated reflection spectroscopy to estimate carotenoid concentration in the skin.[11] The protocol for collecting data using the Veggie Meter®, including triplicate measures and the use of the non-dominant ring finger, was followed to ensure that inter- and intra-individual variability, as well as environmental interferences, were minimized.[30]

Plasma Carotenoids

Participants were asked to abstain from food and beverages, excluding water, for a minimum of 10 h prior to the study visit. Blood samples were collected through venipuncture by a trained

phlebotomist at the Ragle Human Nutrition Research Center using EDTA vacutainer blood collection tubes. Whole blood was centrifuged at 1500 rpm for 15 min at 4 °C and the plasma was extracted, aliquoted, and stored at −80 °C prior to carotenoid analysis performed by Eurofins Craft Technologies.

Individual carotenoids were measured by HPLC in plasma using a modification of the procedures described by Craft.[31, 32] Briefly, after thawing, 150 μL aliquots of plasma were diluted with 150 μL of water containing 0.01% ascorbic acid and 0.001% EDTA then deproteinated by vortexing with 300 μL of ethanol containing tocol as an internal standard and butylated hydroxytoluene (250 ppm) as an antioxidant. The samples were extracted by vortex mixing for 2 min with 2 mL of hexane. Samples were centrifuged to separate phases and the upper hexane was transferred to a borosilicate tube. The extraction was repeated. The combined supernatant was evaporated using a centrifugal evaporator. The residue was dissolved with vortex mixing in 30 μL of ethyl acetate then diluted with 100 μL of acetonitrile:isopropanol (9:1) and vortex mixed 15 s prior to placement in the autosampler. A 20 μL volume was injected.

The HPLC system consisted of a Chromeleon data system, a solvent degasser, an autosampler maintaining samples at 20 °C, a Polaris C18 Ether (3 μm, 4.0 mm × 250 mm), a guard column containing similar stationary phase, a column heater at 31 °C, a diode array detector to measure carotenoids at 450 nm, 325 nm, and at 295 nm to measure tocol. The separation was performed isocratically using a mobile phase of 83% acetonitrile/13% dioxane/4% methanol containing 150 mM ammonium acetate and 0.1% triethylamine at a flow rate of 1.0 mL for 21 min. The method is calibrated with neat standards within the physiological range which are assigned concentrations using absorption coefficients ($E1\%_{cm}$) and corrected

for HPLC purity.[33] The calibration method is based on external standards using peak areas and corrected for tocol as the internal standard.

Statistical Analysis

Data were inspected for normality using Shapiro–Wilk test and transformed as necessary. Descriptive data on participant characteristics are expressed as mean ± SD or percentage. Nutrient analysis from each set of three 24 h NDSR dietary recalls were averaged for a single mean output. Healthy Eating Index 2015 scores were computed from NDSR output using SAS code provided by the NDSR manufacturers. The SAS version 9.4 statistical software was used (SAS Institute Inc.).[34] Paired t-tests were used to determine if dietary intake was independent at each of the timepoints. Considering dietary intake was not independent by timepoint, data from both study timepoints were averaged to determine the relationship between Diet ID™ and NDSR 24 h dietary recalls. Pearson's correlations were computed to explore associations between the nutrients estimated by the dietary assessment instruments. Kendall's tau was computed for variables with a non-linear relationship (HEI-2015) and those with distributions that did not conform to normality after transformation (cholesterol, Vitamin B_{12}, α-carotene, β-carotene, and lycopene). Bland–Altman Plot Analysis was also performed to characterize the agreement between Diet ID™ and NDSR.[35] Nutrients of interest for this analysis were selected based on existing literature from dietary intake studies with the objective of comparing nutrient consumption to other biomarkers of dietary intake.[36-39] Also included were nutrients of concern for underconsumption (calcium, potassium, fiber, and vitamin D) as defined by the 2020–2025 USDA Dietary Guidelines for Americans.[40] Linear regression models were used to estimate the association between Diet ID™, skin carotenoid scores, and plasma carotenoids controlling for BMI, as previous research has demonstrated inverse correlations between BMI and carotenoid

concentrations.[41-43] The vce(robust) command was used to obtain the robust estimator of variance in linear regression models that did not conform to assumptions of homoscedasticity. Statistical significance was established at $p < 0.05$. All other statistical analyses were performed using STATA Version 16.[44]

As this criterion-related validation study is a subset from a larger study, the sample size was initially computed a priori with the primary objective of comparing plasma carotenoids to skin carotenoid scores.[12] A post-hoc analysis for a minimal detectable difference was calculated to determine the number of participants needed to compare Diet ID™ against 24 h NDSR dietary recalls based on $\alpha = 0.05$ and 80% power, in which a minimum of 30 participants were required.[36] As Diet ID™ is a novel assessment tool, additional studies comparing NDSR with other innovative dietary assessment methods were used as comparisons to determine the minimal detectable difference, confirming that the number of participants in this analysis surpasses the number of participants in previous studies that were sufficiently powered.[45, 46]

Results

Participant Characteristics

A total of 48 participants completed the baseline visit of the study, with six participants unable or unwilling to complete the second timepoint; therefore, 42 participants completed timepoint two and are included in the present analysis. Baseline characteristics of the study population are presented in Table 1. The cohort was 75% female, with a mean age of 22.09 ± 2.36 years and BMI of 24.58 ± 5.04 kg/m^2. Of the total participants, 40% were categorized as having high food security, 31% had marginal food security, 17% had low food security, and 12% had very low food security. Results from a paired *t*-test found no significant changes in skin carotenoid scores

from timepoint one to timepoint two, with average scores of 322.98 ± 114.42 and 341.35 ± 113.98, respectively ($p = 0.38$). Participants completed Diet ID™ in 3.68 ± 2.04 min.

Diet ID™ and 24 h NDSR Dietary Recalls

The average nutrient intakes from three 24 h NDSR recalls were significantly correlated with the findings from Diet ID™ for nearly all nutrients evaluated (Table 2). Diet quality was assessed in accordance with HEI-2015, using the nutrient criteria for adequacy and moderation from both dietary intake assessment methods. A significant correlation was observed for diet quality using HEI-2015 as estimated by 24 h NDSR dietary recalls and Diet ID™ ($\tau = 0.55$, $p < 0.0001$). Total calories (kcals), protein intake, and carbohydrate intake were significantly correlated between the two instruments ($\rho = 0.36$, $p = 0.02$; $\rho = 0.55$, $p = 0.002$; $\rho = 0.31$, $p = 0.05$ respectively); however, there was not a significant correlation between the two instruments' measurement of fat intake. To further explore the relationship between different nutrient subtypes for carbohydrates and fat intake as estimated by Diet ID™ and 24 h NDSR dietary recalls, dietary fiber and cholesterol were independently assessed. Significant associations were observed in measurements of dietary fiber ($\rho = 0.64$, $p < 0.0001$, as well as cholesterol ($\tau = 0.32$, $p = 0.003$).

Of specific interest to the study was the consumption of Vitamin A, carotenoids, and carotenoid derivatives. There was a significant correlation of both Vitamin A ($\rho = 0.39$, $p = 0.01$) and total dietary carotenoid intake ($\rho = 0.44$, $p = 0.003$) between Diet ID™ and 24 h NDSR dietary recalls (Table 2). Significant associations were observed regarding the intake of individual carotenoids, including β-carotene ($\tau = 0.39$, $p = 0.0003$), zeaxanthin, and lutein ($\rho = 0.58$, $p = 0.0001$), apart from lycopene ($\tau = -0.09$, $p = 0.40$) and α-carotene, which was approaching significance ($\tau = 0.14$, $p = 0.19$). Additionally, calcium, potassium, folate, iron,

sodium, Vitamins B_2, B_3, B_6, C, and E were significantly correlated, with the exception of Vitamins D, B_1, and B_{12}.

Bland–Altman Plots were generated to characterize the agreement between Diet ID™ and 24 h NDSR dietary recalls for all nutrients of interest. For all nutrients of interest, a majority with the data points fell within the 95% CI, with a maximum of three individuals out of the 42 participants in the sample not within the limits of agreement, with the exception of sodium (n = 5) (Supplemental File S1).

Diet ID™, Skin Carotenoid Scores, and Plasma Carotenoids

Diet ID™, skin carotenoid scores, and plasma carotenoids were compared to determine if objective concentration biomarkers of dietary intake were associated with nutrient estimations from Diet ID™ (Table 3). Total carotenoid intake measured by Diet ID™ was significantly correlated with skin carotenoid scores from the Veggie Meter® after controlling for BMI (Adjusted $R^2 = 0.41$, $p < 0.0001$). Significant positive associations were observed between total plasma carotenoids and total carotenoids estimated by Diet ID™, when controlling for BMI (Adjusted $R^2 = 0.37$, $p = 0.0001$). To directly compare the objective measures of dietary intake, skin carotenoid scores and plasma carotenoids were assessed, and a strong positive correlation was observed after controlling for BMI (Adjusted $R^2 = 0.68$; $p < 0.0001$).

Discussion

Diet ID™ was designed to assess dietary patterns and estimate nutrient intake values by means of a unique pattern recognition image-based algorithm to ultimately identify chronic disease risk.[20] This analysis demonstrates that nutrient intake from Diet ID™ was comparable to both short-term nutrient consumption from NDSR dietary recalls and plasma carotenoids, in addition

to more long-term dietary intake determined by skin carotenoid scores. Diet ID™ was effective in estimating diet quality, as well as nutrients and bioactive compounds associated with fruit and vegetable consumption.

Total Calories and Macronutrients

Total calorie intake is an important nutritional marker used to estimate energy balance and is often pertinent in guiding nutrient recommendations and in nutrition research studies assessing weight gain or weight loss.[47] Total calorie, protein, and carbohydrate intake from Diet ID™ was associated with NDSR output. Although the measurement of total fat was not significantly correlated between Diet ID™ and NDSR, dietary fiber and cholesterol were both found to have significant associations between instruments. As measurements of fat intake approaches significance ($\rho = 0.29$, $p = 0.06$), the sample may have been limited in power to detect the criterion validity between devices for this macronutrient. It is important to note that the Diet ID™ software asks participants to report any dietary restrictions prior to the assessment. Thirteen participants indicated that they did not consume one or more of the following: eggs, nuts, dairy, or meat, which may provide insight into the discrepancies in fat consumption, as the images from Diet ID™ may have not accurately captured additional dietary sources of this macronutrient. Research looking at the dietary intake in the college student population has confirmed the challenges in self-reporting macronutrient intake using innovative technology;[48] further exploration into assessing the discordance in consumption is warranted.

Previous research on dietary intake data collection has indicated inconsistencies between subjective reporting of macronutrient consumption compared to objective measures of macronutrient intake.[7, 49, 50] It has been observed that individuals often underestimate the portion sizes of foods containing both protein and fat, as these are often measured in dietary data

collection using weight estimations, which can be challenging to infer.[51] Additional demographic factors have been observed to introduce a higher risk of bias into the reporting of macronutrient intake, with females, individuals who are overweight or obese, individuals of low socioeconomic status, and individuals actively seeking to lose weight often underreporting macronutrients, whereas younger individuals and individuals with lower BMIs overestimating macronutrient consumption.[52, 53] With the racial and ethnic diversity of the study population, including differences in socioeconomic status indicated by food insecurity, BMI, and a majority of the participants being biologically female, the increased likelihood of reporting bias with macronutrient intake may provide further insight to the non-significant finding for fat intake.

Micronutrients and Phytonutrients

Measurements of Vitamin A, Vitamin C, Vitamin E, Vitamin B_2 (riboflavin), Vitamin B_3 (niacin), Vitamin B_6 (pyridoxine), folate, iron, sodium, potassium, carotenoids, and carotenoid derivative intakes as predicted by Diet ID™ were significantly correlated with NDSR output, whereas lycopene, α-carotene, Vitamin D, Vitamin B_1 (thiamin), and Vitamin B_{12} (cobalamin) were not. Lycopene has been reported to be a challenging carotenoid to measure using traditional dietary assessment tools due to the considerable variability in degradation kinetics, dependent on processing and competing nutrient interactions within the food matrix.[54, 55] It has been observed that lycopene bioavailability is higher in its cooked form compared to raw form, and therefore concentrations may differ depending on the preparation of lycopene-containing foods.[56] Lycopene metabolism and absorption has been shown to be highly correlated and contingent on macronutrient intake, specifically dietary fat and oil consumption.[57] Dietary sources of lycopene are in the *all-trans* configuration, which differs from the lycopene found in human tissue, which is in the cis-isomer configuration.[58] Due to the bulkiness of the *all-trans* lycopene, there is a

lower affinity and efficiency for micelle incorporation, and therefore higher amounts of dietary fat may inhibit the absorption of lycopene.[58] This contradicts the physiological uptake of other carotenoid compounds, in which absorption and bioavailability increases with dietary fat consumption.[59] It is unknown whether correcting for the processing of lycopene-containing foods would alter the estimated nutrient values from Diet ID™ and 24 h NDSR dietary recalls. Additionally, lycopene is predominantly present in tomatoes and tomato-based products, limiting the availability of lycopene intake from food sources, whereas other carotenoids, such as β-carotene, are found more ubiquitously in red, orange, yellow, purple, and dark green foods.[60]

Skin carotenoid scores and plasma carotenoids were used as objective measures of fruit and vegetable consumption. As overweight and obesity impacts the storage capacity of carotenoids in circulation, as well as those deposited in the skin, BMI was added as a covariate into the statistical model. Significant associations were observed between plasma carotenoids, skin carotenoid scores as measured by the Veggie Meter®, and dietary intake of total carotenoids as predicted by Diet ID™. The relationship between dietary intake and skin carotenoid scores is to be expected, as skin carotenoids represent a longer-term dietary intake of carotenoid-containing fruits and vegetables and therefore may be influenced by accretion.[15] Previous research using objective measures of dietary intake, such as plasma carotenoids or spectroscopy-based skin carotenoid measurements have also demonstrated similar moderate or weak associations due to discrepancies between subjective assessment tools for fruit and vegetable consumption and objective skin carotenoid scores.[21, 61] The observed association highlights the use of Diet ID™ as an estimate for fruit and vegetable consumption and provides the capability to extrapolate nutrient values that are comparable to carotenoid concentrations detected in plasma and skin; however, it should be acknowledged that Diet ID™ may have limited utility as a dietary

assessment tool as this comparison has only been demonstrated for carotenoid consumption in a US population.

Vitamin D intake was not significantly correlated between dietary assessment instruments. As an identified 2020–2025 DGA Nutrient of Concern, Vitamin D is only found in a small number of dietary sources, making nutrient adequacy challenging to achieve. The variation in the database from the 2017 version of NDSR and the 2019 version may explain the non-significant correlation, which likely was a result of Diet ID™ not including any fortified food items into the DQPN algorithm, such as Vitamin D found in fortified dairy products, cereals, and juices.[62, 63] Similarly, enriched and fortified grain products are a main dietary source of thiamin and therefore may have not been accurately captured by the DQPN algorithm.[64, 65] When participants were asked about fortified food products during the repeated NDSR dietary recalls, fortification status was often unknown and thus NDSR defaults were used for computation. The difficulty accounting for nutrients naturally found in a limited number of food items and intake of fortified foods may explain the deviation between instruments for Vitamin D and thiamin intake.[66]

Due to the dietary restrictions reported by participants, specifically relating to the lack animal-based food products such as eggs, meat, and lactose intolerance, consumption of overall Vitamin B_{12} intake may have been inaccurately captured. It has been previously observed that individuals following a vegetarian or vegan dietary pattern are at an increased risk for developing a Vitamin B_{12} deficiency, which is often mitigated through a form of Vitamin B_{12} supplementation.[67] As dietary supplements were not incorporated into the final nutrient analysis, B_{12} intake from non-food sources may provide further insight into the deviance in estimates of Vitamin B_{12} intake from Diet ID™ and NDSR.

The findings from this analysis support and expand upon the results from a previous study comparing Diet ID™ to Automated Self-Administered 24 h (ASA24) dietary recalls.[36] The Nutritious Eating with Soul (NEW Soul) study was a 2-year randomized nutrition intervention aimed at comparing the impact of two dietary patterns on the risk of cardiovascular disease among African American adults.[36] Although study populations differed in population size (NEW Soul $n = 68$), age (NEW Soul = 50 ± 9.6 years), and race/ethnicity (NEW Soul = 100% African American), the findings for diet quality, as measured by HEI-2015, as well as cholesterol, potassium, Vitamin C, and Vitamin E were significant between Diet ID™ and both ASA24 and NDSR dietary recalls in their respective study populations. Findings from the NEW Soul study observed significant associations in carbohydrate and protein intake, as well as copper, Vitamin B_1, and Vitamin B_{12}, some of which were not observed in the present analysis;[36] however, it should be noted that the NEW Soul study analyzed mean nutrient intake by aggregating values across all participants, whereas data analysis was performed comparing individual output from both devices in this study. Thus, comparing the magnitude of significance between studies may not be feasible as the statistical approaches were not in congruence.

Furthermore, interviewer-administered dietary recalls are considered a higher quality assessment tool for capturing dietary intake data compared to self-administered dietary recalls due to the methodical probing to acquire exact dietary details.[68] While ASA24 dietary recalls are less participant and researcher burdensome, NDSR dietary recalls are considered to be a more rigorous dietary assessment tool.[69] However, ASA24 and NDSR dietary assessments have limitations in both time and resources; therefore, Diet ID™ may be an alternative tool that can capture similar nutrient output rapidly and with vastly reduced participant and researcher burden. In addition to the more rigorous dietary collection method used in this study, the inclusion of

objective measures of skin carotenoid scores measured via the Veggie Meter® and plasma carotenoids further promotes the use of Diet ID™ to measure nutrient intake, specifically those associated with fruit and vegetable consumption. The advantages of Diet ID™ have the ability to advance dietary intake assessment methodology, though it should be noted that Diet ID™ was designed to measure overall dietary patterns and alternative assessments may be recommended to calculate exact nutrient amounts, kinetics, or degradation of dietary compounds.

Innovative techniques to successfully capture the intricacies of dietary intake are needed to reduce participant and researcher burden in the research setting, as well as extend beyond research to improve dietary monitoring for public health benefit.[70, 71] Dietary intake is closely associated with chronic disease risk, and dietary habits are often established prior to adulthood.[72] College students are a unique category of emerging adults, as many individuals in this life stage are making food choices independently for the first time, drastically altering their eating behaviors.[73] Most recently, the average HEI-2015 score for US adults was 58 out of a maximum score of 100,[74] and it has been observed that diet quality further decreases in the college student population due to financial limitations in affording healthy foods and environmental barriers to access.[75] Diet ID™ and NDSR were strongly correlated for predicting HEI-2015 scores (τ = 0.55, $p < 0.0001$); however, it should be noted that the level of agreement between the two measurements becomes less strong at HEI-2015 scores above 80 with a deviation of 7.14%. For this reason, assessing dietary intake in this population presents challenges that are often difficult to capture using traditional dietary assessment methods.

Despite these challenges, Diet ID™ was able to quickly estimate diet quality, consumption of total calories, protein, carbohydrates, and a majority of micronutrients, phytonutrients, and nutrients of concern with substantially less participant and researcher burden than other

established methodologies, which signals potential for Diet ID™ to be utilized in clinical and outpatient settings as a dietary assessment method. Additionally, as the image-based technology allows for universal visual recognition, Diet ID™ may be able to be implemented in populations of low or limited literacy, and non-native English speakers. This study assessed the use of Diet ID™ in a population of college students, including individuals experiencing acute and chronic food insecurity.

Strengths and Limitations

It is imperative to recognize both the strengths and limitations of the present study. This study is the first to compare the innovative Diet ID™ technology to subjective and objective measures of dietary intake in a population of emerging adults. As this is a secondary validation from the previously mentioned study disrupted due to the COVID-19 pandemic, the total sample size was intended to be larger; however, the observed sample size in the data collected was sufficiently powered to analyze plasma carotenoids as the primary outcome. Thus, it is possible that the present study is underpowered to detect associations in certain nutrients of interest with high interindividual variability.

Due to the racial and ethnic diversity of the college student population at the University of California Davis, foods commonly consumed by participants may have not been present in the NDSR database nor in the images displayed in Diet ID™. To account for this, the Diet ID™ algorithm is currently expanding their patented algorithm to include a larger database of culturally diverse foods to better encompass the diversity of the eating patterns among people living in the US and to identify dietary patterns in other parts of the world. As Diet ID™ does not account for dietary supplements, all reported supplements were excluded from the NDSR nutrient output; therefore, intake of some nutrients may be higher than recorded as a result of

supplementation. For the purpose of this analysis, dietary intake data without supplements was used for uniformity between outputs. While NDSR dietary recalls were unannounced and Diet ID™ was utilized as a self-assessment with limited supervision, it is possible that there was desirability or response bias among the participants. College students generally consume a lower quality diet than other adult populations; therefore, these findings may not be generalizable to all adult populations.[76-78]

Conclusions

The findings from this study support the use of Diet ID™ as a rapid, non-invasive dietary assessment tool that may provide comparable estimates of nutrient consumption against repeated 24 h NDSR dietary recalls, skin carotenoid scores, and plasma carotenoids. Innovative diet capture technology, such as Diet ID™, has the potential to be implemented in both clinical and community settings to increase habitual dietary monitoring, with the goal of developing awareness around food choices to initiate health-promoting behaviors across the lifespan and in racially and ethnically diverse populations.

This manuscript was published:

Radtke MD, Chodur GM, Bissell MCS, Kemp L, Medici V, Steinberg FM, Scherr RS. Validation of Diet ID™ in Predicting Nutrient Intake Compared to NDSR Dietary Recalls and Skin Carotenoid Scores in a Diverse Population of College Students. *Nutrients* 2023; 15(2): 409. https://doi.org/10.3390/nu15020409

References

1. Pico, C.; Serra, F.; Rodriquez, A.M.; Keijer, J.; Palou, A. Biomarkers of Nutrition and Health: New Tools for New Approaches. Nutrients 2019, 11, 1092.
2. Roark, R.A.; Niederhauser, V.P. Fruit and vegetable intake: Issues with definition and measurement. Public Health Nutr. 2013, 16, 2–7.
3. Slavin, J.L.; Lloyd, B. Health benefits of fruits and vegetables. Adv. Nutr. 2012, 3, 506–516.
4. Liu, R.H. Health-promoting components of fruits and vegetables in the diet. Adv. Nutr. 2013, 4, 384S–392S.
5. Shim, J.S.; Oh, K.; Kim, H.C. Dietary assessment methods in epidemiologic studies. Epidemiol Health 2014, 36, e2014009.
6. Penn, L.; Boeing, H.; Boushey, C.J.; Dragsted, L.O.; Kaput, J.; Scalbert, A.; Welch, A.A.; Mathers, J.C. Assessment of dietary intake: NuGO symposium report. Genes Nutr. 2010, 5, 205–213.
7. Macdiarmid, J.; Blundell, J. Assessing dietary intake: Who, what and why of under-reporting. Nutr. Res. Rev. 1998, 11, 231–253.
8. McClung, H.L.; Ptomey, L.T.; Shook, R.P.; Aggarwal, A.;Gorczyca, A.M.; Sazonov, E.S.; Becofsky, K.; Weiss, R.; Das, S.K. Dietary Intake and Physical Activity Assessment: Current Tools, Techniques, and Technologies for Use in Adult Populations. Am. J. Prev. Med. 2018, 55, e93–e104.
9. Thompson, F.E.; Subar, A.F.; Loria, C.M.; Reedy, J.L.; Baranowski, T. Need for technological innovation in dietary assessment. J. Am. Diet. Assoc. 2010, 110, 48–51.

10. Moran, N.E.; Mohn, E.S.; Hason, N.; Erdman, J.W., Jr.; Johnson, E.J. Intrinsic and Extrinsic Factors Impacting Absorption, Metabolism, and Health Effects of Dietary Carotenoids. Adv. Nutr. 2018, 9, 465–492.
11. Radtke, M.D.; Jilcott, P.S.; Jahns, L.; Firnhaber, G.C.; Loofbourrow, B.M.; Zeng, A.; Scherr, R.E. Criterion-Related Validity of Spectroscopy-Based Skin Carotenoid Measurements as a Proxy for Fruit and Vegetable Intake: A Systematic Review. Adv. Nutr. 2020, 11, 1282–1299.
12. Ermakov, I.V.; Ermakova, M.; Sharifzadeh, M.; Gorusupudi, F.K.; Bernstein, P.S.; Stookey, J.; Evans, J.; Arana, T.; Tao-Lew, L.; et al. Optical assessment of skin carotenoid status as a biomarker of vegetable and fruit intake. Arch. Biochem. Biophys. 2018, 646, 46–54.
13. Burrows, T.L.; Rollo, M.E.; Williams, R.; Wood, L.G.; Garg, M.L.; Jensen, M.; Collins, C.E. A Systematic Review of Technology-Based Dietary Intake Assessment Validation Studies That Include Carotenoid Biomarkers. Nutrients 2017, 9, 140.
14. Longevity Link Corporation. The Veggie Meter —Skin Carotenoids as Indicator of Fruit and Vegetable Intake and Biomarker Associated with Better Health [Internet]. 2016. Available online: Longevitylinkcorporation.com/home.html. (accessed on 11 April 2022).
15. Jahns, L.; Johnson, L.K.; Mayne, S.T.; Cartmel, B.; Picklo Sr, M.J.; Ermakov, I.V.; Gellermann, W.; Whigham, L.D. Skin and plasma carotenoid response to a provided intervention diet high in vegetables and fruit: Uptake and depletion kinetics. Am. J. Clin. Nutr. 2014, 100, 930–937.
16. Ermakov, I.V.; Gellermann, W. Dermal carotenoid measurements via pressure mediated reflection spectroscopy. J. Biophotonics 2012, 5, 559–570.

17. Boushey, C.J.; Spoden, M.; Zhu, F.M.; Delp, E.J.; Kerr, D.A. New mobile methods for dietary assessment: Review of image-assisted and image-based dietary assessment methods. Proc. Nutr. Soc. 2017, 76, 283–294.
18. Ho, D.K.N.; Tseng, S.; Wu, M.; Shih, C.; Atika, A.P.; Chen, Y.; Chang, J. Validity of image-based dietary assessment methods: A systematic review and meta-analysis. Clin. Nutr. 2020, 39, 2945–2959.
19. Diet ID—Diet is a Vital Sign [Internet]. Available online: https://www.dietid.com. (accessed on 11 April 2022).
20. Katz, D.L.; Rhee, L.Q.; Katz, C.S.; Aronson, D.L.; Frank, G.C.; Gardner, C.D.; Willett, W.C.; Dansinger, M.L. Dietary assessment can be based on pattern recognition rather than recall. Med. Hypotheses 2020, 140, 109644.
21. Jahns, L.; Johnson, L.K.; Conrad, Z.; Bukowski, M.; Raatz, S.K.; Jilcott Pitts, S.; Wang, Y.; Ermakov, I.V.; Gellermann, W. Concurrent validity of skin carotenoid status as a concentration biomarker of vegetable and fruit intake compared to multiple 24-h recalls and plasma carotenoid concentrations across one year: A cohort study. Nutr. J. 2019, 18, 78.
22. Jahns, L.; Johnson, L.K.; Scheett, A.J.; Stote, K.S.; Raatz, S.K.; Subar, A.F.; Tande, D. Measures of Diet Quality across Calendar and Winter Holiday Seasons among Midlife Women: A 1-Year Longitudinal Study Using the Automated Self-Administered 24-Hour Recall. J. Acad. Nutr. Diet 2016, 116, 1961–1969.
23. Mounien, L.; Tourniaire, F.; Landrier, J.F. Anti-Obesity Effect of Carotenoids: Direct Impact on Adipose Tissue and Adipose Tissue-Driven Indirect Effects. Nutrients 2019, 11, 1562.

24. Holt, E.W.; Wei, E.K.; Bennett, N.; Zhang, L.M. Low skin carotenoid concentration measured by resonance Raman spectroscopy is associated with metabolic syndrome in adults. Nutr. Res. 2014, 34, 821–826.
25. Melendez-Martinez, A.J.; Stinco, C.M.; Mapelli-Brahm, P. Skin Carotenoids in Public Health and Nutricosmetics: The Emerging Roles and Applications of the UV Radiation-Absorbing Colourless Carotenoids Phytoene and Phytofluene. Nutrients 2019, 11, 1093.
26. United States Department of Agriculture (USDA) E.R.S. Food Security in the U.S.: Measurement. Available online: https://www.ers.usda.gov/topics/food-nutrition-assistance/food-security-in-the-u-s/measurement/#measurement. (accessed on 28 September 2022).
27. United States Department of Agriculture (USDA) E.R.S. Food Security in the U.S.: Definitions of Food Security 2021, Available online: https://www.ers.usda.gov/topics/food-nutrition-assistance/food-security-in-the-us/definitions-of-food-security/. (accessed on 28 September 2022).
28. National Center for Health Statistics. What We Eat in America/National Health and Nutrition Examination Survey, 2013–2014; Healthy Eating Index-2015. Available online: https://www.fns.usda.gov/healthy-eating-index-hei#1. (accessed on 29 November 2022).
29. Reedy, J.; Lerman, J.L.; Krebs-Smith, S.M.; Kirkpatrick, S.I.; Pannucci, T.E.; Wilson, M.M.; Subar, A.F.; Kahle, L.L.; Tooze, J.A. Evaluation of the Healthy Eating Index-2015. J. Acad. Nutr. Diet 2018, 118, 1622–1633.
30. Radtke, M.D.; Poe, M.; Stookey, J.; Jilcott Pitts, S.; Moran, N.E.; Landry, M.J.; Rubin, L.P.; Stage, V.C.; Scherr, R.E. Recommendations for the Use of the Veggie Meter® for

Spectroscopy-Based Skin Carotenoid Measurements in the Research Setting. Curr. Dev. Nutr. 2021, 5, nzab104.

31. Craft, N.E. High resolution HPLC method for the simultaneous analysis of carotenoids, retinoids, and tocopherols. FASEB 1996, 10, A527.
32. Craft, N.E. Chromatographic Techniques for Carotenoid Separation. Curr. Protoc. Food Anal. Chem. 2001, 1, F2. 3.1–F2. 3.15.
33. Centers for Disease Control and Prevention. Laboratory Manual: Analysis of Vitamin A/Vitamin E/Carotenoids for NHANES. Available online: https://www.cdc.gov/nchs/data/nhanes/nhanes_03_04/l45vit_c_met_vitAE_carotenoids.pdf. (accessed on 12 October 2022).
34. SAS Institute Inc. SAS/ACCESS® 9.4 Interface to ADABASL: Reference. SAS Institute Inc.: Cary, NC, USA, 2013.
35. Bland, M.J.; Altman, D. Statistical methods for assessing agreement between two methods of clinical measurement. Lancet 1996, 327, 307–310.
36. Turner-McGrievy, G.; Hutto, B.; Bernhart, J.A.; Wilson, M.J. Comparison of the Diet ID™Platform to the Automated Self-administered 24-hour (ASA24) Dietary Assessment Tool for Assessment of Dietary Intake. J. Am. Coll. Nutr. 2021, 41, 1–23.
37. Baldrick, F.R.; Woodside, J.V.; Elborn, J.S.; Young, I.S.; McKinley, M.C. Biomarkers of fruit and vegetable intake in human intervention studies: A systematic review. Crit. Rev. Food Sci. Nutr. 2011, 51, 795–815.
38. Woodside, J.V.; Draper, J.; Llyod, A.; McKinley, M.C. Use of biomarkers to assess fruit and vegetable intake. Proc. Nutr. Soc. 2017, 76, 308–315.

39. Block, G.; Norkus, E.; Hudes, M.; Mandel, S.; Helzlsouer, K. Which Plasma Antioxidants Are Most Related to Fruit and Vegetable Consumption? Am. J. Epidemiol 2001, 154, 1113–1118.
40. US Department of Health and Human Services and US Department of Agriculture. In 2020–2025 Dietary Guidelines for Americans, 9th Ed.; December 2020. Available online: https://www.dietaryguidelines.gov (accessed on 28 September 2022).
41. Rush, E.; Amoah, I.; Diep, T. Jalili-Moghaddam Determinants and Suitability of Carotenoid Reflection Score as a Measure of Carotenoid Status. Nutrients 2020, 12, 13.
42. Tobias, T.A.M.; Wood, L.G.; Rastogi, D. Carotenoids, fatty acids and disease burden in obese minority adolescents with asthma. Clin. Exp. Allergy 2019, 49, 838–846.
43. Vioque, J.; Weinbrenner, T.; Asensio, L.; Castelló, A.; Young, I.S.; Fletcher, A. Plasma concentrations of carotenoids and vitamin C are better correlated with dietary intake in normal weight than overweight and obese elderly subjects. Br. J. Nutr. 2007, 97, 977–986.
44. Stata 13 Base Reference Manual V13. Stata Press. StataCorp: College Station, TX, USA, 2013.
45. Fontana, J.M.; Pan, Z.; Sazonov, E.S.; McCrory, M.A.; Graham Thomas, J.; McGrane, K.S.; Marden, T.; Higgins, J.A. Reproducibility of Dietary Intake Measurement From Diet Diaries, Photographic Food Records, and a Novel Sensor Method. Front. Nutr. 2020, 7, 99.
46. Beasley, J.; Riley, W.T.; Jean-Mary, J. Accuracy of a PDA-based dietary assessment program. Nutrition 2005, 21, 672–677.
47. Mozaffarian, D.; Forouhi, N.G. Dietary guidelines and health-is nutrition science up to the task? BMJ 2018, 360, k822.

48. Slavin, M.; Polasky, A.; Vieyra, K.; Best, A.; Durant, L.; Frankenfeld, C. Single-Meal Nutrient Assessment by a Self-Administered, Electronic Exit Survey Compared with a Multipass Dietary Interview in University Undergraduates in an All-You-Care-to-Eat Campus Dining Hall. J. Acad. Nutr. Diet 2019, 119, 739–749.
49. Glanz, K.; Brug, J.; van Assema, P. Are awareness of dietary fat intake and actual fat consumption associated?—a Dutch-American comparison. Eur. J. Clin. Nutr. 1997, 51, 542–547.
50. Patel, K.A.; Schlundt, D.G. Impact of moods and social context on eating behavior. Appetite 2001, 36, 111–118.
51. Rumpler, W.V.; Kramer, M.; Rhodes, D.G.; Moshfegh, A.J.; Paul, D.R. Identifying sources of reporting error using measured food intake. Eur. J. Clin. Nutr. 2008, 62, 544–552.
52. Johansson LSolvoll, K.; Bjørneboe, G.E.; Drevon, C.A. Under- and overreporting of energy intake related to weight status and lifestyle in a nationwide sample. Am. J. Clin. Nutr. 1998, 68, 266–274.
53. McKenzie Bl Coyle, D.H.; Santos, J.A.; Burrows, T.; Rosewarne, E.; Peter, S.A.E.; Carcel, C.; Jaacks, L.M.; Norton, R.; Collins, C.E.; Woodward, M.; Webster, J. Investigating sex differences in the accuracy of dietary assessment methods to measure energy intake in adults: A systematic review and meta-analysis. Am. J Clin. Nutr. 2021, 113, 1241–1255.
54. Porrini, M.; Riso, P. What are typical lycopene intakes? J. Nutr. 2005, 135, 2042S–5S.
55. Mayeaux, M.; Xu, A.; King, J.M.; Prinyawiwatkul, W. Effects of Cooking Conditions on the Lycopene Content in Tomatoes. J. Food Sci. 2006, 71, C461–C464.

56. Fielding, J.M.; Rowley, K.G.; Cooper, P.; O' Dea, K. Increases in plasma lycopene concentration after consumption of tomatoes cooked with olive oil. Asia Pac. J. Clin. Nutr. 2005, 14, 131–136.
57. Garcia, A.L.; Koebnick, C.; Dagnelie, P.C.; Strassner Carola Elmadfa, I.; Katz, N.; Leitzmann Hoffman, I. Long-term strict raw food diet is associated with favourable plasma beta-carotene and low plasma lycopene concentrations in Germans. Br. J. Nutr. 2008, 99, 1293–1300.
58. Erdman, J.W., Jr. How do nutritional and hormonal status modify the bioavailability, uptake, and distribution of different isomers of lycopene? J. Nutr. 2005, 135, 2046S–7S.
59. Donhowe, E.G.; Kong, F. Beta-carotene: Digestion, Microencapsulation, and In Vitro Bioavailability. Food Bioproc. Tech. 2014, 7, 338–354.
60. Britton, G.; Liaaen-Jensen, S.; Pfander, H. Carotenoids Handbook; Birkhäuser Verlag: Basel, Switzerland; Boston, MA, USA; Belrin, Germany, 2004; Volume 1.
61. Beccarelli, L.M.; Scherr, R.E.; Dharmar, M.; Ermakov, I.V.; Gellermann, W.; Jahns, L.; Linnell, J.D.; Keen, C.L.; Steinberg, F.M.; Young, H.M.; Zidenberg-Cherr, S. Using Skin Carotenoids to Assess Dietary Changes in Students After 1 Academic Year of Participating in the Shaping Healthy Choices Program. J. Nutr. Educ. Behav. 2017, 49, 73–78e1.
62. Calvo, M.S.; Whiting, S.J.; Barton, C.N. Vitamin D fortification in the United States and Canada: Current status and data needs. Am. J. Clin. Nutr. 2004, 80 (Suppl. 6), 1710S-1716S.
63. Moore, C.E.; Murphy, M.M.; Keast, D.R.; Holick, M.F. Vitamin D intake in the United States. J. Am. Diet Assoc. 2004, 104, 980–983.
64. Mielgo-Ayuso, J.; Aparico-Ugarriza, R.; Olza, J.; Aranceta-Bartrina, J.; Gil, A.; Ortega, R.M.; Serra-Majem, L.; Varela-Moreiras, G.; González-Gross, M. Dietary Intake and Food

Sources of Niacin, Riboflavin, Thiamin and Vitamin B(6) in a Representative Sample of the Spanish Population. The Anthropometry, Intake, and Energy Balance in Spain (ANIBES) Study dagger. Nutrients 2018, 10, 846.

65. Subar, A.F.; Krebs-Smith, S.M.; Cook, A.; Kahle, L.L. Dietary Sources of Nutrients among US Adults, 1989 to 1991. J. Am. Diet Assoc. 1998, 98, 537–547.
66. Byrdwell, W.C.; Devries, J.; Exler, J.; Harnly, J.M.; Holden, J.M.; Holick, M.F.; Hollis, B.W.; Horst, R.L.; Lada, M.; Lemar, L.E.; et. al. Analyzing vitamin D in foods and supplements: Methodologic challenges. Am. J. Clin. Nutr. 2008, 88, 554S–557S.
67. Pawlak, R.; Parrott, S.J.; Raj, S.; Cullum-Dugan, D.; Lucus, D. How prevalent is vitamin B(12) deficiency among vegetarians? Nutr. Rev. 2013, 71, 110–117.
68. Thompson, F.W.; Dixit-Joshi, S.; Potischman, N.; Dodd, K.W.; Kirkpatrick, S.I.; Kushi, L.H.; Alexander, G.L.; Coleman, L.A.; Zimmerman, T.P.; Sundaram, M.E.; et al. Comparison of Interviewer-Administered and Automated Self-Administered 24-Hour Dietary Recalls in 3 Diverse Integrated Health Systems. Am. J. Epidemiol. 2015, 181, 970–978.
69. Timon, C.M.; van den Barg, R.; Blain, R.J.; Kehoe, L.; Evans, K.; Walton, J.; Flynn, A.; Gibney, E.R. A review of the design and validation of web—and computer-based 24-h dietary recall tools. Nutr. Res. Rev. 2016, 29, 268–280.
70. Lupton, D. Self-tracking, health and medicine. Health Sociol Rev. 2016, 26, 1–5.
71. Lucassen, D.A.; Brouwer-Brolsma, E.M.; Slotegraaf, A.I.; Kok, E.; Feskens, E.J.M. DIetary ASSessment (DIASS) Study: Design of an Evaluation Study to Assess Validity, Usability and Perceived Burden of an Innovative Dietary Assessment Methodology. Nutrients 2022, 14, 1156.

72. Mennella, J.A.; Ventura, A.K. Early feeding: Setting the stage for healthy eating habits. Nestle Nutr. Workshop Ser. Pediatr. Program 2011, 68, 153–163. Discussion 164–168.
73. Abraham, S.; Noriega, B.R.; Shin, J.Y. College students eating habits and knowledge of nutritional requirements. J. Nutr. Human Health 2018, 2, 13-17.
74. US Department of Agriculture: Healthy Eating Index-2015 Scores. Accessed on: https://www.fns.usda.gov/resource/healthy-eating-index-hei. (accessed on 29 November 2022).
75. Merhout, F.; Doyle, J. Socioeconomic Status and Diet Quality in College Students. J. Nutr. Educ. Behav. 2019, 51, 1107–1112.
76. Sogari, G.; Velez-Argumedo, C.; Gómez, M.I.; Mora, C. College Students and Eating Habits: A Study Using An Ecological Model for Healthy Behavior. Nutrients 2018, 10, 1823.
77. Martinez, S.M.; Grandner, M.A.; Nazmi, A.; Ruben Canedo, E.; Ritchie, L.D. Pathways from Food Insecurity to Health Outcomes among California University Students. Nutrients 2019, 11, 1419.
78. Nelson, M.C.; Larson, N.I.; Barr-Anderson, D.; Neumark-Sztainer, D.; Story, M. Disparities in dietary intake, meal patterning, and home food environments among young adult nonstudents and 2- and 4-year college students. Am. J. Public Health 2009, 99, 1216–1219.

Table 1. Baseline participant characteristics expressed as mean ± standard deviations for age, BMI, SCS, and nutrition knowledge, and the number and percentage of participants in subgroup by sex, race/ethnicity, and food security status (*n* = 48).

Age, Years (Mean ± SD)	22.09 ± 2.36
Biological Sex	
Male	12 (25%)
Female	36 (75%)
Race/Ethnicity	
African American/Black, not of Hispanic origin	1 (2%)
American Indian/Alaska native	0
Asian/Pacific Islander	23 (48%)
White, not of Hispanic origin	9 (19%)
Latin/Hispanic (Mexican-American, Puerto Rican, Cuban)	10 (21%)
Other	1 (2%)
Unknown/Prefer not to answer	4 (8%)
Food Security Status	
High	19 (40%)
Marginal	15 (31%)
Low	8 (17%)
Very Low	6 (12%)
BMI (mean ± SD; kg/m^2)	
Total	24.58 ± 5.04
Male	25.79 ± 4.47
Female	24.18 ± 5.22
Timepoint 1: SCS (mean ± SD)	322.98 ± 114.42
Timepoint 2: SCS (mean ± SD)	341.35 ± 113.98

Table 2. Correlation coefficients between nutrient values predicted by Diet ID™ and 24 h dietary recalls (n = 42) by Pearson's correlation[a] or Kendall's tau correlation [b].

Nutrient	Correlation Coefficient	*p*-Value
HEI-2015 Score[b]	0.55	**<0.001**
Calories (kcals)[a]	0.36	**0.02**
Protein (g)[a]	0.55	**0.0002**
Carbohydrates (g)[a]	0.31	**<0.05**
Fat (g)[a]	0.29	NS (p = 0.06)
Cholesterol (mg)[b]	0.32	**0.003**
Vitamin A (mcg)[a]	0.39	**0.01**
Total Carotenoids (mcg)[a]	0.44	**0.003**
α-carotene (mcg)[b]	0.14	NS (p = 0.19)
β-carotene (mcg)[b]	0.39	**0.0003**
Lycopene (mcg)[b]	−0.09	NS (p = 0.40)
Lutein and Zeaxanthin (mcg)[a]	0.58	**0.0001**
Dietary Fiber (g)[a]	0.64	**<0.0001**
Calcium (mg)[a]	0.36	**0.02**
Vitamin C (mg)[a]	0.44	**0.003**
Vitamin D (mcg)[a]	0.13	NS (p = 0.41)
Vitamin E (mg)[a]	0.35	**0.02**
Sodium (mg)[a]	0.36	**0.02**
Potassium (mg)[a]	0.58	**0.0001**
Folate (mcg)[a]	0.37	**0.02**
Iron (mg)[a]	0.31	**0.04**
Vitamin B_1 (Thiamin) (mg)[a]	0.13	NS (p = 0.40)
Vitamin B_2 (Riboflavin) (mg)[a]	0.34	**0.03**
Vitamin B_3 (Niacin) (mg)[a]	0.42	**0.005**
Vitamin B_6 (Pyridoxine) (mg)[a]	0.57	**0.0001**
Vitamin B_{12} (Cobalamin) (mcg)[b]	0.18	NS (p = 0.09)

Table 3. Relationship between skin carotenoid scores (SCS) measured using the Veggie Meter® and plasma carotenoids measured using Diet ID™ controlling for BMI (*n* = 42).

Variables	**Linear Regression (Adjusted R^2)**	***p*-Value**
SCS and total carotenoids from Diet ID™; controlling for BMI	0.41	**<0.0001**
Total plasma carotenoids and Total Carotenoids from Diet ID™; controlling for BMI	0.37	**0.0001**
SCS and total plasma carotenoids, controlling for BMI	0.68	**<0.0001**

CHAPTER 4

Impact of On-Campus Food Access Resources on Diet Quality and Diet-related Biomarkers

Introduction

University students are a population of emerging adults who experience disproportionate prevalences of food insecurity.[1] Food insecurity, as defined by the United States Department of Agriculture (USDA), is an economic or social condition that results in the limited or uncertain access to sufficient quality or quantity of foods.[2] Previously referred to as food insecurity without hunger, low food security (LFS) status is the experience of a reduced quality diet, but is not associated with a reduction in the quantity of food intake.[2] Very low food security (VLFS) status (formerly food insecurity with hunger), is a disrupted eating pattern that results in a reduction in the quantity and/or quality of foods needed to meet nutrient needs.[2] Food insecurity can be acute or chronic, with both experiences having negative implications on mental and physical well-being. Food insecurity in the college student population has been associated with a reduction in academic performance, sleep quality and duration, perceived health status, diet quality, and increased feelings of anxiety and depression.[3-7]

To address the multitude of health consequences linked to the experience of food insecurity on college campuses, many universities have responded by developing on-campus food access resources. Existing research on the efficiacy of on-campus food access resources on health outcomes has consisted predominately of anecdotal, qualitative responses or self-reported health outcomes.[8] However, with improvements to dietary intake from the food items offered at on-campus food access resources, there may be quantifiable changes in health and diet-related biomarkers. When fruits and vegetables (F/V), whole grains, low-fat dairy, lean protein, and other foods attributed to a healthy eating pattern are ingested in increasing amounts, the composition of circulating and stored nutrient compounds will reflect a higher diet quality.[9]

Although universities are seeking to improve the access to nutrient-rich foods, college students generally exhibit poor diet quality due to increased consumption of low-cost, convenient food choices that are often high in added sugars and dietary fat, and low in F/V.[10] Due to the chronic underconsumption of F/V in the college student population, there is an urgency for on-campus food access resources to prioritize fresh, frozen, and canned F/V items.

Fruits and vegetables are food groups of concern identified by the Dietary Guidelines for Americans (DGAs), with 80% to 90% of the population not meeting F/V recommendations, respectively.[11] Fruits and vegetables contain a multitude of health-promoting bioactive compounds, including carotenoids.[12] Carotenoids are a class of fat-soluble phytonutrients found in a variety of F/V, yielding a useful biomarker for measuring changes in F/V consumption. With over 700 identified carotenoid compounds, α-carotene, β-carotene, lycopene, lutein, and zeaxanthin are the most commonly consumed in the diet, and therefore are present in greater amounts in human tissues and blood.[13, 14] In addition to detecting carotenoids in plasma and serum, carotenoids may also be quantified non-invasively using spectroscopy-based methodologies. In plasma and serum, carotenoids are reflective of short-term consumption of F/V due half-life degradation,[15] whereas carotenoids deposited in the adipose and other tissues indicate long-term, habitual intake.[16]

Evaluating the efficacy of university efforts to support the expansion of food access resources to improve student's food security status and diet quality through increasing F/V consumption is warranted. Therefore, the aim of this study was to investigate various biomarkers in the blood and the skin, as well as dietary intake of carotenoids to determine if the use of on-campus food access resources improve health-related biomarkers.

Methods

The study protocol and procedures were approved by the University of California, Davis Institutional Review Board. Informed consent was acquired prior to study commencement (IRB: 1476178).

Participants

Participants were recruited from the undergraduate and graduate student population at the University of California, Davis through social media, online advertisements, and campus fliers. To be eligible, biological males and females had to be above the age of 18 and within a body mass index (BMI) range of 18.5-34.9 kg/m^2. Participants with a BMI above 35.0 kg/m^2 were excluded to reduce physiological interference in carotenoid storage in the adipose for subcutaneous dermal detection.[17, 18] Exclusion criteria included smoking or living in a household with an indoor smoker (including cigarettes, electronic cigarettes, vaping, and marijuana), and consuming more than five alcoholic drinks per week, as the bioavailability of carotenoid compounds under these metabolic conditions is variable.[19] The use of oral or topical high-dose Vitamin A medication (Accutane, retinol cream, etc.) was exclusionary as this may increase Vitamin A or carotenoid status from non-dietary sources.[20] Individuals participating in artificial tanning methods, such as UV-based tanning beds, were ineligible as excessive UV exposure may result in the mobilization of carotenoids stores for antioxidant scavenging to protect against photo oxidation, therefore reducing total carotenoid levels in the body.[21]

Study Design

This prospective cohort study, conducted in accordance with the academic term (April 2022 to June 2022), included two in-person clinic visits, with at least a six-week duration between baseline (time point one) and follow-up (time point two). This duration between clinic visits was

determined to account for the biomarkers of interest in the blood and skin to respond to changes in dietary intake,[22] as well as capture the usage of on-campus food access resources over multiple weeks. Participants were instructed to avoid intentionally altering any dietary or physical activity behaviors during the study. Participants were not limited to the number of times they could visit the food access resources available on campus; however, attendance was recorded weekly over the study duration for the potential mediating effect. Clinic visits consisted of a venipuncture, anthropometric and skin carotenoid measurements, in addition to a validated dietary assessment to estimate dietary patterns reflective of 30 days of intake.[23] Sociodemographic information was assessed at baseline and food security status was measured at both time points to capture potential changes throughout the term. The usage of the on-campus food access resources was monitored over the duration of the study using surveys in Qualtrics (Qualtrics, Provo, UT, USA).[24]

Plasma Carotenoids

Participants fasted for a minimum of 10 hours prior to the study visit. Blood samples were collected through venipuncture by a trained phlebotomist at the UC Davis Ragle Human Nutrition Research Center using EDTA vacutainer blood collection tubes. Whole blood was centrifuged at 1500 rpm for 15 minutes at 4°C and the plasma was extracted, aliquoted, and stored at -80°C prior to carotenoid analysis at Eurofins Craft Technologies.

Plasma carotenoids were quantified via HPLC using a modification of the procedures described by Craft et al.,[25, 26] and previously published in further detail.[23] Briefly, individual carotenoids were extracted using a mobile phase of 83% acetonitrile/13% dioxane/4% methanol containing 150 mM ammonium acetate and 0.1% triethylamine at a flow rate of 1.0 ml for 21 minutes with a column temperature at 20^{o}C. Separation was performed isocratically on a Polaris

C18 Ether column (3 μm,4.0mm x 250mm) particle size with guard cartridge system. Samples were analyzed for α-carotene, β-carotene, lycopene, lutein, and zeaxanthin using a diode array detector at 450 nm and 325 nm. This method is calibrated with neat standards within the physiological range, which are assigned concentrations using absorption coefficients ($E^{1\%}_{cm}$) and corrected for HPLC purity.[27] The calibration method utilizes external standards using peak areas and corrected for tocol as the internal standard.

Skin Carotenoid Scores

Skin carotenoid scores were assessed using the Veggie Meter®. The Veggie Meter® detects and quantifies the density of carotenoids stored in the subcutaneous dermal adipose using pressure-mediated reflection spectroscopy.[28] Skin carotenoid scores were collected using the non-dominant *digital medicinalis* (ring finger) after cleaning the site with soap and water.[29] Triplicate measurements were averaged to account for potential chromophore interference and the mean skin carotenoid score was used for analysis.[29] The Veggie Meter® employs a spectral de-convolution algorithm to correct for melanin concentration, and the topical pressure applied by the device reduces the presence of hemoglobin and oxyhemoglobin, which can compete for detection in the spectral range.[28] Skin carotenoid scores are an objective measure of dietary intake and have previously been correlated with plasma and serum carotenoids, as well as measures of self-reported dietary intake.[16]

Dietary Intake

Dietary intake was assessed using Diet ID™. Diet ID™ is an innovative dietary assessment tool that utilizes Diet Quality Photo Navigation (DQPN) to identify dietary patterns and estimate nutrient intake.[30] The DQPN predicts diet quality in accordance with the Healthy Eating Index determined by the Dietary Guidelines for Americans 2020 – 2025 using the Nutrition Data

System for Research (NDSR) database (Version 2017).[30] Diet quality and nutrient intake estimated by Diet ID™ have been correlated against nutrient values from Food Frequency Questionnaires (FFQs), repeated 24-hour NDSR dietary recalls, plasma carotenoids, and skin carotenoid scores.[23, 31]

Food Access Resource Usage

The usage of on-campus food access resources was monitored through the Aggie Swipe System, a campus-wide database that records the student identification numbers of participants. The UC Davis Aggie Compass houses the Basic Needs Center, which includes a food pantry, the twice-weekly free, fresh produce pick-up, *Fruit and Veggie-Up!*, and other means for acquiring fresh and shelf-stable food items. In addition to the frequency of use, a weekly qualitative survey was administered using Qualtrics to assess the specific items consumed from food access resources to identify carotenoid-rich food sources, characterized using the USDA-NCC Carotenoid Database.[32] Participants were also asked to report CalFresh usage (formally known at SNAP/EBT), as the Aggie Compass provides resources for enrolling in state and/or federal food assistance programs.

Sociodemographic Characteristics

Sociodemographic information including age, sex, race/ethnicity, academic standing, and physical activity was collected, as well as pertinent information regarding finances, including food security status, and financial support from employment, state and federal scholarships, or parents/guardians as potential covariates. Food security status was determined using the United States Department of Agriculture (USDA) 10-item Adult Food Security Survey Module (AFFSM).[33] In accordance with the USDA classifications, participants were classified as the

following: 0: high food security; 1 – 2: marginal food security; 3 – 5: low food security; and 6 – 10: very low food security.

Statistical Analysis

A minimum of 120 participants were needed based on previous studies indicating statistical and biologic precision between plasma carotenoids, skin carotenoids, and dietary carotenoids with this sample size (based on α=0.05 and 80% power).[34] To account for a 20% attrition rate between time points, a goal of approximately 150 participants were to be recruited at baseline.

Statistical analyses were performed using Stata v13 (StataCorp, College Station, TX, USA). Data were inspected for normality using Shapiro Wilks and transformed as necessary. Descriptive data on participant characteristics are expressed as absolute (n) and relative frequencies (%) for categorical variables and mean +/- standard deviation (SD) for continuous variables. Pearson's and Kendall's Tau correlations were calculated to assess carotenoid and diet quality measures from Veggie Meter®, Diet ID™, and plasma biomarkers.

Food access resource usage over the 8-week study duration was stratified by frequency into quartiles (IQR), representing no use (0), low use (1 – 3), moderate use (4 – 5), and high use (6 – 8) and paired t-tests were used to analyze the differences in biomarkers of interest. To identify group differences, analysis of covariance (ANCOVA) was conducted to determine the interaction of food security status and usage of food access resources on changes to plasma, skin, and dietary carotenoids. Multivariate linear regression models were constructed to explore the association between frequency of interactions with on-campus food access resources and changes in diet quality outcomes in the blood, skin, and Diet ID™, controlling for biological sex, BMI, food security status, first generation student status, and academic standing. Potential confounders were identified by using Pearson's correlations to test the association on plasma,

skin, and dietary carotenoids, with $p < 0.10$. Statistical significance for all other analyses were set at $p < 0.05$.

Results

Participant Characteristics

A total of 413 participants were assessed for eligibility, of which 271 were not eligible for inclusion, resulting in a baseline study population of 142 participants (**Figure 1**). Of the 142 participants who completed time point one, 10 participants did not complete time point two and therefore were not included due to incomplete data. A total of 132 participants, a retention rate of 93%, completed all study protocols and are included in the present analysis.

Participant characteristics are detailed in **Table 1**. The cohort was predominately female (81.8%), with an average age of 23.1 ± 4.0 years. The population was racially/ethnically diverse: 1.5% non-Hispanic black, 48.5% Asian or Pacific Islander, 22.7% Hispanic, 22.0% non-Hispanic White, 3.8% multi-racial, and 1.5% other. Participants were both undergraduate (79.5%) and graduate (20.5%) students, with a majority of students receiving financial aid (58.3%) and 39.4% identifying as first-generation college students. Of the total participants, 52.3% had high food security, 22.0% were marginally food secure, 15.9% were experiencing low food security, and 9.8% had very low food security. Due to the brevity of the academic term, as expected there were no significant changes in BMI from time point one to time point two.

Food Access Resource Usage

The response rate to the weekly questionnaire on food access resource usage was 92%. Participants accessed the food access resources an average of 3.1 ± 2.6 times over the study duration, with a range of 0 to 8 uses. Of the participants who utilized the food access resources, 96% of the food items acquired contained one or more carotenoid-containing food sources as

defined by the USDA-NCC Carotenoid Database. **Table 2 s**hows the utilization of food access resources by food security status.

Impact of Food Access Resource Usage on Plasma, Skin, and Dietary Carotenoids

To determine the criterion-validity between biomarkers, associations between skin carotenoids, plasma carotenoids and carotenoids estimated by Diet ID™ were calculated (**Table 3**). Skin carotenoids scores and plasma carotenoids were strongly correlated ($p = 0.77$; $p < 0.0001$). Significant associations between total carotenoids from Diet ID™ to skin and plasma carotenoids were also observed ($\tau = 0.16$; $p = 0.007$; $\tau = 0.16$; $p = 0.005$, respectively). Associations were also observed between diet quality estimated by Diet ID™ and both SCS ($\tau = 0.22$; $p < 0.001$) and plasma carotenoids ($\tau = 0.18$; $p < 0.001$).

Analysis of covariance (ANCOVA) models demonstrated changes in skin carotenoid scores from pre to post were impacted by the interaction between of food access resource usage and food security status ($p = 0.002$). Mean group differences in plasma carotenoids and dietary carotenoids by time were not associated with the interaction of food security status and food access resources. The change in skin carotenoids was correlated to the change in plasma β-carotene ($p = 0.20$; $p = 0.02$), as the Veggie Meter® is most sensitive to the wavelength corresponding to β-carotene detection in the adipose.

Table 4 shows the interaction effect of food access resource usage and food security status between pre- and post-measurements for plasma, skin, and dietary carotenoids using univariate and multivariate linear regression models, controlling for demographic and socioeconomic characteristics. Of the biomarkers of interest, skin carotenoids elicited significant changes from pre to post from by the interaction of food security status and the frequency of food access resource usage ($Adj\ R^2 = 0.31$; $p = 0.001$). These results remained significant after

the models were adjusted for potential cofounding variables (*Adj* R^2 = 0.27; p = 0.006). This relationship was not observed for plasma carotenoids, dietary carotenoids, or diet quality.

Discussion

This clinical study followed a diverse population of university students over the academic term to determine the efficacy of on-campus food access resources to alleviate food insecurity and assess the impact on associated diet-related biomarkers of interest. The results indicated that skin carotenoid scores were significantly associated with the frequency of food access resources usage by food security status.

As skin carotenoid scores are reflective of approximately 30 days of intake, due to the prolonged incorporation and storage into epidermis and adipose tissue, this biomarker was able to elucidate changes in dietary intake across the academic term (weeks 4 – 8).[35] With the half-life of carotenoids in the plasma degrading after approximately 7 – 10 days, the use of the resources to acquire carotenoid-containing foods must have occurred during week 8 of the study to capture the incorporation of such foods in the diet.[36] Out of the 132 participants, only three individuals utilized the food access resources in the final week of study; thus, providing insight into the non-significant differences in the pre- to post-plasma carotenoid concentrations.

The use of a novel, image-based dietary assessment tool, Diet ID™, was implemented to reduce the researcher and participant challenges and inherent biases that may occur with traditional dietary data collection methods. Although correlated against both skin carotenoids scores and plasma carotenoids in this study, Diet ID™ may not yield the sensitivity required to detect minor, yet clinically relevant, changes in servings of F/V intake. The DQPN algorithm in Diet ID™ adheres to the human capacity for pattern recognition by capitalizing on sematic and episodic memory to identify overall dietary patterns, while avoiding the inconsistencies and

errors associated with dietary recalls.[30] As the primary outcome of the study was to identify potential changes in carotenoid intake, the incorporation of carotenoid-containing foods acquired through these resources may not have diversified overall dietary patterns, and therefore may not be represented in quantifiable changes in Diet ID™ output between timepoints.

As this was an observational study, participants were not instructed to alter habitual dietary intake, physical activity, or frequency of food access resource usage. The study duration of 8 weeks was determined *a priori* based on the physiological properties of dietary carotenoids during circulation, deposition, and accumulation in the blood and tissues.[37] Despite the appropriate time lapse to allow for incorporation of carotenoid compounds into the adipose tissue, dietary pattern trajectories are often established during childhood and continue into young adulthood, such that individuals who consumed diets rich in fruits, vegetables, whole-grains, and other nutrient dense foods are more likely to continue that pattern.[38] Due to the variation in dietary intake of free-living participants, incorporation of food items from the food access resources may have remained consistent with existing dietary patterns and average number of servings. Additional information on food-related behavior is needed to better elucidate usage strategies and accessibility priorities to establish the existence of compounding effects on dietary intake from food access resources. Further exploration into cognitive processes may determine if resources provide supplemental servings of carotenoid-containing foods in addition to habitual intake or whether fruits and vegetables acquired from food access resources are in lieu of self-purchasing.

To date, the evaluation of campus-affiliated food access resources on health-related outcomes has consisted of only subjective measures of self-reported general health status.[8] Although the General Self-Rate Health (SRH) question has been validated against objective

health measures, the strongest association is demonstrated with mental health and stress symptoms.[39, 40] The SRH allows for the capacity of large sample populations through survey-based dissemination, thus reducing the resource-intensive nature of collecting objective health-related biomarkers in the blood, tissue, or additional clinical assessments;[41] however, the non-specific assessment minimizes the ability to identify definitive diet-disease relationships.[39, 40, 42] Previous research has explored the impact of community-based food banks and food pantries on health-related biomarkers in low-income adult populations by assessing hemoglobin A1C and lipids panels,[43] barring that existing studies did not include the college student population nor the frequency of resource usage to determine the impact on diet-related outcomes. Due to the limited funding for both community and campus-based food access resources, the availability of nutrient-dense foods, inclusive of F/V, is limited.[44] Without additional research to evaluate the threshold effect for influencing clinically relevant nutrient biomarkers in the college student population and therefore justification for this target food group, the prospect of developing and expanding on-campus F/V food access resources is limited.

Contrary to the assumption of increased frequency of resource usage resulting in a greater magnitude of change to the biomarkers of interest, it is imperative to note that the food access resources were available to all participants, independent of food security status. It was observed that 73% of the study population accessed one or more of the resources in the duration of the study, indicating that individuals not experiencing food insecurity were still inclined to utilize food access avenues on campus. Previous findings have identified that first generation students and students further along in their academic careers are more likely to experience an increased risk of food insecurity.[45, 46] Similar associations among racial and ethnic groups have been observed, such that Latino(a)/Chicano(a)/Hispanic students are twice as likely to experience food

insecurity and utilize the on-campus food access resources more than food secure counterparts.[45] To better address the populations in need and reduce the identified barriers, such as stigmatization and cultural discordance, on-campus food access resources should evolve to include culturally relevant foods, foods that encompass dietary restrictions, and other considerations to promote utilization for improved diet-related health outcomes.[47, 48]

Strengths and Limitations

It is pertinent to acknowledge the strengths and limitations in the present study. To the authors knowledge, this study is of the first to evaluate the acute impact of food access resources on diet-related biomarkers in a diverse population of university students with varying food security statues using innovative and objective assessments of dietary intake.

Due to the unforeseen campus closure resulting from a surge in COVID-19 cases in January 2022, the original start date was postponed until the spring academic term (April – June 2022). This resulted in an abbreviated washout period, as students were only without access to the on-campus food access resources for one week corresponding to spring break, when operations are discontinued. Therefore, baseline values may have been elevated by carotenoid-containing food items consumed using the resources prior to the break. This shift in the start date may also impact the seasonality and price of carotenoid-containing produce, which was initially expected to be controlled for if the study was conducted in the winter months.[49]

As this study was observational on free-living subjects, participants were able to access resources independent of food security status. This type of study design limits the ability to determine a direct relationship. Although a randomized controlled trial may have the ability to elucidate the casual relationship between food access resources and diet-related biomarkers, it is not ethically appropriate to withhold such basic needs resources. If food access resource usage

was limited to only those experiencing low or very low food security status, it is plausible that the rates of FI would increase if access to resources was prohibited during the study. As observed in this study, many participants identifying as food secure still utilized the on-campus food access resources.

Conclusions

The findings from this study support the acute utilization of food access resources to improve diet-related biomarkers in a diverse population of university students. Improvements in skin carotenoid scores were reflective of the impact of carotenoid-containing foods provided by the food access resources. On-campus food access resources have the ability to improve food security status and increase the consumption of health-promoting foods for disproportionately vulnerable populations of emerging adults. Further research should explore the longitudinal impact of food access resources on student health outcomes to determine if prolonged, frequent use influences the reduction of chronic disease risk later in life.

References

1. Nikolaus CJ, An R, Ellison B, Nickols-Richardson SM. Food Insecurity among College Students in the United States: A Scoping Review. Adv Nutr 2020;11(2):327-348. doi:10.1093/advances/nmz111
2. United States Department of Agriculture. Definitions of Food Security 2021. Available from: https://www.ers.usda.gov/topics/food-nutrition-assistance/food-security-in-the-us/definitions-of-food-security/.
3. van Woerden I, Hruschka D, Bruening M. Food insecurity negatively impacts academic performance. Journal of Public Affairs 2019;19(3):e1864. doi:10.1002/pa.1864
4. Camelo K, Elliott M. Food Insecurity and Academic Achievement Among College Students at a Public University in the United States. Journal of College Student Development 2019;60(3):307-318. doi:10.1353/csd.2019.0028.
5. Weaver RR, Vaughn NA, Hendricks SP, McPherson-Myers PE, Jia Q, Willis SL, Rescigno KP. University student food insecurity and academic performance. J Am Coll Health 2020;68(7):727-733. doi:10.1080/07448481.2019.1600522
6. Martinez SM, Grandner MA, Nazmi A, Canedo ER, Ritchie LD. Pathways from Food Insecurity to Health Outcomes among California University Students. Nutrients 2019;11(6):1419. doi:10.3390/nu11061419
7. Meza A, Altman E, Martinez S, Leung CW. "It's a Feeling That One Is Not Worth Food": A Qualitative Study Exploring the Psychosocial Experience and Academic Consequences of Food Insecurity Among College Students. J Acad Nutr Diet 2019;119(10):1713-1721. doi:10.1016/j.jand.2018.09.006

8. Martinez SM, Chodur GM. Esaryk EE, Kaladijian S, Ritchie LD, Grandner M. Campus Food Pantry Use Is Linked to Better Health Among Public University Students. J Nutr Educ Behav 2022;54(6):491-498. doi:10.1016/j.jneb.2022.03.001

9. Neuhouser ML, Patterson RE, King IB, Horner NK, Lampe JW. Selected nutritional biomarkers predict diet quality. Public Health Nutr 2003;6(7):703-709. doi: 10.1079/phn2003486

10. Pelletier JE, Laska NM. Campus food and beverage purchases are associated with indicators of diet quality in college students living off campus. Am J Health Promot 2013;28(2):80-87. doi:10.4278/ajhp.120705-QUAN-326

11. United States Department of Agriculture and the United States Department of Health and Human Services. 2020 - 2025 Dietary Guidelines for Americans. 9th Edition. December 2020 [Accessed March 25, 2023]. Available at https://www.dietaryguidelines.gov/sites/default/files/2020-12/Dietary_Guidelines_for_Americans_2020-2025.pdf

12. Liu RH. Health-promoting components of fruits and vegetables in the diet. Adv Nutr 2013;4(3):384S-92S. doi:10.3945/an.112.003517

13. Amengual J. Bioactive Properties of Carotenoids in Human Health. Nutrients 2019;11(10):2388. doi:10.3390/nu11102388

14. Melendez-Martinez AJ, Mapelli-Brahm P, Benítez-González A, Stinco CM. A comprehensive review on the colorless carotenoids phytoene and phytofluene. Arch Biochem Biophys 2015;572:188-200. doi:10.1016/j.abb.2015.01.003

15. Thurmann PA, Steffen J, Zwernemann C, Aebischer CP, Cohn W, Wendt G, Schalch W. Plasma concentration response to drinks containing beta-carotene as carrot juice or

formulated as a water dispersible powder. Eur J Nutr 2002;41(5):228-235. doi:10.1007/s00394-002-0381-3

16. Radtke MD, Jilcott Pitts S, Jahns L, Firnhaber GC, Loofbourrow BM, Zeng A, Scherr RE. Criterion-Related Validity of Spectroscopy-Based Skin Carotenoid Measurements as a Proxy for Fruit and Vegetable Intake: A Systematic Review. Adv Nutr 2020;11(5):1282-1299. doi:10.1093/advances/nmaa054

17. Mounien L, Tourniaire F, Landrier JF. Anti-Obesity Effect of Carotenoids: Direct Impact on Adipose Tissue and Adipose Tissue-Driven Indirect Effects. Nutrients 2019;11(7):1562. doi:10.3390/nu11071562

18. Holt EW, Wei EK, Bennett N, Zhang LM. Low skin carotenoid concentration measured by resonance Raman spectroscopy is associated with metabolic syndrome in adults. Nutr Res 2014;34(10):821-826. doi:10.1016/j.nutres.2014.08.017

19. Moran NE, Mohn ES, Hason N, Erdman Jr JW, Johnson EJ. Intrinsic and Extrinsic Factors Impacting Absorption, Metabolism, and Health Effects of Dietary Carotenoids. Adv Nutr 2018;9(4):465-492. doi:10.1093/advances/nmy025

20. Melendez-Martinez AJ, Stinco CM, Mapelli-Brahm P. Skin Carotenoids in Public Health and Nutricosmetics: The Emerging Roles and Applications of the UV Radiation-Absorbing Colourless Carotenoids Phytoene and Phytofluene. Nutrients 2019;11(5):1093. doi:10.3390/nu11051093

21. Stahl W, Sies H. Carotenoids and protection against solar UV radiation. Skin Pharmacol Appl Skin Physiol 2002;15(5):291-296. doi:10.1159/000064532

22. Jilcott Pitts S, Moran NE, Laska MN, Wu Q, Harnack L, Moe S, et. al. Reflection Spectroscopy-Assessed Skin Carotenoids Are Sensitive to Change in Carotenoid Intake in a 6-Week Randomized Controlled Feeding Trial in a Racially/Ethnically Diverse Sample. J Nutr 2023;153(4):1133-1142. doi:10.1016/j.tjnut.2023.02.017

23. Radtke MD, Chodur GM, Bissell MCS, Kemp LC, Medici V, Steinberg FM, Scherr RE. Validation of Diet ID™ in Predicting Nutrient Intake Compared to Dietary Recalls, Skin Carotenoid Scores, and Plasma Carotenoids in University Students. Nutrients 2023;15(2):409. doi:10.3390/nu15020409

24. Qualtrics. 2005: Provo, Utah, USA.

25. Craft NE. High resolution HPLC method for the simultaneous analysis of carotenoids, retinoids, and tocopherols. FASEB 1996;10(3):A527.

26. Craft, N.E., Chromatographic Techniques for Carotenoid Separation. Curr Protoc Food Anal Chem 2001;00(1):F2.3.1-F2.3.15. doi:10.1002/0471142913.faf0203s00

27. CDC. Centers for Disease Control and Prevention. Laboratory Manual: Analysis of Vitamin A / Vitamin E / Carotenoids for NHANES.; Available online: https://www.cdc.gov/nchs/data/nhanes/nhanes_03_04/l45vit_c_met_vitAE_carotenoids.pdf. [Accessed May 16, 2023]

28. Ermakov IV, Gellermann W. Dermal carotenoid measurements via pressure mediated reflection spectroscopy. J Biophotonics 2012;5(7):559-570. doi: 10.1002/jbio.201100122

29. Radtke MD, Poe M, Stookey J, Jilcott Pitts S, Moran NE, Landry MJ, Rubin LP, Stage VC, Scherr RE. Recommendations for the Use of the Veggie Meter® for Spectroscopy-Based Skin Carotenoid Measurements in the Research Setting. Curr Dev Nutr 2021;5(8):nzab104. doi:10.1093/cdn/nzab104

30. Katz DL, Rhee LQ, Katz CS, Aronson DL, Frank GC, Cardner CD, Willett WC, Dansinger ML. Dietary assessment can be based on pattern recognition rather than recall. Med Hypotheses 2020;**140**:109644. doi:10.1016/j.mehy.2020.109644

31. Turner-McGrievy G, Hutto B, Bernhart JA, Wilson MJ. Comparison of the Diet ID™Platform to the Automated Self-administered 24-hour (ASA24) Dietary Assessment Tool for Assessment of Dietary Intake. J Am Nutr Assoc 2022;41(4):360-382. doi:10.1080/07315724.2021.1887775

32. Holder JM, Eldridge AL, Beecher GR, Buzzard IM, Bhagwat S, Davis CS, et al. Carotenoid Content of US Foods: An Update of the Database. Journal of Food Composition and Analysis 1999;**12(3)**:169-196. doi:10.1006/jfca.1999.0827

33. United States Department of Agriculture (USDA) E.R.S. Food Security in the U.S.: Measurement. Available online: https://www.ers.usda.gov/topics/food-nutrition-assistance/food-security-in-the-u-s/measurement/#measurement. (accessed on 28 September 2022).

34. Ermakov IV, Ermakova M, Sharifzadeh M, Gorusupudi A, Farnsworth K, Bernstein PS, et al. Optical assessment of skin carotenoid status as a biomarker of vegetable and fruit intake. Arch Biochem Biophys 2018;**646**:46-54. doi:10.1016/j.abb.2018.03.033

35. Darvin ME, Sterry W, Lademann J, Vergou T. The Role of Carotenoids in Human Skin. Molecules 2011;**16**(12):10491-10506. doi:10.3390/molecules161210491

36. Morgan EH, Graham ML, Marshall GA, Hanson KL, Seguin-Fowler RA. Serum carotenoids are strongly associated with dermal carotenoids but not self-reported fruit and vegetable intake among overweight and obese women. Int J Behav Nutr Phys Act 2019;**16**(1):104. doi:10.1186/s12966-019-0869-3

37. Furr HC, Clark RM. Intestinal absorption and tissue distribution of carotenoids. The Journal of Nutritional Biochemistry 1997;8(7):364-377. doi:10.1016/S0955-2863(97)00060-0

38. Appannah G, Murray K, Trapp G, Dymock M, Oddy WH, Ambrosini GL. Dietary pattern trajectories across adolescence and early adulthood and their associations with childhood and parental factors. Am J Clin Nutr 2021;113(1):36-46. doi:10.1093/ajcn/nqaa281

39. Wu S, Wang R, Zhao Y, Ma X, Wu M, Yan X, He J. The relationship between self-rated health and objective health status: a population-based study. BMC Public Health 2013;13:320. doi:10.1186/1471-2458-13-320

40. Hamplova D, Klusacek J, Mracek T. Assessment of self-rated health: The relative importance of physiological, mental, and socioeconomic factors. PLoS One 2022;17(4):e0267115. doi:10.1186/1471-2458-13-320

41. Garbarski D. Research in and Prospects for the Measurement of Health Using Self-Rated Health. Public Opin Q 2016;80(4):977-997. Doi:10.1093/poq/nfw033

42. Wuorela M, Lavonius S, Salminen M, Vahlberg T, Viitanen M, Viikari L. Self-rated health and objective health status as predictors of all-cause mortality among older people: a prospective study with a 5-, 10-, and 27-year follow-up. BMC Geriatr 2020;20(1):120. doi:10.1186/s12877-020-01516-9

43. Eicher-Miller HA. A review of the food security, diet and health outcomes of food pantry clients and the potential for their improvement through food pantry interventions in the United States. Physiol Behav 2020;220:112871. doi:10.1016/j.physbeh.2020.112871

44. Bazerghi C, McKay FH, Dunn M. The Role of Food Banks in Addressing Food Insecurity: A Systematic Review. J Community Health 2016. 41(4):732-40. doi: 10.1007/s10900-015-0147-5

45. Tanner Z, Loofbourrow BM, Chodur GM, Kemp L, Scherr RE. Food insecurity and utilization of campus food resources differ by demographic and academic group. Journal of Agriculture, Food Systems, and Community Development 2023;12(2):63-78. doi: 10.5304/jafscd.2023.122.018

46. Esaryk EE, Jiménez Arriaga EE, Kalaydjian S, Martinez SM. Campus Food Pantry Use Addresses a Gap Among California Public University Students. J Nutr Educ Behav 2021;53(11):921-930. doi: 10.1016/j.jneb.2021.06.005

47. El Zein A, Mathews AE, House L, Shelnutt KP. Why Are Hungry College Students Not Seeking Help? Predictors of and Barriers to Using an On-Campus Food Pantry. Nutrients 2018;10(9):1163. doi:10.3390/nu10091163

48. Wright KE, Lucero J, Crosbie E. "It's Nice to Have a Little Bit of Home, Even if It's Just on Your Plate" – Perceived Barriers for Latinos Accessing Food Pantries. Journal of Hunger & Environmental Nutrition 2019;15(4):496-513. doi: 10.1080/19320248.2019.1664963

49. Jahns L, Johnson LK, Conrad Z, Bukowski M, Raatz SK, Jilcott Pitts S, et. al. Concurrent validity of skin carotenoid status as a concentration biomarker of vegetable and fruit intake compared to multiple 24-h recalls and plasma carotenoid concentrations across one year: a cohort study. Nutr J 2019;18(1):78. doi:10.1186/s12937-019-0500-0

Tables and Figures

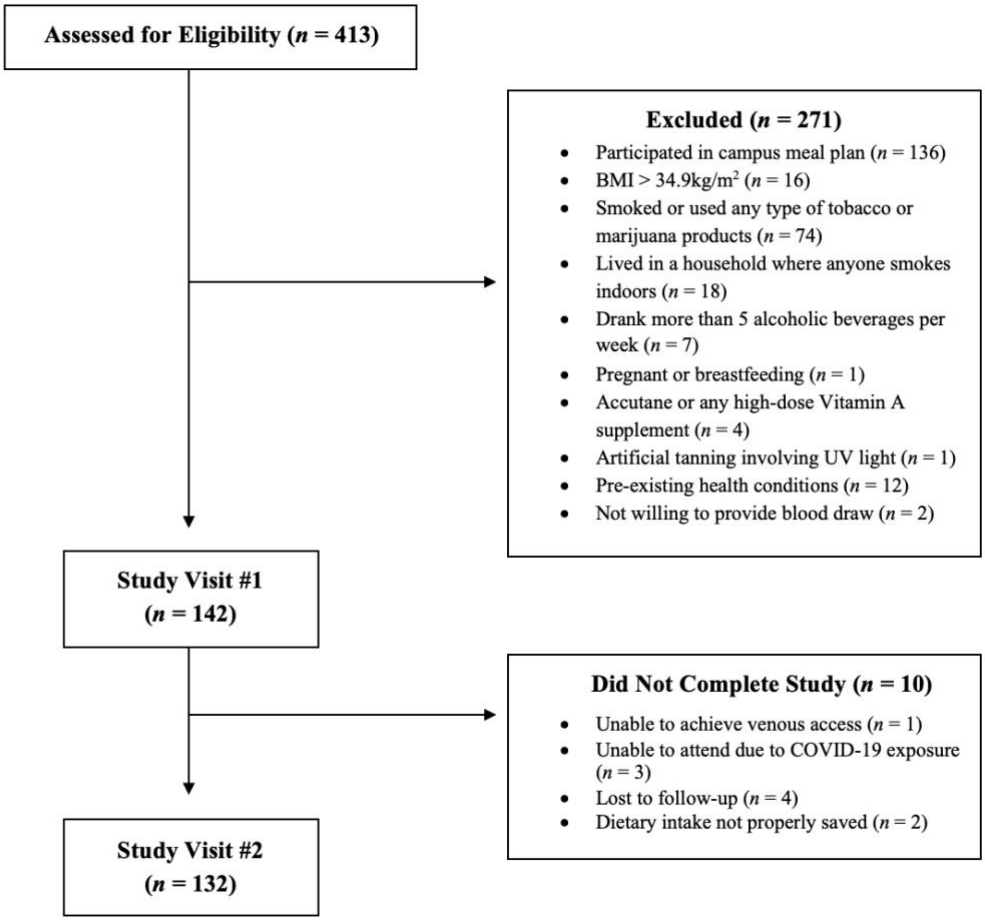


Figure 1. Flow diagram detailing inclusion and exclusion of participants.

Table 1. Participant characteristics expressed as mean ± standard deviations for age, BMI, and SCS, and the number and percentage of participants in subgroup by biological sex, academic standing, financial aid, first generation college student status, race/ethnicity, and food security status (n = 132).

Age, years (mean ± SD)	23.1 ± 4.0
Biological Sex	
Male	21 (15.9%)
Female	108 (81.8%)
Non-binary	3 (2.3%)
Academic Standing	
First Year	3 (2.3%)
Second Year	16 (12.1%)
Third Year	44 (33.3%)
Fourth Year	39 (29.5%)
Fifth Year +	3 (2.3%)
Graduate	27 (20.5%)
Financial Aid	
Yes	77 (58.3%)
No	55 (41.7%)
First-Generation College Student	
Yes	52 (39.4%)
Unsure	1 (0.8%)
No	79 (59.8%)
Race/Ethnicity	
African American/Black, not of Hispanic origin	2 (1.5%)
American Indian/Alaska native	0
Asian/Pacific Islander	64 (48.5%)
White, not of Hispanic origin	29 (22.0%)
Latin/Hispanic (Mexican-American, Puerto Rican, Cuban)	30 (22.7%)
Multi-racial	5 (3.8%)
Other	2 (1.5%)
Food Security Status	
High	69 (52.3%)
Marginal	29 (22.0%)
Low	21 (15.9%)
Very Low	13 (9.8%)
BMI (mean ± SD; kg/m^2)	
Timepoint 1	23.3 ± 4.4

Timepoint 2	23.2 ± 4.4
Skin Carotenoid Scores (mean ± SD)	
Timepoint 1	304.2 ± 113.1
Timepoint 2	319.3 ± 120.7[a]

[a] Mean skin carotenoid scores were significantly higher at timepoint 2 ($p < 0.001$)

Table 2. Number of participants who utilized food access resources weekly by food security status, ranging from a frequency of 0 – 8 visits (n = 132)

Frequency of Food Access Resource Use	Food Security Status			
	High Food Security (n = 69)	Marginal Food Security (n = 29)	Low Food Security (n = 21)	Very Low Food Security (n = 13)
0	21	7	4	4
1	13	3	2	1
2	4	1	1	1
3	1	1	3	0
4	7	4	3	1
5	6	5	2	4
6	10	4	6	2
7	5	3	0	0
8	2	1	0	0

Table 3. Associations between plasma carotenoids, skin carotenoid scores (SCS), and dietary carotenoid intake

	Correlation Coefficient	P-value
Plasma and SCS[a]	0.77	**p < 0.001**
Plasma and Dietary Carotenoids[b]	0.16	**p = 0.005**
SCS and Dietary Carotenoids[b]	0.16	**p = 0.007**
Plasma and Diet Quality[b]	0.22	**p < 0.001**
SCS and Diet Quality[b]	0.18	**p < 0.001**

[a] Calculated using Pearson's correlation coefficient (ρ). [b] Calculated using Kendall's tau (τ).

Table 4. Multivariable Linear Regression Models looking at the interaction between food security status and food access resource usage on pre- to post-changes to diet-related biomarkers

	Univariate Adjusted R^2	*Multivariate Adjusted R^{2*}*
Total Plasma Carotenoids	-0.015	-0.04
Skin Carotenoids	**0.31*** **	**0.27*** **
Total Dietary Carotenoids	-0.02	-0.06
HEI 2015-Score	-0.02	-0.02

Univariate Adjusted R^2 = from univariate models regressing the outcome by the use of food access resources; *Multivariate Adjusted R^{2*}* = from multivariable models regressing the outcome on the use of food access resources and potential confounders related to the outcome at $p < 0.10$ in correlations (see Table S1: plasma, scs: bmi; plasma:fss; plasma, scs: academic standing; plasma, scs:biological sex; plasma:fgss). *p < 0.05; ** p < 0.01; ***p < 0.001

Supplemental Table 1. Pearson's Correlations for Determining Covariates to include in the multivariate linear regression models

	Correlation Coefficient	P-value
Plasma and BMI	-0.31	**0.001**
SCS and BMI	-0.33	**0.001**
Plasma and FSS	-0.20	**0.001**
Plasma and academic standing	0.10	**0.097**
SCS and academic standing	0.16	**0.009**
Plasma and biological sex	0.l2	**< 0.05**
SCS and biological sex	0.13	**< 0.05**
Plasma and first-generation student status	-0.21	**< 0.001**

CHAPTER 5

Interrelationship between Executive Function, Food Insecurity, and Diet Quality in a Diverse Population of College Students

Introduction

Food insecurity (FI) is defined as the limited or uncertain access to adequate quality, desirability, variety, or quantity of food in the diet and can be either acute or chronic.[1] Food insecurity and the underconsumption of essential macronutrients, vitamins, minerals, and phytonutrients may negatively impact brain development, neural connectivity, and executive function (EF).[2] Executive function encompasses a myriad of behavioral and cognitive attributes that influence decision making, learning, and memory, among other neuropsychological processes.[3] Concentrated in the frontal lobe region of the brain, EF processes continue to develop and mature through adolescence and into early adulthood.[4]

College students are a subpopulation of emerging adults that are still experiencing cognitive development. In the United States, college students experience disproportionate rates of FI compared to the national average.[5, 6] This has implications for a reduced quality diet, which may be accompanied with the experience of overnutrition, undernutrition, or both conditions manifesting simultaneously. Overnutrition may occur when the experience of FI yields the consumption of calorically dense, nutrient poor foods, increasing the risk for overweight or obesity, hypertension, and type 2 diabetes, among other diet-related chronic diseases.[7] Contrarily, undernutrition may occur when either calorie or micronutrient needs are not achieved due to a reduction in the amount or quality of food being consumed.[8] Additionally, the co-occurrence of both under and overnutrition has been observed in the experience of adequate caloric intake, but essential, regulatory micronutrients not being consumed in sufficient quantity to meet requirements.[9] All three dietary outcomes may affect cognitive development and maturation, along with EF processes in college students experiencing FI.

To date, the relationship between food insecurity and cognition has been predominately explored in older adults and children. Food insecurity later in the lifespan has been associated with accelerated cognitive decline and memory loss,[10, 11] whereas FI during childhood has been shown to delay cognitive development and increase the risk of behavioral problems.[12, 13] A recent study exploring functional state connectivity in college students found significant differences in neural connectivity among groups of differing food security status, in addition to differences in executive function using the self-report Behavior Rating Inventory of Executive Function-2 Adult Version (BRIEF-2A).[14] Discrepancies between self-reported and objective measures of EF have been observed with regard to financial behaviors and overall wellbeing.[15] Therefore, assessing EF processes in college students experiencing FI using objective assessment tools is warranted.

Objective measures of executive function, such as the CANTAB assessment tool, have been implemented in populations across the lifespan, and uniquely distinct characterisitics have emerged in the college-aged individuals 20-29 years of age.[16] Executive function, with regard to strategic planning and goal-oriented behaviors were heightened in individuals 20-29 years, emphasizing the developmental attainment of maximal short-term memory capacity during the early twenties.[16] Despite the knowledge of the crucial maturation of EF processes in this age range, there is limited research in this specific population, with a emphasis on the lack of research in univeristy students under various life stressors, such as the experience of FI.

The aim of the present study was to explore the interrelationship between EF, food security status, and diet quality in a diverse population of college students. It was hypothesized that students experiencing FI will have poorer indications of EF processing, which may

subsequently affect dietary behaviors and diet quality due to increased impulsivity, and decreased planning capabilities, among other affected domains of cognition.

Methods

Prior to study commencement, all protocols and procedures were reviewed and approved by the Institutional Review Board at the University of California, Davis (IRB:1702801).

Participants

This cross-sectional study was conducted online in May 2021 – November 2021 using virtual assessment modalities. Participants were recruited using a rolling admission process from the University of California, Davis through the SONA Research Participation System. The SONA system is a platform that allows students to sign up for campus affiliated research opportunities. Students using this system must login using UC Davis credentials. Interested students were screened online using a Qualtrics survey to determine eligibility based on inclusion and exclusion criteria (**Table 1**). Once eligibility was established, participants received an instructional video detailing the procedure, followed by individualized, encrypted links to the study protocol to be completed online.

Executive Function

The imperative components of EF, such as time and quality of decision-making, ability to multi-task effectively, and capacity for acute or long-range planning were analyzed using various tasks in the CANTAB battery (Cambridge Cognition, Bottisham Cambridgeshire, UK).

Cambridge Gambling Task (CGT): The Cambridge Gambling Task (CGT) was used to determine the decision-making time and decision-making quality, as well as risk adjustment and impulsivity, as it pertains to diet quality and eating behaviors.

Delayed Matching to Sample (DMS): The Delayed Matching to Sample (DMS) was used to evaluate the probability of an error after a correct or incorrect responses through the simulation of forced decision making.

Intra-Extra Dimensionality Shift Test (IED): The Intra-Extra Dimensionality Shift Test (IED) was used to examine rule acquisition and reversal to determine attentional shift to mismatched stimuli.

One Touch Stockings of Cambridge (OTS): The One-Touch Stockings of Cambridge Test (OTS) was used to determine efficiency in frontal cortex strategic long-range planning processes.

Rapid Visual Information Processing (RVP): The Rapid Visual Information Processing (RVP) was used to examine response latency in the theory of choice overload, a phenomenon resulting from difficulty making decisions due to too many choices presented.

Dietary Intake

Diet patterns and nutrient intake were assessed using Diet ID™. Diet ID™ utilizes Diet Quality Photo Navigation (DQPN) to predict dietary intake using the Nutrition Data System for Research (NDSR) database (Version 2017). Diet ID™ estimates diet quality using the Healthy Eating Index (HEI-2015 Score) determined by the Dietary Guidelines for Americans 2020 – 2025 for the over- or underconsumption of select nutrients and food groups. Diet quality and nutrient intake from Diet ID™ has been correlated against nutrient values from 24-hour NDSR dietary recalls and food Frequency Questionnaires (FFQs), as well as objective measures of dietary intake in the plasma and skin.[17, 18]

Stress and Adverse Childhood Experiences

Stress levels were evaluated using the Cohen's Perceived Stress Scale (PSS).[19] The PSS is a psychological assessment tool that measures the perception of stress in an individual's life over

the past month (30 days). The following ranges were used to classify stress levels: 0 – 13 = low stress; 14 – 26 = moderate stress; 27 – 40 = high stress. In addition to quantifying stress level, the PSS also predicts the probability of increased risk of disease onset due to high levels of stress. The Adverse Childhood Experiences (ACEs) questionnaire was administered to measure childhood trauma.[20] This survey assesses various components of emotional and physical violence, substance abuse, food insecurity, and other acute or chronic stressors in early life. A score of 4 or more affirmative answers are considered clinically significant.

Food Security Status

The United States Department of Agriculture (USDA) 10-item food security checklist was administered to participants to determine individual food security status.[21] The following classifications were used: 0: high food security; 1 – 2: marginal food security; 3 – 5: low food security; 6 – 10: very low food security.

Anthropometrics

Anthropometric data included self-reported height and weight to calculate body mass index (BMI; kg/m^2).

Demographic Information

Information regarding participant sex, gender, race/ethnicity, socioeconomic status, academic status, and other demographic information pertinent to the analysis was requested. This also included information on the usage of state or federal government supported food assistance programs, such as CalFresh, known federally as the Supplemental Nutrition Assistance Program (SNAP).

Statistical Analysis

Statistical analyses were performed using Stata v13 (StataCorp, College Station, TX, USA). Data were inspected for normality using Shapiro Wilks and adjusted using logarithmic transformations when necessary. Descriptive data on participant characteristics are expressed as absolute (n) and relative frequencies (%) for categorical variables and mean +/- standard deviation (SD) for continuous variables. Key outcomes for each EF domain were determined by analytical guidance from Cambridge Cognition.[22, 23]

Independent sample t-tests were conducted to determine differences in dietary intake, perceived stress, adverse childhood experiences, and EF output by food security status. To explore the direct relationships between EF and potential covariates, Pearson's and Kendall's Tau correlations were constructed for BMI, FSS, stress, ACEs, diet quality, and nutrients associated with EF. For each domain of the CANTAB battery, multivariate linear regression were performed to test the association between food security status and CANTAB outcome, controlling for covariates identified by correlations ($p < 0.10$). Moderation by the interaction of both PSS and ACEs with FSS was tested for each regression. Statistical significance was considered with $p < 0.05$ level of probability after correction for multiple comparisons.

Results

Following screening and exclusion (n = 367), a total of 230 participants completed all study components (**Figure 1**). Sociodemographic characteristics, BMI, dietary intake, perceived stress, and ACE scores are presented in **Table 2.** Participants were predominately female (84%), with a mean age of 22.2 ± 1.5 years. The racial and ethnic diversity of the population comprised of African American/Black (2%), American Indian/Alaskan Native (3%), Asian/Pacific Islander

(56%), White (36%), multi-racial (1%), and other (3%), of whom 23% of the total population identifying as Hispanic, Latinx, or Spanish Origin. Participants on average were in the normal to overweight BMI classifications, with a mean BMI of 23.5 ± 4.6 kg/m^2. Participants ranged in food security status, with 45% classified as having high food security, 25% marginal food security, 16% low food security, and 15% very low food security.

Approximately 19% of the participants experienced clinically relevant adverse childhood experiences, and 84% of the population had moderate (70%) to high (14%) levels of stress. Stress levels and adverse childhood experiences were significantly higher in the very low food security group compared to their food secure counterparts (**Table 3**). Diet quality, in accordance with the Healthy Eating Index 2015, was an average of 71.2 ± 22.9. Although diet quality was trending lower in the very low food security status group, differences were approaching but did not reach statistical significance ($p = 0.06$). Other nutrients of interest that may impact EF output were explored, including carotenoids (from fruit and vegetables), saturated fat, sodium, added sugars, and macronutrients, among others. Significant differences in nutrient intake were observed by food security status for sodium, such that individuals experiencing very low food security had higher sodium intake compared to individuals with high food security ($p < 0.05$). Protein intake was significantly higher in the low food secure group when compared to the food secure group ($p < 0.05$). No other selected nutrients of interest were significantly different by food security status.

Differences in mean scores for each domain of EF was compared by food security status. Significant differences were observed in the CGT, reflecting impulsivity. It was observed that individuals experiencing low food security were significantly more impulsive compared to all

other groups ($p < 0.05$ for all). In the IED assessment, total trials and total errors were significantly higher for the marginally food secure and low food secure groups compared to the high food secure group, reflecting poorer flexibility with stimuli integration and informed decision making ($p = 0.03$ for both). The RVP assessment demonstrated increased sensitivity to a target sequence for the food secure group compared to the marginal and low food secure groups ($p = 0.02$ and $p = 0.04$, respectively). No significant differences by food security status were observed with outcomes from the OTS and DMS tests.

Correlations between EF and potential covariates were explored to construct appropriate linear regression models. Body mass index was negatively correlated with outputs for all domains of EF; however, body mass index was not significantly difference between individuals who were FS and FI. After adjusting for biological sex, BMI, stress, and adverse childhood experiences, multivariate linear regressions of EF outcomes as the independent variable found significant weak associations for DMS, which food security status explained 4% of the variance, and with RVP, which food security status accounted for 7% of the variance (**Table 5**).

Discussion

An assortment of CANTAB Battery tasks were used to assess differences in EF by food security status to determine if cognitive behavioral traits are associated with diet quality. The CANTAB Battery has been previously validated for use in typically cognitive functioning adult populations, with notable differences by age and cognitive disease status.[24] As food security status was the primary group identifer in this study, EF outcomes by group were expected to be more conservative than age or disease status. Using EF output from the CANTAB assessments relating to impulsive tendencies, risk taking behaviors, and response latency, this study evaluated

different behavioral charactersitics in the college student population that may be impacted by the experience of FI.

Consistent with previous literature, stress levels and advserse childhood experiences were significantly higher in the individuals experiencing very low food security.[25-27] The experience of FI initiates stressful thought patterns that often correlate with the idea of worrying about how to secure a next meal. In addition to mental stress, the experience of FI may also result in physiological stress, where bouts of starvation result in the increased levels of C-reactive protein (CRP), a biomarker of systemic inflammation.[28] Adverse childhood experiences have been closely associated with stress, mental illness, and chronic disease development into adulthood.[29, 30] Such experiences during childhood increases risk factors for food insecurity, which can continue to perpetuate stress levels as emerging adults, consequently impacting EF.

Specific nutrients have been demonstrated to impact EF outcomes.[31] Identified as crucial to brain development in infants, toddlers, and children, cognitive maturation is still occurring in young adults, thus similar nutrients may also impact the college student population.[32, 33] Carotenoids, specifically lutein and zeaxanthin, B-vitamins, and iron are among the nutrients for anatomical brain development in utero and into infancy; however, macronutrients, including protein and fat have demonstrated emergent impact of EF following puberty.[34] Nutrients to negatively impact cognition have also been identified, corresponding to many of the nutrients of concern listed in the Dietary Guidelines for Americans, including sodium and added suagrs.[35-37] As shown in this study, food insecurity was associated with a trend toward reduced diet quality, and significantly increased consumption of sodium. Such dietary patterns may result in poorer EF outcomes compared to individuals experiencing food security with elevated HEI-scores.

Executive function scores from the DMS and RVP were partially explained by the incoporation of food security, when biological sex, BMI, stress, and adverse childhood experiences were incorporated into the model. The main outcomes of the DMS test corresponds to components of memory and planning capabilities. The capacity to effectively plan may be impacted in the experience of FI, such that financial budgeting behaviors for food purchasing are underdeveloped.[38] The RVP tests for speed of response, time to decision, and the probability of false alarms under multiple mismatched stimuli. This provides insight to impulsive behaviors and rushed reponsivity that may occur in emerging adults experiencing FI, where dietary behaviors are driven by external factors, such a price or convenience, potentially contributing to binge eating experiences.[39]

Several domains of EF achieved statisitically significant differences by food security status. As the population consisted of relatively healthy, cognitively-abled college students, these differences highlight the considerable affect of FI on cognitive outcomes. As food security status was assessed over the last 30 days, the acute experience of FI was reflected. As there is limited research on the duration and severity of FI on the development of mental illness and other related cognitive outcomes, results from this study found that FI prevalence over the last 30 days was indicative of cognitive differences. Exploration into the differences between acute and chronic FI may provide more insight on the physiological mechanisms altering EF, which will further elucidate the understanding of cognitive-related behaviors influencing dietary outcomes.

Limitations

It is important to identify the limitations of the present study. As this study was conducted during the campus closure of the COVID-19 pandemic, the entirety of data collection occurred online. There are notable benefits from online survey adminstation, including existing evidence that

participants may be more likely to share personal information without the fear of the privacy paradox.[40] The privacy paradox refers to the discordance between attitudes and behaviors, which is often associated with the desire to mask financial challenges, such as in the experience of FI; therefore, the migration of this questionnaire to be administered online may have diminished the issue of personal information remaining private. Although collecting data in an in-person setting may alleviate aspects of other participant self-reported biases, including BMI, self-reported height and weight in young adults often remains statistically significant when correlated to objectively measured BMI.[41] Participants were instructed to remove any object that may cause visual or auditory distractions during the CANTAB assessment, including cellphones, computers, and other technological devices, as well as to take the assessment in a quiet, private room. As this was up to the discretion of the participants, some individuals may have been distracted using the assessment period.

Stress and adverse childhood experiences were assessed using validated questionnaires for adults. Literature also suggests the clinical manifestation of other depression or anxiety-based mental health conditions may impact EF. Although the use of medication for any diagnosed depression or anxiety diagnoses was assessed, validated questionnaires for undiagnosed mental health concerns, such as the Generalized Anxiety Disorder (GAD), were not implemented.[42] To minimize the time burden of excessive assessments for all components of mental, physical, and dietary health outcomes, the omission of such assessments may have resulted in not accounting for participants experiencing acute, undiagnosed depression or anxiety.

Conclusions

Cumulative acute and chronic life experiences, such as food insecurity, may perpetuate negative phsyiological and cognitive health. This study sought to elucidate the relationship between EF,

food insecurity, and diet quality in college students, which had yet to be explored. Differences in EF domains by food security status were observed, providing insight to the potential for detrimental cognitive impacts of FI that develop in emerging adulthood and can perpetuate across the lifespan. These findings support efforts to improve access to nutrient-dense foods in this population to increase food security and reduce the risk for acute and lasting cognitive impairments.

References

1. United States Department of Agriculture. Definitions of Food Security 2021 [Accessed March 25, 2023]. Available at https://www.ers.usda.gov/topics/food-nutrition-assistance/food-security-in-the-us/definitions-of-food-security/.
2. Royer MF, Guerithault N, Braden BB, Laska MN, Bruening M. Food Insecurity Is Associated with Cognitive Function: A Systematic Review of Findings across the Life Course. International Journal of Translational Medicine 2021;1(3):205-222. doi:10.3390/ijtm1030015
3. Baggetta P, Alexander PA. Conceptualization and Operationalization of Executive Function. Mind, Brain, and Education 2016;10(1):10-33. doi:10.1111/mbe.12100
4. Arain M, Haque M, Johal L, Mathur P, Nel W, Rais W, Sandhu R, Sharma S. Maturation of the adolescent brain. Neuropsychiatr Dis Treat 2013;9:449-61. doi:10.2147/NDT.S39776
5. Nikolaus CJ, An R, Ellison B, Nickols-Richardson SM. Food Insecurity among College Students in the United States: A Scoping Review. Adv Nutr 2020;11(2):327-348. doi:10.1093/advances/nmz111
6. United States Department of Agriculture Economic Research Service. Key Statistics & Graphics - Food Security Status of U.S. Households in 2021 [Accessed March 25, 2023]. Available at https://www.ers.usda.gov/topics/food-nutrition-assistance/food-security-in-the-u-s/key-statistics-graphics/#foodsecure
7. Laraia BA. Food insecurity and chronic disease. Adv Nutr 2013;4(2):203-12. doi:10.3945/an.112.003277

8. Lopes SO, Abrantes LCS, Azevedo FM, de Souza de Morais N, de Castro Morais D, Gonçalves VSS, et al. Food Insecurity and Micronutrient Deficiency in Adults: A Systematic Review and Meta-Analysis. Nutrients 2023;15(5):1074. doi:10.3390/nu15051074

9. Tanumihardjo SA, Anderson C, Kaufer-Horwitz M, Bode L, Emenaker NJ, Haqq AM, Satia JA, Silver HJ, Stadler DD. Poverty, obesity, and malnutrition: an international perspective recognizing the paradox. J Am Diet Assoc 2007;107(11):1966-72. doi:10.1016/j.jada.2007.08.007

10. Wong JC, Scott T, Wilde P, Li Y, Tucker KL, Gao X. Food Insecurity Is Associated with Subsequent Cognitive Decline in the Boston Puerto Rican Health Study. J Nutr 2016;146(9):1740-5. doi:10.3945/jn.115.228700

11. Na M, Dou N, Brown MJ, Chen-Edinboroo LP, Anderson LR, Wennberg A. Food Insufficiency, Supplemental Nutrition Assistance Program (SNAP) Status, and 9-Year Trajectory of Cognitive Function in Older Adults: The Longitudinal National Health and Aging Trends Study, 2012-2020. J Nutr 2023;153(1):312-321. doi:10.1016/j.tjnut.2022.12.012

12. Hobbs S, King C. The Unequal Impact of Food Insecurity on Cognitive and Behavioral Outcomes Among 5-Year-Old Urban Children. J Nutr Educ Behav 2018;50(7):687-694. doi:10.1016/j.jneb.2018.04.003

13. Bryant LM, Eicher-Miller HA, Korucu I, Schmitt SA. Associations between Subjective and Objective Measures of the Community Food Environment and Executive Function in Early Childhood. Nutrients 2020;12(7):1944. doi:10.3390/nu12071944

14. Guerithault N, McClure SM, Ojinnaka CO, Braden BB, Bruening M. Resting-State Functional Connectivity Differences in College Students with and without Food Insecurity. Nutrients 2022;14(10):2064. doi:10.3390/nu14102064

15. Strömbäck C, Skagerlund K, Västfjäll, Tinghög G. Subjective self-control but not objective measures of executive functions predicts financial behavior and well-being. Journal of Behavioral and Experimental Finance 2020;27:100339. doi:10.1016/j.jbef.2020.100339

16. De Luca CR, Wood SJ, Anderson V, Buchanan J, Proffitt Tm, Mahony K, Pantelis C. Normative data from the CANTAB. I: development of executive function over the lifespan. J Clin Exp Neuropsychol 2003;25(2):242-54. doi:10.1076/jcen.25.2.242.13639

17. Radtke MD, Chodur GM, Bissell MCS, Kemp LC, Medici V, Steinberg FM, Scherr RE. Validation of Diet ID™ in Predicting Nutrient Intake Compared to Dietary Recalls, Skin Carotenoid Scores, and Plasma Carotenoids in University Students. Nutrients 2023;15(2):409. doi:10.3390/nu15020409

18. Turner-McGrievy G, Hutto B, Bernhart JA, Wilson MJ. Comparison of the Diet ID™ Platform to the Automated Self-administered 24-hour (ASA24) Dietary Assessment Tool for Assessment of Dietary Intake. J Am Nutr Assoc 2022;41(4):360-382. doi:0.1080/07315724.2021.1887775

19. Cohen S, Kamarck T, Mermelstein R. Perceived Stress Scale. Mind Garden 1994.

20. Hardt J, Rutter M. Validity of adult retrospective reports of adverse childhood experiences: review of the evidence. J Child Psychol Psychiatry 2004;45(2):260-73. doi:10.1111/j.1469-7610.2004.00218.x

21. United States Department of Agriculture (USDA) E.R.S. Food Security in the U.S.: Measurement. Available online: https://www.ers.usda.gov/topics/food-nutrition-assistance/food-security-in-the-u-s/measurement/#measurement. (accessed on 16 May 2023).

22. Robbins TW, James M, Owen AM, Sahakian BJ, Lawrence AD, McInnes L, Rabbitt PM. A study of performance on tests from the CANTAB battery sensitive to frontal lobe dysfunction in a large sample of normal volunteers: implications for theories of executive functioning and cognitive aging. Cambridge Neuropsychological Test Automated Battery. J Int Neuropsychol Soc 1998;4(5):474-90. doi:10.1017/s1355617798455073

23. Robbins TW, James M, Owen AM, Sahakian BJ, McInnes L, Rabbitt P. Cambridge Neuropsychological Test Automated Battery (CANTAB): a factor analytic study of a large sample of normal elderly volunteers. Dementia 1994;5(5):266-81. doi:10.1159/000106735

24. Smith PJ, Need AC, Cirulli ET, Chiba-Falek O, Attix DK. A comparison of the Cambridge Automated Neuropsychological Test Battery (CANTAB) with "traditional" neuropsychological testing instruments. J Clin Exp Neuropsychol 2013;35(3):319-28. doi:10.1080/13803395.2013.771618

25. Pourmotabbed A, Moradi S, Babaei A, Ghavami A, Mohammadi H, Jalili C, Symonds ME, Miraghajani M. Food insecurity and mental health: a systematic review and meta-analysis. Public Health Nutr 2020;23(10):1778-1790. doi:10.1017/S136898001900435X

26. Martin MS, Maddocks E, Chen Y, Gilman SE, Colman I. Food insecurity and mental illness: disproportionate impacts in the context of perceived stress and social isolation. Public Health 2016;132:86-91. doi:10.1016/j.puhe.2015.11.014

27. Royer MF, Ojinnaka CO, Zhang X, Thornton AG, Blackhorse K, Bruening M. Food insecurity and adverse childhood experiences: a systematic review. Nutr Rev 2022;80(10):2089-2099. doi:10.1093/nutrit/nuac029

28. Gowda C, Hadley C, Aiello AE. The association between food insecurity and inflammation in the US adult population. Am J Public Health 2012;102(8):1579-86. doi:10.2105/AJPH.2011.300551

29. Danese A, McEwen BS. Adverse childhood experiences, allostasis, allostatic load, and age-related disease. Physiol Behav 2012;106(1):29-39. doi:10.1016/j.physbeh.2011.08.019

30. Kalmakis KA, Chandler GE. Health consequences of adverse childhood experiences: a systematic review. J Am Assoc Nurse Pract 2015;27(8):457-65. doi:10.1002/2327-6924.12215

31. Gomez-Pinilla F. Brain foods: the effects of nutrients on brain function. Nat Rev Neurosci 2008;9(7):568-78. doi:10.1038/nrn2421

32. Franklyn N, Kesavelu D, Joji P, Verma R, Wadhwa A, Ray C. Impact of Key Nutrients on Brain and Executive Function Development in Infants and Toddlers: A Narrative Review. Journal of Food and Nutrition Sciences 2022;10(1):19-26. doi:10.11648/j.jfns.20221001.14

33. Costello SE, Geiser E, Schneider N. Nutrients for executive function development and related brain connectivity in school-aged children. Nutr Rev 2021;79(12):1293-1306. doi:10.1093/nutrit/nuaa134

34. van der Wurff ISM, Meyer BJ, de Groot RHM. Effect of Omega-3 Long Chain Polyunsaturated Fatty Acids (n-3 LCPUFA) Supplementation on Cognition in Children

and Adolescents: A Systematic Literature Review with a Focus on n-3 LCPUFA Blood Values and Dose of DHA and EPA. Nutrients 2020;12(10):3115. doi:10.3390/nu12103115

35. United States Department of Agriculture and the United States Department of Health and Human Services. 2020 - 2025 Dietary Guidelines for Americans. 9th Edition. December 2020 [Accessed March 25, 2023]. Available at https://www.dietaryguidelines.gov/sites/default/files/2020-12/Dietary_Guidelines_for_Americans_2020-2025.pdf

36. Fiocco AJ, Shatenstein B, Ferland G, Payette H, Belleville S, Kergoat MJ, Morais JA, Greenwood CE. Sodium intake and physical activity impact cognitive maintenance in older adults: the NuAge Study. Neurobiol Aging 2012;33(4):829 e21-8. doi:10.1016/j.neurobiolaging.2011.07.004

37. Muth AK, Park SQ. The impact of dietary macronutrient intake on cognitive function and the brain. Clin Nutr 2021;40(6):3999-4010. doi:10.1016/j.clnu.2021.04.043

38. Coffino JA, Udo T, Hormes JM. Nudging while online grocery shopping: A randomized feasibility trial to enhance nutrition in individuals with food insecurity. Appetite 2020;152:104714. doi:10.1016/j.appet.2020.104714

39. Rook DW, Fisher RJ. Normative Influences on Impulsive Buying Behavior. Journal of Consumer Research 1995;22(3):305-313. doi:https://doi.org/10.1086/209452

40. Kokolakis S. Privacy attitudes and privacy behaviour: A review of current research on the privacy paradox phenomenon. Computers & Security 2017;64:122-134. doi:10.1016/j.cose.2015.07.002

41. Olfert MD, Barr ML, Charlier CM, Famodu OA, Zhou W, Mathews AE, Byrd-Bredbenner C, Colby SE. Self-Reported vs. Measured Height, Weight, and BMI in Young Adults. Int J Environ Res Public Health 2018;15(10):2216. doi:10.3390/ijerph15102216

42. Spitzer RL, Kroenke K, Williams JBW, Löwe B. A brief measure for assessing generalized anxiety disorder: the GAD-7. Arch Intern Med 2006;166(10):1092-7. doi:10.1001/archinte.166.10.1092

Tables and Figures

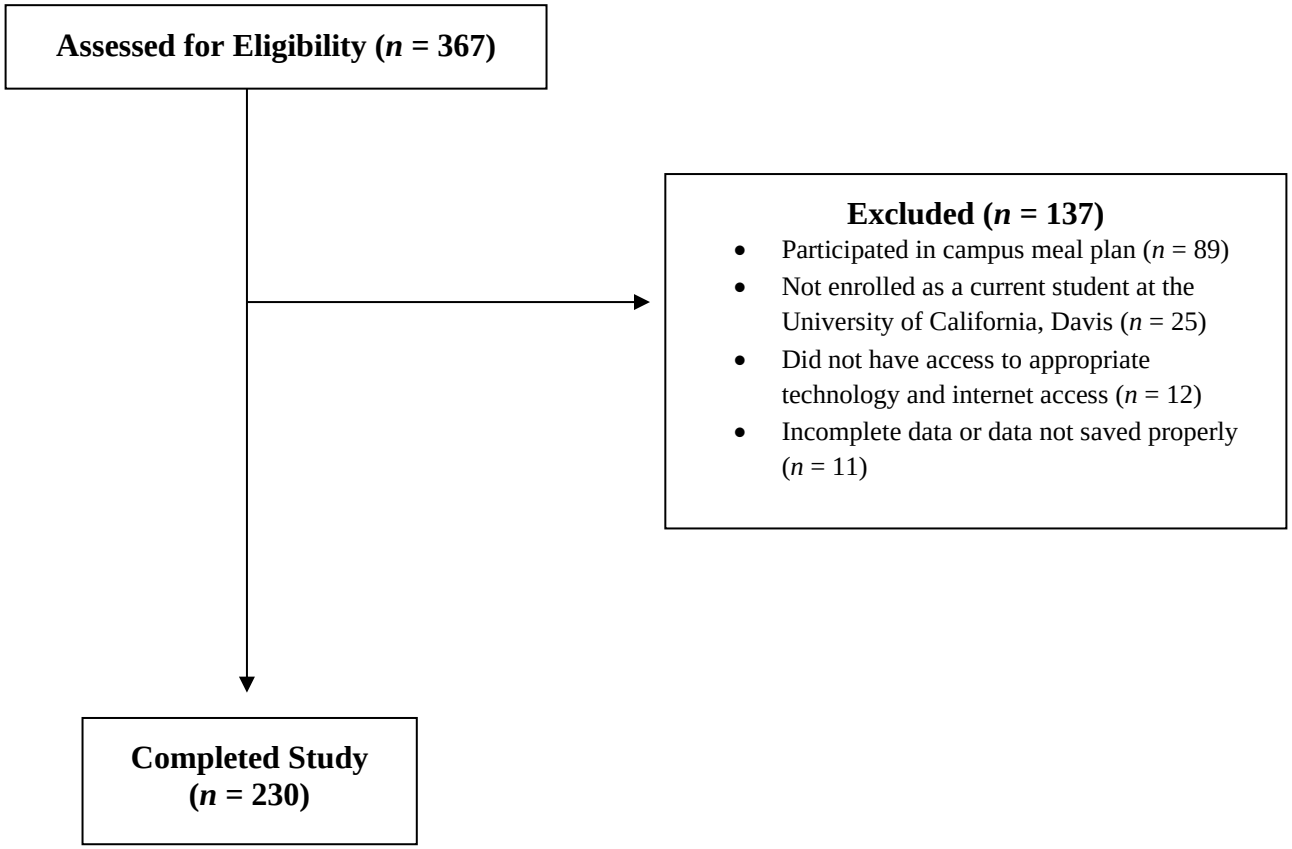


Figure 1. CONSORT flow diagram detailing participant eligibility

Table 1. Inclusion criteria for participant enrollment defined *a priori*

Inclusion Criteria	Description
Student Status	Enrolled as an undergraduate student (18 years or older) at the University of California, Davis (minimum 6 units)
Technology	Access to high-speed internet; Updated computer processing software
Meal Plan Use	Does not participate in a provided meal plan, such as the Dining Commons at the University of California, Davis
Vision Status	Ability to complete the visual portion of the CANTAB assessment and virtual questionnaires

Table 2. Participant characteristics expressed as mean ± standard deviations for age and BMI, and the number and percentage of participants in subgroup by biological sex, academic standing, financial aid, first generation college student status, race/ethnicity, and food security status (*n* = 230).

Age, years (mean ± SD)	22.2 ± 1.5
Biological Sex	
Male	30 (13%)
Female	193 (84%)
Non-binary	5 (2%)
Prefer to self-describe/other	2 (1%)
Ethnicity	
Hispanic, Latinx, or Spanish Origin	52 (23%)
Not of Hispanic, Latinx, or Spanish origin	178 (77%)
Race	
African American/Black	2 (1%)
American Indian/Alaska native	7 (3%)
Asian/Pacific Islander	129 (56%)
White	82 (36%)
Multi-racial	3 (1%)
Other	7 (3%)
Food Security Status	
High	103 (45%)
Marginal	57 (25%)
Low	36 (15%)
Very Low	34 (15%)
BMI (mean ± SD; kg/m^2)	23.4 ± 4.6
Diet Quality (HEI-2015)	71.2 ± 22.9
Perceived Stress Scale	
Low	36 (16%)
Moderate	162 (70%)
High	32 (14%)
Adverse Childhood Experiences Survey	
ACE Scores (> 4)	44 (19%)

Table 3. Mean values by food security status for diet quality, nutrients of intake, perceived stress, and adverse childhood experiences (n=230)

	Total Sample (n = 230)	High Food Security (n = 103)	Marginal Food Security (n = 57)	Low Food Security (n = 36)	Very Low Food Security (n = 34)
Diet Quality and Select Nutrient Intake					
HEI-Score	71.2 ± 22.9	73.1 ± 23.1	69.5 ± 21.9	74.6 ± 21.8	64.4 ± 24.4
Carotenoids	18187.3 ± 14146.9	18442.4 ± 14305.0	17913.7 ± 13152.9	19900.6 ± 14982.3	16059.1 ± 14870.3
Added Sugars	33.5 ± 30.4	30.8 ± 29.6	37.0 ± 32.3	29.7 ± 28.7	40.0 ± 31.3
Sodium	2683.0 ± 975.9	2536.6 ± 973.6[a]	2784 ± 1054.8	2716.2 ± 849.3	2922.9 ± 938.2[b]
Carbohydrates	249.4 ± 64.6	243.3 ± 63.8	252.3 ± 64.5	260.1 ± 61.7	251.3 ± 70.7
Fiber	31.1 ± 18.1	32.1 ± 18.8	29.2 ± 16.8	34.2 3± 18.7	28.2 ± 17.5
Protein	86.7 ± 27.5	83.7 ± 26.1[a]	84.9 ± 25.2	95.0 ± 30.4[b]	89.8 ± 30.9
Fat	78.5 ± 22.1	76.5 ± 21.9	77.9 ± 19.1	82.5 ± 25.7	81.1 ± 23.7
Saturated Fat	19.4 ± 11.0	18.7 ± 11.4	19.2 ± 8.5	19.5 ± 10.7	21.9 ± 13.4
Perceived Stress					
PSS Score	20.0 ± 6.4	18.7 ± 6.8[a]	21.0 ± 6.0[b]	19.9 ± 5.5	22.4 ± 6.1[b]
Adverse Childhood Experiences					
ACE Score	1.76 ± 2.00	1.22 ± 1.66[a]	1.96 ± 2.03[b]	1.61 ± 1.61[a,c]	3.2 ± 2.5[b,d]

Superscripts with differing letters indicate statistical significance between group means by food security status. Values that were significantly different from one another are indicated by superscripts as follows: when the values for 2 outcomes within a row do not share a common superscript, they are significantly different, whereas if the values do share a common superscript, they are not significantly different.

Table 4. Mean scores for select domains of executive function by food security status (n = 230)

	Total Sample (n = 230)	High Food Security (n = 102)	Marginal Food Security	Low Food Security	Very Low Food Security
Cambridge Gambling Task (CGT)					
Delay Aversion Total	0.33 ± 0.23	0.32 ± 0.21[a]	0.29 ± 0.18[a]	0.44 ± 0.23[b]	0.29 ± 0.30[a]
Decision Making Quality	0.96 ± 0.06	0.96 ± 0.06	0.96 ± 0.06	0.95 ± 0.07	0.97 ± 0.05
Sensitivity to Risk	1.64 ± 1.10	1.76 ± 1.05	1.44 ± 1.04	1.44 ± 1.34	1.80 ± 0.99
Delayed Match to Sample (DMS)					
DMS Percent Correct (all delays)	85.75 ± 12.99	85.37 ± 12.03	85.93 ± 12.25	83.69 ± 17.71	88.79 ± 10.92
DMS Percent Correct (simultaneous)	97.27 ± 7.84	97.23 ± 7.50	97.86 ± 8.25	97.22 ± 7.01	96.47 ± 9.17
DMS Probability of Error Given Error	0.11 ± 0.19	0.09 ± 0.17	0.13 ± 0.20	0.16 ± 0.23	0.08 ± 0.16
Intra-Extra Dimensionality Shift (IED)					
Total Latency	90677.06 ± 35924.13	89519.97 ± 32069.43	97226.95 ± 44677.55	92413.39 ± 39483.69	83291.48 ± 29312.3
Total Trials (Adjusted):	101.76 ± 60.58	93.33 ± 46.03[a]	114.18 ± 75.15[b]	117.5 ± 77.33[b]	90.18 ± 46.47
Stages Completed	8.43 ± 1.40	8.62 ± 1.05[a]	8.13 ± 1.77[a]	8.19 ± 1.82	8.62 ± 1.04
Total Errors (Adjusted)	27.97 ± 34.36	23.35 ± 26.27[a]	34.70 ± 42.52[b]	36.50 ± 43.73[b]	21.85 ± 26.85
One Touch Stockings (OTS)					
Mean Choices to Correct	1.55 ± 0.55	1.52 ± 0.50	1.55 ± 0.54	1.65 ± 0.67	1.54 ± 0.56
Median Latency to First Choice	8678.84 ± 3984.60	8573.87 ± 3297.22	9131.59 ± 4764.68	7852.89 ± 3889.69	9116.38 ± 4520.10
Problems Solved on First Choice	10.70 ± 2.80	10.84 ± 2.66	10.66 ± 2.95	10.42 ± 3.07	10.64 ± 2.80

Rapid Visual Processing (RVP)					
Sensitivity to the target sequence	0.91 ± 0.05	0.92 ± 0.05[a]	0.90 ± 0.05[b]	0.90 ± 0.06[b]	0.92 ± 0.05
Response Latency	507.31 ± 110.50	490.96 ± 99.19	524.90 ± 128.01	523.82 ± 116.86	510.36 ± 102.42
Probability of False Alarm	0.015 ± 0.045	0.012 ± 0.036[a]	0.016 ± 0.052	0.030 ± 0.069[b]	0.007 ± 0.014
Total Misses	17.63 ± 9.38	16.05 ± 9.13[a]	19.98 ± 9.73[b]	18.72 ± 9.60	17.38 ± 8.77

Superscripts with differing letters indicate statistical significance between group means by food security status. Values that were significantly different from one another are indicated by superscripts as follows: when the values for 2 outcomes within a row do not share a common superscript, they are significantly different, whereas if the values do share a common superscript, they are not significantly different.

Table 5. Multivariate linear regression models with food security status as the independent variable and domains of EF as the dependent variable, controlling for biological sex, BMI, stress, and adverse childhood experiences.

Cognitive Assessment Test	Adjusted R^2	P-Value
Cambridge Gambling Task (CGT)	0.02	0.12
Delayed Match to Sample (DMS)	**0.04**	**0.03***
Intra Extra Dimensionality Shift Test (IED)	0.003	0.35
One Touch Stockings of Cambridge (OTS)	0.02	0.13
Rapid Visual Processing (RVP)	**0.07**	**< 0.001*** **

*Indicates significance of $p < 0.05$; **Indicates significance of $p < 0.01$; ***Indicates significance of $p < 0.001$

CHAPTER 6

Discussion

The work encompassed in this dissertation aims to elucidate the relationship between food insecurity (FI) and the clinical manifestation of physical and mental health outcomes in college students. College students have been an under-researched population as it relates to FI and health outcomes, despite the increased risk of FI in this demographic. To date, the research on associated negative health outcomes in this population has consisted of survey-based findings measured through subjective, self-reported assessments of health status, as previously described. This warranted the exploration into the use of objective measures to assess components of physiological and cognitive health impacted by the experience of FI.

Carotenoids were selected as the biomarker of interest for this dissertation research due to their bioactive properties that have been demonstrated to slow the progression of chronic disease development by means of anti-inflammatory and antioxidant function.[1] Intake of dietary carotenoids has been associated with improvements in cognitive function, memory, vision, DNA protection, and immune function.[1] Found ubiquitously in red, orange, yellow, green, and purple fruits and vegetables, carotenoids serve as a relative proxy for F/V intake.[2] Carotenoids are fat-soluble compounds that are detectable in the diet, plasma, and adipose tissue, with each biological medium reflecting different durations of intake.[3] Measuring changes in carotenoid concentrations in individuals experiencing FI compared to their food secure counterparts increases the understanding dietary intake and diet quality as it relates to health outcomes. The consistent monitoring of carotenoid levels through non-invasive, objective assessment tools may improve self-efficacy for health-promoting behavior change in college students experiencing FI.[4]

The emergence of technology-based dietary assessment tools is advancing dietary intake data collection modalities, with the promise for reducing the time, cost, and resource-intensive

structure of traditionally used diet assessment tools, such as repeated 24-hour dietary recalls or food frequency questionnaires (FFQs).[5-7] In addition to the researcher and participant burden, traditional dietary intake tools introduce the risk of systematic, recall and social desirability biases, which may impact the quality of dietary data collected.[8, 9] In the college student population and environment, peer approval and group think mentalities are powerful predictors of behavior.[10, 11] This poses exceptional challenges regarding issues pertaining to FI, as the stigmatization and fear of alienation may inhibit the individual from reporting accurate depictions of dietary intake.[12] Assessing dietary intake and diet quality in populations vulnerable to FI warrants the implementation of a complex, multidisciplinary, and holistic approach to address the social, financial, and health-related attributes.[13]

In order to capture the experience of FI, the incorporation of technology may provide additional insight into detecting changes in acute and chronic health implications.[14] Prior to implementing novel technologies in the human clinical research setting, rigorous validation of innovative dietary assessment tools must be conducted.[15] Diet ID™, along with other image-based and wearable technologies to assess diet quality and dietary intake allow for the continuous monitoring of dietary intake without relying on participant memory or recall abilities.[16, 17] For this reason, Diet ID™ output for nutrients of interest, such as diet quality assessed by the Healthy Eating Index 2015 (HEI-2015), total calories, macronutrients, select micronutrients, phytonutrients, and nutrients of concern as expressed in the 2020 – 2025 Dietary Guidelines for Americans were assessed against the gold standard of dietary intake, the Nutrition Data System for Research (NDSR).[5, 18] In addition to NDSR, objective measures of plasma and skin carotenoids were also assessed for convergence of nutrient outputs. The findings demonstrated significant correlations between Diet ID™ and carotenoid intake from NDSR,

plasma carotenoids and skin carotenoid scores.[19] Additional research using the Automated Self-Administered 24-hour (ASA24®) Dietary Assessment Tool also resulted in significant correlations between Diet ID™ and ASA24 output for select nutrients.[20] Research utilizing Diet ID™ in populations experiencing cardiovascular and cardiometabolic disease is on-going to determine the effectiveness of assessing risk factors for chronic disease monitoring and prevention.[21] As FI increases the risk of diet-related chronic disease development, the implementation of dietary assessment tools, such as Diet ID™ in the food environment or healthcare setting, may serve as a successful intervention to improve awareness to the association between dietary intake and health outcomes.[22]

The aforementioned difficulties measuring dietary intake and nutrient status provide the catalyst for inventing novel, objective, non-invasive tools. The Veggie Meter® is a device that utilizes pressure-mediated reflection spectroscopy to quantify the density of carotenoids stored within the adipose tissue of the finger.[23] In under 90 seconds, the Veggie Meter® provides an objective score ranging from 0 – 800 that serves as a proxy for fruit and vegetable consumption.[24, 25] Veggie Meter® scores have been strongly correlated to plasma carotenoids and have the capacity to collect similar information on changes in the intake of carotenoid-containing foods in a rapid and minimally burdensome manner.[26] Although the Veggie Meter® reduces the biases that may arise with traditional dietary recalls, the score has yet to be equated to the exact number of servings of fruits and vegetables. This tool is effective when assessing individual dietary patterns but does not yet have the ability to determine quantity of intake.[25]

Effective interventions aimed at improving FI across the lifespan may benefit from the implementation of Diet ID™, the Veggie Meter®, or other similar dietary assessment methods to increase autonomy over food-acquiring behaviors without weight-centric stigmatizations.

Currently, primary interventions for improving FI for college students include food pantries, food banks, and local, state, and federal food assistance programs, such as the Supplemental Nutrition Assistance Program (SNAP).[27] These resources are predominately focused on increasing the quantity of food available for patrons, but the concept of nutrition security, or ensuring that the foods offered are nutrient-dense, may not be possible due to limited operational finances.[28] The Aggie Compass Basic Needs Resource Center at the University of California, Davis organizes and operates multiple food access resources that prioritize the regular distribution of fresh fruits and vegetables to students, free of charge. For this reason, evaluating the impact of food access resources was explored to determine if individuals utilizing campus-based avenues to access food had improvements in health-related biomarkers, in addition to solely assessing energy intake. On-campus food access resource usage positively impacted skin carotenoid scores; however, the inconsistent frequency of use made it challenging to detect changes in plasma concentrations. The study results hold promise for the potential use of biologic outcomes data as justification for increased food access resources to alleviate the consequential effects of FI on health outcomes. Emerging research using Geographic Information System (GIS) may help to increase the number of food access resources in college environments to exacerbate health-promoting effects.[29]

In addition to diet-related biomarkers and other measure of physiological health status, the experience of FI may also affect cognitive function.[30] Stress, anxiety, and poor sleep have been found to be elevated in the experience of FI,[31] and simultaneously have negative impacts on executive function (EF).[32-34] As EF regulates neuronal activity located in the frontal lobe, such as decision making, impulsivity, and acute and long-term planning, the detrimental impacts of FI on crucial brain function may be mitigated with increased dietary intake of nutrient-dense foods.[35]

Previous research using functional magnetic resonance imaging (fMRI) scans of the anatomy and neuronal connectivity of the brain found significant differences in college students experiencing FI compared to those who were food secure;[36] however, assessing domains of EF as it relates to dietary patterns and dietary quality had yet to be explored in emerging adults. Select domains of EF were impacted by the experience of FI. Such experiences with limited or uncertain avenues of meeting nutrient needs continues the bi-directional relationship between FI influencing EF, with qualities of EF perpetuating cognitive attributes that yield an increased risk for FI. Future research is warranted to explore the impact of food access resource usage on cognitive processes to determine if the incorporation of nutrient-dense foods following periods of acute or chronic FI improve select domains of executive function.

Previously, research has been conducted in children and older adults to determine the physiological and cognitive impacts of FI; however, prior to this dissertation work there was a dearth of knowledge regarding impacts on the young adult population. Symptoms related to diet-related chronic disease etiology, as seen with obesity, diabetes, cardiovascular disease, and dyslipidemia, among others, have been demonstrated to develop prior to adulthood.[37] As brain development and cognitive processes continue to mature well into the late twenties, the emerging adult population, including a majority of college-aged individuals, may be exceptionally impacted by the experience of FI.[38] Therefore, the areas of this dissertation were justified, as existing research did not provide substantial insight into this vulnerable population that may experience compounded effects of FI on health outcomes due to concurrent physiological and cognitive development occurring in this life stage.

Conclusion

Synthesizing the collective information found in this dissertation, the diet-related and cognitive impacts of acute or chronic FI exert substantial risk to health status in the college student population. Research efforts should prioritize assessing the duration and severity of the FI experience to determine immediate areas for targeted interventions. Longitudinal analysis from the emerging adult population to middle-age adulthood will explicate the impact of FI on chronic disease development and progression.

The continued development and expansion of food access programs aimed at increasing dietary intake of health-promoting foods is warranted. Approaching the idea of food assistance through collegiate education strategies on nutrition and financial literacy may improve food security status and promote adherence to more healthful eating patterns. The experience of FI is omnipresent in the college student population and requires immediate attention to reduce the negative health impacts forthcoming.

References

1. Zia-Ul-Haq M. Past, Present and Future of Carotenoids Research. Carotenoids: Structure and Function in the Human Body 2021. doi:10.1007/978-3-030-46459-2_28
2. Holden JM, Eldridge AL, Beecher GR, Buzzard IM, Bhagwat S, Davis CS, Douglass LW, Gebhardt S, Haytowitz D, Schakel S. Carotenoid Content of U.S. Foods: An Update of the Database. Journal of Food Composition and Analysis 1999;12(3):169-196.
3. Amorim-Carrilho KT, Cepeda A, Fente C, Regal P. Review of methods for analysis of carotenoids. TrAC Trends in Analytical Chemistry 2014;56:49-73. doi:10.1016/j.trac.2013.12.011
4. Strecher VJ, DeVellis BM, Becker MH, Rosenstock IM. The Role of Self-efficacy in Promoting Health Behavior Change. Health Education Quarterly 1986;13:73-91. doi:10.1177/109019818601300108
5. Thompson FE, Subar AF. Chapter 1 - Dietary Assessment Methodology. Nutrition in the Prevention and Treatment of Disease (Fourth Edition) 2017:5 - 48.
6. Pendergast FJ, Leech RM, McNaughton SA. Novel Online or Mobile Methods to Assess Eating Patterns. Curr Nutr Rep 2017;6(3):212-227. doi:10.1007/s13668-017-0211-0
7. Eldridge AL, Piernas C, Illner AK, Gibney MJ, Gurinović MA, de Vries JHM, Cade JE. Evaluation of New Technology-Based Tools for Dietary Intake Assessment-An ILSI Europe Dietary Intake and Exposure Task Force Evaluation. Nutrients 2018;11(1):55. doi:10.3390/nu11010055

8. Hebert JR, Clemow L, Pbert L, Ockene IS, Ockene JK. Social desirability bias in dietary self-report may compromise the validity of dietary intake measures. Int J Epidemiol 1995;24(2):389-98. doi:10.1093/ije/24.2.389

9. Kipnis V, Midthune D, Freedman L, Bingham S, Day NE, Riboli E, Ferrari P, Carroll RJ. Bias in dietary-report instruments and its implications for nutritional epidemiology. Public Health Nutr 2002;5(6A):915-23. doi:10.1079/PHN2002383

10. Cox MJ, DiBello AM, Meisel MK, Ott Mq, Kenney SR, Clark MA, Barnett NP. Do misperceptions of peer drinking influence personal drinking behavior? Results from a complete social network of first-year college students. Psychol Addict Behav 2019;33(3):297-303. doi:10.1037/adb0000455

11. Graupensperger S, Abdallah DA, Lee CM. Social norms and vaccine uptake: College students' COVID vaccination intentions, attitudes, and estimated peer norms and comparisons with influenza vaccine. Vaccine 2021;39(15):2060-2067. doi:10.1016/j.vaccine.2021.03.018

12. Loofbourrow BM, Jones AM, Chodur GM, Martinez SM, Kemp LC, Scherr RE. Evaluating Knowledge, Attitudes, and Practices Regarding CalFresh Participation in University Students. Nutrients 2022;15(1):192. doi:10.3390/nu15010192

13. Calloway EE, Parks CA, Byker Shanks C, Bowen DJ, Yaroch AL. A special issue addressing healthful food access and food insecurity: risk factors, behavioral variables, interventions, and measurement. Transl Behav Med 2020;10(6):1249-1254. doi:10.1093/tbm/ibaa119

14. Hendriks SL. The food security continuum: a novel tool for understanding food insecurity as a range of experiences. Food Security 2015;7(3):609-619. doi:10.1007/s12571-015-0457-6

15. Bailey RL. Overview of dietary assessment methods for measuring intakes of foods, beverages, and dietary supplements in research studies. Curr Opin Biotechnol 2021;70:91-96. doi:10.1016/j.copbio.2021.02.007

16. Vadiveloo MK, Juul F, Sotos-Prieto M, Parekh. Perspective: Novel Approaches to Evaluate Dietary Quality: Combining Methods to Enhance Measurement for Dietary Surveillance and Interventions. Adv Nutr 2022;13(4):1009-1015. doi:10.1093/advances/nmac007

17. Katz DL, Rhee LQ, Katz CS, Aronson DL, Frank GC, Gardner CD, Willett WC, Dansinger ML. Dietary assessment can be based on pattern recognition rather than recall. Med Hypotheses 2020;140:109644. doi:10.1016/j.mehy.2020.109644

18. United States Department of Agriculture and the United States Department of Health and Human Services. 2020 - 2025 Dietary Guidelines for Americans. 9th Edition. December 2020 [Accessed March 25, 2023]. Available at https://www.dietaryguidelines.gov/sites/default/files/2020-12/Dietary_Guidelines_for_Americans_2020-2025.pdf

19. Radtke MD, Chodur GM, Bissell MCS, Kemp LC, Medici V, Steinberg FM, Scherr RE. Validation of Diet ID™ in Predicting Nutrient Intake Compared to Dietary Recalls, Skin Carotenoid Scores, and Plasma Carotenoids in University Students. Nutrients 2023;15(2):409. doi:10.3390/nu15020409

20. Turner-McGrievy G, Hutto B, Bernhart JA, Wilson MJ. Comparison of the Diet ID™ Platform to the Automated Self-administered 24-hour (ASA24) Dietary Assessment Tool for Assessment of Dietary Intake. J Am Nutr Assoc 2022;41(4):360-382. doi:0.1080/07315724.2021.1887775

21. Dansinger ML, Breton GL, Joly JE, Rhee LQ, Katz DL. Rapid, Digital Dietary Assessment in Association with Cardiometabolic Biomarkers. Am J Health Promot 2023:8901171231156513. doi:10.1177/08901171231156513

22. Rozanski A, Sakul S, Narula J, Berman D. Assessment of lifestyle "vital signs" in healthcare settings. Prog Cardiovasc Dis 2023:S0033-0620(23)00008-7. doi:10.1016/j.pcad.2023.02.002

23. Ermakov IV, Gellermann W. Dermal carotenoid measurements via pressure mediated reflection spectroscopy. J Biophotonics 2012;5(7):559-570. doi: 10.1002/jbio.201100122

24. Radtke MD, Jilcott Pitts S, Jahns L, Firnhaber GC, Loofbourrow BM, Zeng A, Scherr RE. Criterion-Related Validity of Spectroscopy-Based Skin Carotenoid Measurements as a Proxy for Fruit and Vegetable Intake: A Systematic Review. Adv Nutr 2020;11(5):1282-1299. doi:10.1093/advances/nmaa054

25. Radtke MD, Poe M, Stookey J, Jilcott Pitts S, Moran NE, Landry MJ, Rubin LP, Stage VC, Scherr RE. Recommendations for the Use of the Veggie Meter® for Spectroscopy-Based Skin Carotenoid Measurements in the Research Setting. Curr Dev Nutr 2021;5(8):nzab104. doi:10.1093/cdn/nzab104

26. Jahns L, Johnson LK, Conrad Z, Bukowski M, Raatz SK, Jilcott Pitts S, et. al. Concurrent validity of skin carotenoid status as a concentration biomarker of vegetable and fruit intake compared to multiple 24-h recalls and plasma carotenoid concentrations across one

year: a cohort study. Nutr J 2019;18(1):78. doi:10.1186/s12937-019-0500-0and plasma carotenoid concentrations across one year: a cohort study. Nutrition Journal 2019.

27. Nazmi A, Martinez SM, Byrd A, Robinson D, Bianco S, Maguire J, et al., A systematic review of food insecurity among US students in higher education. Journal of Hunger & Environmental Nutrition 2018;14(5):725-740. doi:10.1080/19320248.2018.1484316

28. Rivera AF, Smith NR, Ruiz A. A systematic literature review of food banks' supply chain operations with a focus on optimization models. Journal of Humanitarian Logistics and Supply Chain Management 2023;13(1):10-25. doi:10.1108/JHLSCM-09-2021-0087

29. Bacon CM, Baker GA. The rise of food banks and the challenge of matching food assistance with potential need: towards a spatially specific, rapid assessment approach. Agriculture and Human Values 2017;34(4):899-919. doi:10.1007/s10460-017-9783-y

30. Royer MF, Guerithault N, Braden BB, Laska MN, Bruening M. Food Insecurity Is Associated with Cognitive Function: A Systematic Review of Findings across the Life Course. International Journal of Translational Medicine 2021;1(3):205-222. doi:10.3390/ijtm1030015

31. Martinez SM, Grandner MA, Nazmi A, Canedo ER, Ritchie LD. Pathways from Food Insecurity to Health Outcomes among California University Students. Nutrients 2019;11(6):1419. doi:10.3390/nu11061419

32. Shields GS, Moons WG, Slavich GM. Better executive function under stress mitigates the effects of recent life stress exposure on health in young adults. Stress 2017;20(1):75-85. doi:10.1080/10253890.2017.1286322

33. Shields GS, Moons WG, Tewell CA, Yonelinas AP. The effect of negative affect on cognition: Anxiety, not anger, impairs executive function. Emotion 2016;16(6):792-7. doi:10.1037/emo0000151

34. Tai XY, Chen C, Manohar S, Husain M. Impact of sleep duration on executive function and brain structure. Commun Biol 2022;5(1):201. doi:10.1038/s42003-022-03123-3

35. Dalile B, Kim C, Challinor A, Geurts L, Gibney ER, Galdos MV, et al. The EAT-Lancet reference diet and cognitive function across the life course. Lancet Planet Health 2022;6(9):e749-e759. doi:10.1016/S2542-5196(22)00123-1

36. Guerithault N, McClure SM, Ojinnaka CO, Braden BB, Bruening M. Resting-State Functional Connectivity Differences in College Students with and without Food Insecurity. Nutrients 2022;14(10):2064. doi:10.3390/nu14102064

37. Himmelgreen D, Romero-Daza N, Heuer J, Lucas W, Salinas-Miranda AA, Stoddard T. Using syndemic theory to understand food insecurity and diet-related chronic diseases. Soc Sci Med 2022;295:113124. doi:10.1016/j.socscimed.2020.113124

38. Carriedo N, Corral A, Montoro PR, Herrero L, Rucián M. Development of the updating executive function: From 7-year-olds to young adults. Dev Psychol 2016;52(4):666-78. doi:10.1037/dev0000091

APPENDIX

Recommendations for the Use of the Veggie Meter® for Spectroscopy-based Skin Carotenoid Measurements in the Research Setting

Introduction

Carotenoids are bioactive phytochemicals found in a variety of fruits and vegetables (F/V) that cannot be synthesized *de novo* in humans and therefore, are only obtained from the diet.[1] For this reason, measurement of carotenoid status has gained traction in population-based nutrition research as an objective biomarker for the estimation of F/V intake. Following dietary intake of carotenoids, these lipophilic compounds are metabolized, transported by lipoproteins in the bloodstream, and accumulate in various tissues, including blood, skin, and other organs.[2] The kinetics of carotenoid distribution and clearance are dependent on a variety of factors, such as age, obesity status, stress, illness, and oxidative damage.[3] Existing methods for quantifying tissue and blood carotenoid concentrations are typically invasive or subject to participant error and bias.[4-6] Blood samples and dermal, adipose, or muscle biopsies provide an overview of circulating and deposited carotenoid compounds, but these methods may be painful and/or burdensome to participants.[7, 8] In comparison, methods for determining carotenoid and fruit and vegetable intakes through dietary assessments, such as self-reported dietary recalls and food records, are subjective and inherently biased, leading to inaccurate estimates.[9] Emerging evidence supports the use of spectroscopy-based skin carotenoid measurements as a non-invasive, objective method for determining skin carotenoid concentrations, indicative of F/V intake.[10]

The Veggie Meter® is a spectroscopy-based skin carotenoid measurement device, created by Longevity Link Corporation (Salt Lake City, Utah, USA) in 2015, with the purpose of commercializing pressure-mediated reflection spectroscopy (RS) for detecting and quantifying skin carotenoids as a proxy for F/V intake in humans.[11] The Veggie Meter® is a small, portable device that detects skin carotenoid concentrations in approximately 15-20 seconds for a single reading, including processing time and display information, or about 90 seconds to complete three individual measurements (**Figure 1**).[12] Validation studies of RS methodology against high-performance liquid chromatography of excised human tissue samples were comparable,[13, 14] and serum samples were highly correlated with skin carotenoid scores measured using the Veggie Meter® ($R = 0.81$; $p < 0.001$).[15] The Veggie Meter® connects to an interfaced laptop computer to display an individual's "skin carotenoid score" on a histogram, with the x-axis illustrating the range of scores from 0 to 800, and the y-axis representing the reference population frequency. Skin carotenoid scores are plotted in relation to a reference population, which has been constructed by aggregating a large convenience sample of individuals' scores recorded using the Veggie Meter® and cross calibrating these values with skin carotenoid scores measured using Raman resonance spectroscopy (RRS).[11] The reference distribution was designed to feature a bell-shaped distribution with a slight skew toward higher skin carotenoid scores, such that the halfwidth of the distribution is approximately 75% of the peak score (**Figure 2**).[13] The histogram may be used to illustrate where a participant's skin carotenoid score compares to the reference population of individuals of all ages, sexes, and race/ethnicities, previously measured using this device.[13] Although accumulated data, to date, indicate that the average Veggie Meter® score may vary among populations for a number of dietary and physiological reasons, specific reference histograms for sub-populations or condition-specific populations have yet to be developed.

Compilation of measurements from various groups under varying physiological conditions and a range of dietary carotenoid and fruit and vegetable intakes are needed to develop tailored reference distributions, which may support the interpretation of skin carotenoid score results.

In addition to the RS approach utilized by the Veggie Meter®, other spectroscopy-based skin carotenoid methods include RRS and spectrophotometers, which have also been demonstrated to be valid assessments of F/V intake, with a majority of correlation coefficients > 0.40.[10] Although these alternative methods for spectroscopy-based carotenoid detection exist and are used in the research setting, the Veggie Meter® remains an attractive instrument for research use due to its affordability, portability, and responsivity to detect changes in dietary patterns related to F/V consumption.[10, 14] For example, RRS requires a spectrally precise LED light source, whereas the Veggie Meter® uses a relatively low-powered white LED for carotenoid excitation. Therefore, the overall cost of the Veggie Meter®, priced at approximately $15,000 USD, is relatively inexpensive compared to other spectroscopic devices. Although the light strength and precision of the detection methods differ between RRS and RS, validation studies comparing the two methods found comparable skin carotenoid signals ($R = 0.94$; $p < 0.001$).[15] Spectrophotometers used for skin carotenoid estimation detect red and yellow dermatological pigments that fall within the UV range in the color spectra of carotenoid compounds (~350 – 500 nanometers).[16] As various compounds fall within the carotenoid detection UV spectral window, some spectrophotometers may include measurements of additional compounds or chromophores, such as hemoglobin and deoxyhemoglobin in the absorption measurement, reflecting a higher skin carotenoid score than actually exists.[14, 17] Comparatively, the Veggie Meter® applies supradermal pressure at 1 atm (~14.7 PSI) ± 10% to limit blood circulation to the anatomical assessment region, thus preventing other chromophores from interfering with carotenoid

detection.[13] Furthermore, the Veggie Meter® identifies and corrects for individual melanin concentration by employing an algorithmic deconvolution adjustment; therefore, at a group level, melanin was not found to independently correlate with skin carotenoid score.[15] Although the Veggie Meter® provides objective measures of skin carotenoid status, if the continued use of the device remains inconsistent among users, there is potential for differences in data interpretation. The need for a protocol outlining the use of the device is critical for minimizing between-user and between-study error.

The aforementioned advantages of the Veggie Meter® support the use of this device in the research setting; however, there is currently no standardized protocol for using the Veggie Meter® to assess skin carotenoids. Thus, the purpose of this study was to determine current practices and examine variability among Veggie Meter® users, ultimately to create and disseminate a standardized protocol for quantifying skin carotenoids in human subjects using the Veggie Meter®. A standardized protocol can support comparability among study data and the creation of a universal data repository to aggregate skin carotenoid scores recorded across all Veggie Meter® devices. Although the Veggie Meter® provides an estimate against a reference population, the device does not account for individual characteristics that may impact skin carotenoid measurements. In order to address the need for population- and condition-specific reference distributions and recommended ranges and trajectories for Veggie Meter® scores, this study aimed to identify protocol-related barriers to Veggie Meter® score aggregation and comparison. It is imperative to provide guidelines for collecting uniform information data for a central repository and as a basis for comparison for any further protocol modifications or deviations indicated by future advances in this field.

Materials and Methods

Development of the Current Practices Survey

The Veggie Meter® device is accompanied by an operating procedure;[18] however, these instructions are subject to individual interpretation and, therefore, implementation may not be consistent among users. Following a comprehensive review of the literature in early 2020, the novelty of the device was apparent by the limited number of publications.[12, 15, 19-25] In the existing literature and supported by anecdotal evidence, inconsistencies among researchers were observed and procedural details pertaining to the Veggie Meter® were not reported in detail. Therefore, to determine the current methodologies among researchers using the Veggie Meter® for research purposes, a survey developed at the University of California, Davis in partnership with the San Francisco Department of Public Health (SFDPH) was distributed to Veggie Meter® users and members of the International Carotenoid Database Group (veggiemeter@ucdavis.edu). This survey did not meet the criteria to be considered human subjects research and therefore, no Institutional Review Board action was required. The survey questions were derived from the Veggie Meter® operating procedure by identifying the areas that either were ambiguous or open for interpretation and asking for clarification or explanation through open-ended response questions. The final survey consisted of 21 questions, with 17 questions directly related to the experience of users, calibration and set-up technique, measurement information, interpretation of results, and additional documentation details for Veggie Meter® use. The remaining four questions pertained to the research site specific Institutional Review Board protocol verbiage and researcher contact information.

Distribution of the Current Practices Survey

On April 7, 2020 the survey was distributed via the Veggie Meter® listserv (veggiemeter@ucdavis.edu), a previously established listserv to aggregate communication

among nutrition scientists using the Veggie Meter®. The survey was distributed online (www.surveymonkey.com, San Mateo, CA, USA) using a modified tailored Dillman approach.[26] If the individual on the listserv was not familiar with the operating procedures for the Veggie Meter®, that person was advised to distribute the survey to a more experienced user. Consequently, the exact number of survey recipients may extend beyond those included in the listserv. The survey was open for two weeks and closed on April 21, 2020. Responses were compiled and data cleaning was performed on the open-ended questions by identifying emergent themes and categorizing responses on areas of convergence. Descriptive analysis of the categorical variables were summarized by count and frequency (%) in May of 2020.

Development of the Standardized Protocol

A standardized protocol was developed based on current literature, input from the Veggie Meter® creators, and results from the Current Practices Survey regarding recommendations to reduce user discrepancies; As the recommendations stated in the standardized protocol were derived based upon these various sources, some of the current practices reported by Veggie Meter® users may not agree with the suggestions in the protocol for collecting skin carotenoid scores for research purposes.

The draft protocol was sent to the Veggie Meter® listserv for feedback. Members of the listserv were asked to review the protocol and complete a questionnaire regarding willingness to adhere to particular aspects of the protocol in future research efforts. Users were asked to elaborate on any areas where they were not willing to adhere to aspects of the protocol.

Results

Survey Results

The results from the survey supported the need to develop universal recommendations for the use of the Veggie Meter® in the research setting due to a majority of users implementing differing methodologies. Listserv members are from a variety of research backgrounds and work with racially/ethnically diverse populations and populations of varying ages, including infants and toddlers, children, and adults. Table 1 includes the operational- and procedural-related questions from the Current Practices Survey, along with the participant responses. Among participants who completed the survey (n=19), 63% of Veggie Meter® users were considered experienced users, defined as performing over 500 skin carotenoid measurements using the device.

Discrepancies were observed in the following operational and procedural steps:

Device Set-up and Calibration

Differences regarding the set up and calibration of the Veggie Meter® using the dark and light reference materials were recorded, along with a low number of responders documenting the date, time, and location of the calibrations (Table 1). Per manufacturer instructions, calibration of the device is recommended before use and every hour thereafter. Improper or infrequent calibration may result in inaccurate skin carotenoid measurements. On occasion, an error in calibration may occur when the expected calibration display does not match the actual display. Many users reported simply repeating the calibration until the expected display was achieved, and some users take the additional step of cleaning the device prior to repeating the calibration step. An additional question regarding concerns about device set-up and calibration resulted in the requests for proper instructions on what to do when the calibration display does not match, as well as an explanation for the importance of repeated calibration.

Measurement Preparation and Anatomical Site Selection

Variation in the selection of the anatomical site was observed, such that users differed in the use of dominant versus non-dominant hand and in digit preference for the measurement location (Table 1). In addition, site preparation prior to the measurement was inconsistent, indicating the use of alcohol-based solutions, such as a pre-soaked wipe or hand sanitizer, washing hands using soap and water, using multiple cleaning methods, employing whatever method is convenient at the research location, or not having participants wash the anatomical site prior to performing the measurement were all reported (Table 1).

Data Collection and Interpretation

The number of individual skin carotenoid measurements was generally consistent among users, with a majority of researchers (n = 16; 84%) performing triplicate measurements per participant and using the average of the three readings, using the "Average of 3 Scans" mode or conducting three separate measurements and averaging manually, to reduce intra-individual variability. However, differences in the interpretation of skin carotenoid scores were identified, such that 58% of users interpret Veggie Meter® results relative to the baseline value for the same individual, 16% interpret results relative to the current study population, 11% relative to the absolute reference score, another 11% relative to a previously reported value associated with a specific diet or outcome, and one user stated that interpretation is dependent on the particular study (Table 1).

Documentation

Although the manufacturer indicates that the Veggie Meter® is highly sensitive to excessive heat, cold, or bright light due to the potential for optical interference with the LED, 89% of users did not document environmental conditions, such as temperature or humidity (Table 1).[11] This

documentation of environmental conditions is not currently stated in the most recent version of the manufacturer manual,[18] however, when using the device outside of a controlled setting, environmental conditions may impact findings. Additionally, with 32% of users having more than one Veggie Meter®, proper documentation of which device is being used is important in order to limit inconsistencies and inter-instrument variability; of the users who indicated they had multiple devices, only 31% record which instrument is being used for each study participant.

User-reported Acceptability of the Standardized Protocol

To address the inconsistencies observed in the survey results and make future collected data amenable to inter-site analysis, a standardized protocol was developed to support the use of the Veggie Meter® in the research setting. Detailed explanations corresponding with the recommendations in the standardized protocol are provided in the subsequent discussion. This standardized protocol outlines the operating procedure for use of this device as a research-grade instrument to ensure researchers using the Veggie Meter® are following steps for consistency, repeatability, and generalizability. The standardized protocol can be followed verbatim whenever the Veggie Meter® is being used to collect research data that can be compared with data from other research locations. The standardized protocol can be found in the *Supplementary Data Appendix.*

To determine the likelihood that the standardized protocol will be implemented in the research setting, current Veggie Meter® users were asked to review the protocol recommendations and indicate whether they would be willing to implement the protocol steps in future research efforts (Table 2). Responses were received from Veggie Meter® users (n = 21, response rate = 39.6%) and the qualitative feedback was used to modify the final draft of the standardized protocol.

Discussion

Justification for Standardized Protocol Recommendations

The survey results indicated a need for the development of a standardized protocol for use of the Veggie Meter® to improve the generalizability and applicability of skin carotenoid scores recorded using this device. Moreover, the results from the Current Practices Survey suggested that methods currently in use may introduce additional variability into skin carotenoid score data. The discrepancies identified in the calibration and set-up technique, anatomical site selection and preparation, measurement methods, interpretation of results, and documentation processes yield insight to the importance of developing and disseminating a standardized protocol for the Veggie Meter®.

Device Set-up and Calibration

Calibration of the Veggie Meter® using the dark and white reference blanks should be conducted prior to performing measurements. The calibration graphs should be compared to the reference graphs to confirm that calibration of the device was successful. If the graphs are not consistent with the reference images, calibration should be repeated. Calibration of the Veggie Meter® should occur at least every two hours after continuous use. Nevertheless, more frequent calibration at the one-hour time lapse is recommended for research purposes.[11] The Veggie Meter® should be re-calibrated if the device is elevated from a flat surface or transported to a different data collection site within the two-hour time period.

Measurement Preparation and Anatomical Site Selection

Prior to conducting skin carotenoid measurements, it is advantageous to describe the operational procedure of the Veggie Meter® to participants to avoid apprehension due to the frequent assumption that the measurement will be painful.[15, 19, 24] When working with toddlers, children,

or adolescents, it is recommended to both verbally explain and demonstrate the process of measuring skin carotenoids using the Veggie Meter® to ensure the participants feel comfortable during data collection procedures.[19] In infants and toddlers, digit size will play a role in accuracy of measurements, therefore, prior to working with infants, toddlers, or children, contact the manufacturers for instructions and device modifications.

Emerging evidence has demonstrated variability in skin carotenoid scores due to differences in anatomical site selection.[27] To reduce the inconsistencies due to differences in hand and digit selection, the non-dominant digitus medicinalis, commonly known to as the ring finger, should be used when conducting skin carotenoid measurements with the Veggie Meter®. Since the dominant hand has increased vasculature and musculature resulting from more frequent use, the non-dominant hand recommendation was selected to ensure minimal deoxyhemoglobin and oxyhemoglobin chromophore interference.[28] The digitus medicinalis or ring finger is preferred due to the decreased callusing compared to the index finger, thereby reducing skin thickness to allow for increased light penetration to the subdermal detection region.[29] Therefore, to minimize potential error due to vasculature and skin thickness, the non-dominant ring finger is the preferred measurement site. Additional research is required to determine individual variability using different digits under various levels of carotenoid intake.

In addition to the physiological differences driving the selection of the non-dominant ring finger, observations at the community level, particularly regarding the use of the Veggie Meter® in the school setting, were also considered. It was evident in the residual staining on the thumb and index finger of the dominant hand that children regularly consumed highly pigmented foods or had remnant splotches from colored markers or paint, which may subsequently affect skin-carotenoid measurements due to these pigments being incorrectly identified within the

carotenoid-detection spectral range.[30] Although the Veggie Meter® is equipped with a spectral deconvolution algorithm to correct for melanin and other chromophore interference, supracutaneous pigmentation may not be accurately detected or corrected for in this process.

Prior to performing skin carotenoid measurements, it is recommended that participants wash hands using soap and warm water to ameliorate potential confounding residue;[31] however, since the Veggie Meter® may be used in settings where access to a proper hand-washing equipment is not available, a pre-moistened alcohol prep pad or hand wipes may be used instead. Hand sanitizer is not recommended, as it may not remove pigment or interfering debris. Additionally, to ensure minimal lens interference, the surface of the contact lens should be cleaned using an optical cloth after every participant.[23]

Data Collection and Interpretation

Triplicate measures should be conducted to increase reliability of skin carotenoid scores. The triplicate measurement feature of the Veggie Meter® (also known as "Average of Three Scans" mode) averages the scores across three consecutive measurements and reports the mean value. The digit should be retracted from the device as indicated by the measurement software between respective measurements to ensure reperfusion occurs.[23] The triplicate mode is equivalent to measuring skin carotenoids scores three separate times, writing down the score each time, summing them up, and dividing by three; however, the triplicate mode computes the mean automatically and reports a single average skin carotenoid score for the participant. The triplicate mode increases the accuracy and reproducibility of skin carotenoid detection by two-fold due to the light excitation disk sampling slightly different measurement sites, which minimizes tissue irregularities due to repeated blood reperfusion to the measurement site and accounts for sweat gland ducts or papillary ridges, that may affect carotenoid detection. Some users indicate using

the single measurement mode to obtain three unique values for an individual, in order to determine intra-individual variation. This is a method that can be considered if a researcher is interested in the respective values that comprise the mean. Regardless of whether the triplicate measurement mode or repeating the single measurement mode three separate times is used, it is recommended to collect three measurements to generate a mean skin carotenoid score to account for tissue inhomogeneities. The single measurement mode may be used in situations with limited time allotted for data collection; however, for research purposes, the triplicate measurement should be used exclusively when possible.

Rarely, the Veggie Meter® will be unable to compute an individual's skin carotenoid score and an error message due to a non-detectable or zero measurement will appear in the display window. If this occurs, confirm the finger has no contaminants, reposition the finger in the center of the contact lens, and repeat the measurement process. The output may display a skin carotenoid score on the second attempt; however, in some cases the error message may persist, in which case there will be no quantitative skin carotenoid value computed. Therefore, the researcher should consider removing this participant from the study, deeming them ineligible for inclusion. It is not recommended to attempt to record an individual's skin carotenoid score using a different finger, as this may impact the reproducibility of measurements.

As the Veggie Meter® computes skin carotenoid scores immediately, researchers have the option of communicating the results to participants or blinding participants to their scores. Recommendations may vary based on study design. If conducting a randomized controlled trial or intervention study aimed at increasing skin carotenoid scores over time, it is not recommended to share results, as this may influence dietary behaviors. If the research study focuses on dietary tracking or monitoring, chronic disease prevention, or nutrition education effectiveness, sharing

the results with the participants may create a visual representation as a strategy to promote and maintain positive behavior change.

Researchers should carefully consider what and how to share skin carotenoid score results with study participants. Depending on the study design, the results may bias the behaviors of the participant, or may cause the participant to experience stress or concern about their own score. If the participant expresses stress, the researcher can emphasize that scores, at this point, cannot be compared between individuals and are the result of many different personal considerations, such as age, sex, BMI, alcohol consumption, smoking status, and dietary factors. It should be conveyed that a low skin carotenoid score does not necessarily equate with poor health or risk of chronic disease.

Internet connections may pose connectivity issues or make the device more vulnerable to harmful software viruses. It is recommended to back up the data on a USB drive and transfer to project-specific password-encrypted file. For this reason, confidential or identifiable participant information should not be directly inputted into the Veggie Meter® software. It is advised to assign subject identification codes to ensure participant anonymity, in accordance with the guidelines enforced by the respective Institutional Review Boards.

Documentation

Skin carotenoid scores may be influenced by a variety of intrapersonal factors. To account for individual characteristics that may alter skin carotenoid measurements, the following data should be collected whenever possible: age, sex, BMI, smoking status, supplement use, and diagnosed chronic diseases.

The Veggie Meter® is an optoelectronic device, making it highly sensitive to changes in temperature and light; therefore, it is imperative to record environmental conditions to examine

whether the results were confounded by environmental exposure (Table 3). This is particularly of interest when working in community settings, outdoors, at a hospital, or in any other environment where exposure to mechanical shock from sudden movement, excessive heat, or bright lights may occur. In addition to the environmental conditions, researchers with more than one Veggie Meter® should record which device is used to collect the skin carotenoid scores, as this will minimize potential errors arriving from inter-device variability. While this is a recommendation from the manufacturers, future research is needed to investigate the influence of different environmental conditions on Veggie Meter® output.

Previous research has indicated possible seasonal differences in the intake of carotenoid-rich foods, therefore, documenting the date the device was used should be factored into data analysis to account for the potential unintended changes in skin carotenoid scores due to seasonality.[3, 10, 32] Additionally, individual health status, such as BMI, acute and chronic illnesses, supplementation or long-term medication intake may impact skin carotenoid scores.[23, 33] If a participant experiences drastic weight loss or is diagnosed with a chronic disease during the study, it may be important to document these changes since they may alter skin carotenoid scores.

Veggie Meter® users expressed areas of concern regarding the additional time and resources required to document individual characteristics and environmental conditions; however, as these are important requirements for the development of a universal skin carotenoid data repository, it is recommended to collect consistent information among Veggie Meter® users whenever possible.

Future Applications

Effective methods for measuring F/V consumption, such as the rapid and non-invasive carotenoid detection method using the Veggie Meter®, have the capacity to assess public health and nutrition education interventions, assist physicians, registered dietitians, and other health professionals with monitoring the health status of patients, and in research as an objective biomarker for F/V intake.[10, 20, 21, 24, 35, 36] The Veggie Meter® has been used to measure skin carotenoids in ethnically diverse toddlers, children, and adult populations in both clinical and non-clinical settings.[12, 15, 19-24, 37, 38] Future research efforts should focus on conducting a systematic review of the studies using the Veggie Meter® to determine if the differences in published methods for skin carotenoid data acquisition affect the reported outcomes (Table 4).

Recommendations to increase the validity and generalizability of the Veggie Meter® in the research setting include continued usage and validation of the RS method across the lifespan, in individuals of various BMI classifications, and in populations of diverse races and ethnicities. Introducing this technology into the clinical setting in primary healthcare facilities and non-clinical sectors, such as the public school systems and other community environments, has the potential to support preventative health services and health interventions aimed at improving health outcomes in children and adults.

With the proposed integration of the Veggie Meter® into various health, education, and community settings, expanded utilization of the device will facilitate an increase in the number of unique Veggie Meter® measurements, thereby broadening the diversity and generalizability of the skin carotenoid data repository. Implementing the standardized protocol for the Veggie Meter® is fundamental to the development of a universal skin carotenoid data repository to establish recommended ranges of skin carotenoid concentration values at the population level. With the previously identified differences in skin carotenoid scores based on individual

characteristics, the proposed repository will incorporate these factors to determine an estimated range of skin carotenoid scores based on these factors. Therefore, it is recommended that researchers report consistent non-identifiable, individual level characteristics to be incorporated into the database and to facilitate comparisons among studies when using the Veggie Meter®. With the emergence of new literature, Veggie Meter® users will be contacted using the established listserv to determine if the recommendations presented in this protocol remain the most appropriate for the research setting. Further validation is needed to determine the extent to which differences in the digit(s) measured and site preparation are correlated with plasma carotenoids and dietary carotenoid intake assessed by controlled dietary interventions or rigorous 24-hour dietary recalls.

Limitations

It is important to acknowledge potential limitations in the standardized protocol developed for the use of the Veggie Meter® in the research setting. The survey that informed current practices was distributed to known Veggie Meter® users and members of the International Carotenoid Database Group in April of 2020, amid a peak in the Covid-19 pandemic. For this reason, it is possible that some researchers who use the Veggie Meter® for research did not receive nor respond to the survey; however, as there were many discrepancies observed among the 19 survey respondents, it is expected that an increase in survey responses would further exacerbate the inconsistencies. The proposed standardized protocol may not address some sources of error.

As the Veggie Meter® is an emerging technology among researchers, it is important to expedite the adoption of this standardized protocol to inform future practices for using this device in the research setting. It is important to note that some of the information presented in this protocol was extracted from published abstracts, in addition peer-reviewed manuscripts,

which was expected due to the novel, innovative, and emerging nature of the device as a research-grade instrument. It was difficult to acquire precise methodologies since many papers did not state the specific procedures used when operating the device. As the Current Practices Survey was conducted in spring of 2020, it should be acknowledged that an updated review of more recently published literature using the Veggie Meter® was performed, confirming that procedural discrepancies still persisted in more recent literature.[39-50] This finding emphasizes the importance of establishing a standardized protocol for the use of the Veggie Meter® in the research setting to ensure consistency among users moving forward, which will allow for comparisons between studies.

Regarding the Veggie Meter®, there are limitations to the use of the device in the research setting that should be recognized. The majority of studies that have used the Veggie Meter® to determine skin carotenoid status have been performed in populations consisting of children and adult participants. Current efforts to evaluate the validity of the Veggie Meter® in infants are on-going.[51] To the authors' knowledge, no studies have been conducted using the Veggie Meter® in older adult populations. Therefore, due to the limited data available on the use of the Veggie Meter® in infants and older adults, amendments to the standardized protocol may be appropriate in these populations.

Conclusions

The standardized protocol for use of the Veggie Meter® will provide researchers with comprehensive instructions on how to operate the device in the research setting. Standardization of the procedure will increase the comparability of the results among studies and allow for a more robust database of skin carotenoid measurements to be developed. The recommendations provided in the standardized protocol are based upon the most recent understanding of

carotenoid physiology and the method by which the Veggie Meter® measures the carotenoids deposited in the skin. These recommendations can be strengthened by systematic experimentation and evaluation of commonly occurring discrepancies among Veggie Meter® use (Table 4). With the goal of the standardized protocol being to reduce variability among researchers, this protocol neither invalidates nor undermines the significance of previously collected data that utilized the Veggie Meter® with different approaches or methodologies. The use of the standardized protocol will strengthen the field of spectroscopy-based skin carotenoid measurements and as researchers employ the standardized protocol for the Veggie Meter®, this will allow the opportunity to create suggested ranges for specific populations, including different stages across the lifespan, BMI classifications, and for individuals living with chronic diseases.

This manuscript was published:

Radtke MD, Poe M, Stookey J, Jilcott Pitts S, Moran NE, Landry MJ, Rubin LP, Stage VC, Scherr RE. Recommendations for the Use of the Veggie Meter® for Spectroscopy-based Skin Carotenoid Measurements in the Research Setting. Current Developments in Nutrition 2021; 5(8). doi: https://doi.org/10.1093/cdn/nzab104

References

1. Goodwin TW. Metabolism, Nutrition, and Function of Carotenoids. Ann Rev Nutr 1986;6:273-297.
2. Mayne ST, Cartmel B, Scarmo S, Jahns L, Ermakov IV, Gellerman W. Resonance Raman spectroscopic evaluation of skin carotenoids as a biomarker of carotenoid status for human studies. Arch Biochem Biophys 2013;539(2):163-170.
3. Darvin ME, Sterry W, Lademann J, Vergou T. The Role of Carotenoids in Human Skin. Molecules 2011;16(12):10491-10506.
4. Beaton GH, Milner J, Corey P, McGuire V, Cousins M, Stewart E, de Ramos M, Hewitt D, Grambsch PV, Kassim N, Little JA. Sources of variance in 24-hour dietary recall data: implications for nutrition study design and interpretation. Am J Clin Nutr 1979;*32(12): 2546-2559*.
5. Thomson CA, Giuliano A, Rock CL, Ritenbaugh CK, Flatt SW, Faerber S, Newman V, Caan B, Graver E, Hartz V, Whitacre R, Parker F, Pierce JP, Marshall JR. Measuring dietary change in a diet intervention trial: comparing food frequency questionnaire and dietary recalls. Am J Epidemiol 2003;157(8):754-762.
6. Bledsoe MJ, Grizzle WE. Use of human specimens in research: the evolving United States regulatory, policy, and scientific landscape. Diagn Histopathol 2013;19(9):322-330.
7. Block G, Norkus E, Hudes M, Mandel S, Helzlsouer K. Which Plasma Antioxidants Are Most Related to Fruit and Vegetable Consumption? Am J Epidemiol 2001;154(12):1113-1118.

8. Nierenberg DW, Nann SL. A method for determining concentrations of retinol, tocopherol, and five carotenoids in human plasma and tissue samples. Am J Clin Nutr 1992;56(2):417-426.
9. Natarajan L, Flatt SW, Sun X, Gamst AC, Major JM, Rock CL, Al-Delaimy W, Thomson CA, Newman VA, Pierce JP. Women's Healthy Eating and Living Study Group. Validity and systematic error in measuring carotenoid consumption with dietary self-report instruments. Am J Epidemiol 2006;163(8):770-778.
10. Radtke MD, Jilcott Pitts SB, Jahns L, Firnhaber GC, Loofbourrow BM, Zeng A, Scherr RE. Criterion-Related Validity of Spectroscopy-Based Skin Carotenoid Measurements as a Proxy for Fruit and Vegetable Intake: A Systematic Review. Adv Nutr 2020;11(5): 1282-1299.
11. Longevity Link Corporation. The Veggie Meter® – skin carotenoids as indicator of fruit and vegetable intake and biomarker associated with better health [Internet]. [Accessed 2021 Jan 7]. 2016. Available at: http://www.longevitylinkcorporation.com
12. Jilcott Pitts SB, Jahns L, Wu Q, Moran NE, Bell RA, Truesdale KP, Laska MN. A non-invasive assessment of skin carotenoid status through reflection spectroscopy is a feasible, reliable and potentially valid measure of fruit and vegetable consumption in a diverse community sample. Public Health Nutr 2018;21(9):1664-1670.
13. Ermakov IV, Gellermann W. Optical detection methods for carotenoids in human skin. Arch Biochem Biophys 2015;572:101-111.
14. Ermakov IV, Gellermann W. Dermal carotenoid measurements via pressure mediated reflection spectroscopy. J Biophotonics 2012;5(7):559-570.
15. Ermakov IV, Ermakova M, Sharifzadeh M, Gorusupudi A, Farnsworth K, Bernstein PS, Stookey J, Evans J, Arana T, Tao-Lew L, Isman C, Clayton A, Obana A, Whigham L,

Redelfs AH, Jahns L, Gellermann W. Optical assessment of skin carotenoid status as a biomarker of vegetable and fruit intake. Arch Biochem Biophys 2018;646:46-54.

16. Meléndez-Martínez AJ, Britton G, Vicario IM, Heredia FJ. Relationship between the colour and the chemical structure of carotenoid pigments. Food Chem 2007;101(3): 1145-1150.
17. Alaluf S, Heinrich U, Stahl W, Tronnier H, Wiseman S. Dietary Carotenoids Contribute to Normal Human Skin Color and UV Photosensitivity. J Nutr 2002;132(3):399-403.
18. Longevity Link. 2020. *Operating Instructions for the Veggie Meter* ®. Salt Lake City, Utah, USA.
19. May K, Jilcott Pitts SB, Stage VC, Kelley CJ, Burkholder S, Fang X, Zeng A, Lazorick S. Use of the Veggie Meter® as a tool to objectively approximate fruit and vegetable intake among youth for evaluation of preschool and school-based interventions. J Hum Nutr Diet 2020;33(6):869-875.
20. Jilcott Pits SB, Wu Q, Truesdale KP, Haynes-Maslow L, McGuirt JT, Ammerman A, Bell R, Laska MN. One-Year Follow-Up Examination of the Impact of the North Carolina Healthy Food Small Retailer Program on Healthy Food Availability, Purchases, and Consumption. Int J Environ Res Public Health 2018;15(12):2681.
21. McGuirt JT, Jilcott Pitts SB, Gustafson A. Association between Spatial Access to Food Outlets, Frequency of Grocery Shopping, and Objectively-Assessed and Self-Reported Fruit and Vegetable Consumption. Nutrients 2018;10(12):1974.
22. Obana A, Gohto Y, Gellermann W, Ermakov IV, Sasano H, Seto T, Bernstein PS. Skin Carotenoid Index in a large Japanese population sample. Sci Rep 2019; 9(1):9318.
23. Rush E, Amoah I, Diep T, Jalili-Moghaddam S. Determinants and Suitability of Carotenoid Reflection Score as a Measure of Carotenoid Status. Nutrients 2020;12(1): 113.

24. Bakirci-Taylor AL, Reed DB, McCool B, Dawson JA. mHealth Improved Fruit and Vegetable Accessibility and Intake in Young Children. J Nutr Educ Behav 2019;51(5): 556-566.
25. Thomson JL, Goodman MH, Landry AS, Donoghue A, Chandler A, Bilderback R. Feasibility of Online Nutrition Education in the Workplace: Working Toward Healthy Lifestyles. J Nutr Educ Behav 2018;50(9):868-875.
26. Dillman DA, Smyth JD, Christian LM. Internet, Mail, and Mixed-Mode Surveys: The Tailored Design Method. John Wiley: Hoboken, NJ. 2009, 3rd edition.
27. Ansu V, Dickinson S, Fly A. Digit Variability in Carotenoid Scores Obtained with the Veggie Meter®: A Pilot Study (P02-001-19). Curr Dev Nutr 2019;3(1).
28. Kagaya A, Ohmori F, Okuyama S, Muraoka Y, Sato K. Blood flow and arterial vessel diameter change during graded handgrip exercise in dominant and non-dominant forearms of tennis players. Adv Exp Med Biol 2010;662:365-370.
29. Fruhstorfer H, Abel U, Garthe CD, Knöttel A. Thickness of the stratum corneum of the volar fingertips. Clin Anat 2000;13(6):429-433.
30. Sutter C, Nishina A, Scherr RE, Zidenberg-Cherr S, Ontai LL. Sharing, Trading, Stealing. J Early Adolesc 2015;36(4):449-464.
31. Erickson T, Dickinson S, Golzarri-Arroyo L, Fly A. Effect of Handwashing on Skin Carotenoids Measurements Using Pressure-Mediated Reflectance Spectroscopy. Curr Dev Nutr 2020;4(2):103.
32. Moran NE, Mohn ES, Hason N, Erdman JW Jr., Johnson EJ. Intrinsic and Extrinsic Factors Impacting Absorption, Metabolism, and Health Effects of Dietary Carotenoids. Adv Nutr 2018;9(4):465-492.

33. Jahns L, Johnson LK, Conrad Z, Bukowski M, Raatz SK, Jilcott Pitts SB, Wang Y, Ermakov IV, Gellermann W. Concurrent validity of skin carotenoid status as a concentration biomarker of vegetable and fruit intake compared to multiple 24-h recalls and plasma carotenoid concentrations across one year: a cohort study. Nutr J 2019;18(1):78.
34. Salter-Venzon D, Gellenbeck K, Hu C. Clinical Evaluation of Three Carotenoid Containing Supplements Using Fingertip Reflection Spectroscopy Measurements (P02-009-19). Curr Dev Nutr 2019;3(1).
35. Jamshidi-Naeini Y, Oldewage-Theron W. P162 Comparative Outcome Evaluation of 3 Delivery Models for a Diabetes Prevention Education Program for Older Adults. J Nutr Educ Behav 2020;52(7):S93.
36. Scherr RE, Laugero KD, Graham DJ, Cunningham BT, Jahns L, Lora KR, Reicks M, Mobley AR. Innovative Techniques for Evaluating Behavioral Nutrition Interventions. Adv Nutr 2017;8(1):113-125.
37. Bayles J, Peterson AD, Jilcott Pitts SB, Bian H, Goodell LS, Burkholder S, Hegde AV, Stage VC. Food-Based Science, Technology, Engineering, Arts, and Mathematics (STEAM) Learning Activities May Reduce Decline in Preschoolers' Skin Carotenoid Status. J Nutr Educ Behav 2020;53(4):343-351.
38. Burkholder S, Jilcott Pitts SB, Wu Q, Bayles J, Baybutt R, Stage VC. Skin Carotenoid Status Over Time and Differences by Age and Sex Among Head Start Children (3-5 Years). J Nutr Educ Behav. 2020;53(2):103-109.
39. Di Noia J, Gellermann W. Use of the Spectroscopy-Based Veggie Meter® to Objectively Assess Fruit and Vegetable Intake in Low-Income Adults. Nutrients 2021;13(7):2270.

40. Martinelli S Acciai F, Tasevska N, Ohri-Vachaspati P. Using the Veggie Meter® in Elementary Schools to Objectively Measure Fruit and Vegetable Intake: A Pilot Study. Methods Protoc 2021;4(2):33.

41. Kopetsky Fultz A, Rex SM, Mazelin A, McGarry C, Brewer B, Patterson F, Robson S. Examining fruit and vegetbale intake in low-income older adults using the Veggie Meter®. Nutr Health 2021;2601060211022275.

42. Di Noia J, Monica D, Jensen HH, Sikorskii A. Economic Evaluation of a Farm-to-Special Supplemental Nutrition Programme for Women, Infants, and Children Intervation Promoting Vegetable Consumption. Pub Health Nutr 2021.

43. Liu R, Edwards CG, Cannavale CN, Flemming IR, Chojnacki MR, Reeser GE, Iwinski SJ, Renzi-Hammond LM, Khan NA. Weight Status and Visceral Adiposity Mediate the Relation between Exclusive Breastfeeding Duration and Skin Carotenoids in Later Childhood. Curr Dev Nutr 2021;5(3).

44. Hill CM, Paschall MJ, O'Brien DM, Bersamin A. Characterizing Vegetable and Fruit Intake in a Remote Alaska Native Community Using Reflection Spectroscopy and 24-Hour Recalls. J Nutr Educ Behav 2021.

45. McGuirt JT, Qu W, Laska MN, Truesdale KO, Rafferty AP, Bell RA, Ammerman AS, Jilcott Pitts SB. Association between shopping patterns, dietary behaviours and geographic information system-assessed store accessibility among small food store customers. Pub Health Nutr 2020.

46. Nagao-Sato S, Baltaci A, Omar Peralta Reyes A, Zhang Y, Ali Hurtado Choque A, Reicks A. Skin Carotenoid Scores Assessed with Reflection Spectroscopy Are Associated with Self-

Reported Fruit and Vegetable Intake Among Latino Early Adolescents. J Acad Nutr Diet 2021.

47. Jilcott Pitts SB, Wu Q, Truesdale KP, Rafferty AP, Haynes-Maslow L, Boys KA, McGuirt JT, Fleischhacker S, Johnson N, Kaur AP, Bell RA, Ammerman AS, Laska MN. A four-year observational study to examine the dietary impact of the North Carolina Healthy Food Small Retailer Program, 2017-2020. Int J Behav Nutr Phys Act 2021; 18(1):44.
48. Obana A, Gohto Y, Nakazawa R, Moriyama T, Gellermann W, Berstein PS. Effect of an antioxidant supplement containing high dose lutein and zeaxanthin on macular pigment and skin carotenoid levels. Sci Rep 2020;10:10262.
49. Di Noia J, Monica D, Sikorskii A. Process Evaluation of a Farm-to-WIC Intervention. J Acad Nutr Diet 2021.
50. Fam VW, Holt RR, Keen CL, Sivamani RK, Hackman RM. Prospective Evaluation of Mango Fruit Intake on Facial Wrinkles and Erythema in Postmenopausal Women: A Randomized Controlled Trial. Nutrients 2020;12(11):3381.
51. Moran NE, Hason N, Chang J, O' Connor T. Infant Heel Dermal Carotenoids Are Measured Rapidly and Reliably by Portable Reflection Spectroscopy. Curr Dev Nutr. 2020;4(2):122.

Figure 1. Veggie Meter® instrument and laptop interface

Figure 2. Veggie Meter® example output, containing overall skin carotenoid score and comparison to a reference population, indicated on the software output as the general population.

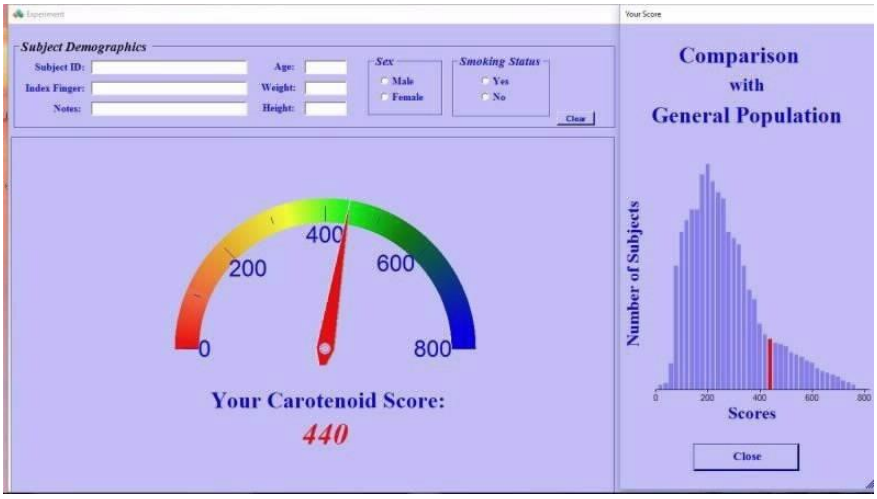


Table 1. Selected questions and responses from the Current Practices Survey to assess the operating methodologies currently implemented by Veggie Meter® users.

Questions from the Current Practices Survey	n (%)
How experienced are you using your Veggie Meter®?	
0 – 500 measurements	7 (36.8)
500 – 1,000 measurements	9 (47.4)
> 1,000 measurements	3 (15.8)
Have you managed to calibrate the Veggie Meter® before each (and every) hour of use using the white and dark calibration sticks?	
Always	8 (42.1)
Most of the time	5 (26.3)
Sometimes	4 (21.1)
Never	2 (10.5)
Which hand do you always use to take measurements?	
Dominant	2 (10.5)
Non-dominant	5 (26.3)
Right	5 (26.3)
Left	4 (21.1)
I don't always use the same hand	3 (15.8)
What finger do you always use to take measurements?	
Thumb	0
Index	12 (63.2)
Middle	3 (15.8)
Ring	3 (15.8)
Pinky/little	0
I don't always use the same finger	1 (5.3)
How do you prepare the fingers to prepare for a scan? Select all that apply.	
Wash hands (soap and water)	5 (26.3)
Use an alcohol-based solution	12 (63.2)
Other	7 (36.8)
How do you interpret (or plan to interpret) the Veggie Meter® results?	
Relative to the distribution of scores in the current study population	3 (15.8)
Relative to an absolute reference result or score (same value across populations/studies)	2 (10.5)
Relative to a prior value for the same	11 (57.9)
Relative to a result associated with a particular diet or outcome in a peer-reviewed publication	2 (10.5)
Other	1 (5.3)
Do you record environmental conditions such as temperature, humidity, etc.?	
Always	0
Most of the time	0
Sometimes	2 (10.5)

Never	17 (89.5)
Do you keep track of which instrument is doing which measurement? (Only for users who indicated that they had more than one device).	
Always	5 (31.3)
Most of the time	2 (12.5)
Sometimes	1 (6.3)
Never	3 (18.8)

Table 2. Responses from Veggie Meter® users about implementing the recommendations in the standardized protocol in future research efforts.

Veggie Meter® User Feedback on Standardized Protocol	n (%)
Would you consider implementing the proposed recommendation of allowing a 15-minute acclimation period if the Veggie Meter® is introduced to a new environment, especially one with substantial changes in temperature, lighting, or relative humidity?	
Yes, I will implement	17 (81.0)
No, I will not implement	0
I am not sure if I will implement	4 (19.0)
Qualitative Responses[1]	
Time constraint	2
Need to see the data for why this is important and how much of an effect it would have on the outcome	2
Would you consider implementing the proposed recommendation of calibrating the Veggie Meter® every 1 hour of operation or if the device is relocated or moved prior to the 1-hour time interval?	
Yes, I will implement	18 (85.7)
No, I will not implement	2 (9.5)
I am not sure if I will implement	1 (4.8)
Qualitative responses[1]	
Time constraint	2
Need to see the data for why this is important and how much of an effect it would have on the outcome	1
Would you consider implementing the proposed recommendation of using the triplicate (three-scan) mode on the Veggie Meter® to record skin carotenoid scores?	
Yes, I will implement	16 (76.2)
No, I will not implement	1 (4.8)
I am not sure if I will implement	4 (19.0)
Qualitative responses[1]	
Remain consistent during on-going study	1
Work with young children (2-5yrs)	1
Time constraint, particularly with large sample sizes	2
Need to see the data for why this is important and how much of an effect it would have on the outcome	1
Would you consider implementing the proposed recommendation of recording the following individual characteristics: age, sex, BMI, smoking status, supplement use, and diagnosed chronic diseases?	
Yes, I will implement	12 (57.1)
No, I will not implement	1 (4.8)
I am not sure if I will implement	8 (31.0)
Qualitative responses[1]	

Collecting disease status is outside the scope of Institutional Review Board approved protocols	2
Collecting personal information in a field-based setting may be inappropriate	2
Work with young children (2-5yrs)	3
Depends on purpose of the study – such information may not be needed	2
Would you consider implementing the proposed recommendation of using the non-dominant, ring finger to record skin carotenoid scores using the Veggie Meter®?	
Yes, I will implement	14 (66.7)
No, I will not implement	3 (14.3)
I am not sure if I will implement	4 (19.0)
Qualitative responses[1]	
Remain consistent during on-going study	2
Additional data is needed on finger variation	4
Not consistent with information provided in the user manual	1
Would you consider implementing the proposed recommendation of recording environmental conditions, such as temperature and relative humidity when using the Veggie Meter®?	
Yes, I will implement	9 (42.9)
No, I will not implement	3 (14.2)
I am not sure if I will implement	9 (42.9)
Qualitative responses[1]	
Time constraint	2
Not consistent with information provided in the user manual	2
Unsure how to collect environmental conditions	7
Need to see the data for why this is important and how much of an effect it would have on the outcome	1
Would you consider implementing the proposed recommendation of having participants wash hands with soap and warm water prior to recording skin carotenoid scores? If hand washing resources are unavailable, would you use a pre-moistened alcohol prep pad or hand wipes as an alternative?	
Yes, I will implement	19 (90.4)
No, I will not implement	1 (4.8)
I am not sure if I will implement	1 (4.8)
Qualitative responses[1]	
Some individuals may be sensitive to alcohol-based cleaners	1
Hand washing station not available in field setting	1

[1] Emerging themes from qualitative responses and number of respondents corresponding to each theme.

Table 3. Sample document for the recording information when using the Veggie Meter® for research purposes with a sample entry provided.

Veggie Meter® Data Collection Record					
Date	Location	Environmental Condition	Initial Calibration	Additional Re-Calibration Times	Notes
01/01/ 2021	Elementary School	78º F; overcast and humid (60-70%)	9:00am	9:55am, 10:52am, 11:48am	Device was moved to 3 classrooms; measurements at 11:48am occurred outside; Device II was used

Table 4. Proposed Future Validation Efforts for Use of the Veggie Meter® in the Research Setting for Skin Carotenoid Detection

Inconsistencies Among Users	Recommendation in the Research Setting and Rationale	Type of Study Design Needed to Verify Best Practice
Recording of Environmental Conditions	As the Veggie Meter® is an optoelectronic device, it is sensitive to environmental exposures; therefore, it is recommended to record where the device is being used, especially when in variable conditions, such as outdoor settings.	Study to assess discrepancies when using the device indoors vs. outdoors to determine if skin carotenoid values on the same participants are impacted by environmental conditions.
Measurement Site Preparation	Participants should wash hands with soap and water prior to having skin carotenoid measurements performed. If there is limited access to soap and water, an alcohol prep pad or hand wipe may be used to clean the measurement site.	Different site preparation techniques should be performed to assess the variability that may be caused due to residue or pigments that remain on the measurement site, which may interfere with carotenoid detection. These values obtained using different site preparations should be correlated with plasma carotenoids.
Measurement Site	The non-dominant ring finger should be used when possible to minimize potential error due to vasculature, musculature, and skin thickness.	Digit variability has been observed (26), however, additional research is required to determine the error between measurement sites as compared to dietary intake and/or plasma carotenoids.
Number of Measurements	The "Average of 3 Scans" mode or recording skin carotenoid scores three times and manually averaging the scores should be performed when possible.	Study to assess the reproducibility of results observed using the "Average of 3 Scans" mode and manually averaging the scores to determine whether the same margin of error is recorded.
Individual level data collected and reported	Age, sex, BMI, smoking status, supplement use, and diagnosed chronic disease status should be recorded when possible, as these individual characteristics may impact skin carotenoid status.	Efforts to create standardized skin carotenoid score ranges require the documentation of individual level characteristics. Controlled-feeding studies evaluating which of these characteristics are the most influential with respect to skin carotenoid scores are necessary.

Supplementary Data Appendix A

Standardized Protocol for Use of the Veggie Meter® in the Research Setting

The manufacturer's operating manual was modified for use of the Veggie Meter® for research purposes.

1. General

The Veggie Meter® uses reflection spectroscopy to measure the level of carotenoid pigments in an individual's skin. As a light source, the Veggie Meter® uses a white LED. To reduce inconsistencies in the research setting, the non-dominant ring finger should be selected for the measurement site. To obtain a measurement, the finger is inserted into the instrument's finger cradle to simultaneously bring the pad of the fingertip in close contact with the light source and light collecting contact lens. A spring-loaded clip gently applies pressure to the finger, such that blood flow is temporarily pushed away from the measured tissue site to avoid interfering with carotenoid detection.

A laptop computer interfaced to the instrument analyzes and quantifies the amount of white LED excitation light reflected from the finger and instantaneously derives a carotenoid score. The measurement takes approximately 10 seconds for a single measurement or 45 seconds for a triplicate measurement. Allow an additional 15 - 20 seconds for processing time and display information. In the multi-measurement mode, the finger is inserted and retracted three times and an average score is determined for the three measurement values.

Carotenoids are found in a wide variety of fruits and vegetables. They cannot be synthesized *de novo* in the human body and therefore are consumed through the diet. Thus, skin carotenoid scores are correlated with the dietary uptake of fruits and vegetables. In general, habitual consumption of fruits and vegetables results in an increase in skin carotenoid scores. The

instrument provides a comparative output of an individual's skin carotenoid score in relation to data from the general population. Additionally, changes in skin carotenoid scores may be tracked over time to determine responsivity to changes in dietary intake. Since carotenoid levels differ slightly between fingers, it is advisable to measure the same finger when tracking changes over time.

2. Equipment

- Veggie Meter®
- Interface Laptop and Corresponding Charger
- Connector Cords (2)
    - Black Cord
    - Grey Cord (device specific)
- Dark Calibration Reference Stick
- White Calibration Reference Stick
- Demonstrating tool and adaptor for different digit diameters (including toddlers and children)
- Access to hand-washing station, including anti-bacterial hand soap, warm water, and drying apparatus
    - If access to a hand-washing station is unavailable, alcohol-based prep pad or hand wipes may be used
- Optical cloth and glass cleaning fluid
- Data recording binder

3. Set-up

The Veggie Meter® instrument consists of a box shaped base (pedestal) that contains the electronics, and an oval shaped housing unit that sits on top of the base to provide the optical interface for measuring skin carotenoids.

3.1 The Veggie Meter® and interfaced laptop have limited battery capacity (approximately 6 hours following a complete charge). Connect the laptop with a 110 Volt power outlet or make sure a power outlet is nearby if required.

3.2 Power up the laptop by pressing and releasing the power button, which is located on the top right corner above the keyboard.

To activate the user screen, tap on the mouse pad. When prompted, enter the password “project” to login.

* 3.3 **This step is device specific.**

If the device you are using comes equipped with one cable (newer version), connect the Veggie Meter® to the interfaced laptop using the black cable by inserting one end into either of the USB ports and the other end into the housing port.

If the device you are using comes equipped with two cables (older version), connect the Veggie Meter® to the interfaced laptop using both cables:

Grey – connect the grey cable from the ***base (pedestal) port*** of the Veggie Meter® to the ***back port*** of the laptop

Black – connect the black cable from the ***housing port*** of the Veggie Meter® to the ***front port*** of the laptop

Note: the USB ports are not interchangeable. The instrument will not function properly if the cords are reversed.

3.4 Power up the Veggie Meter® by pressing and releasing the power button on the back of the instrument.

Note: There are two buttons located on the back of the instrument. The power button is the bottom button, located closest to the black cable connection. Ignore the top button.

* 3.5 The Veggie Meter® will take 5 minutes to warm-up. A colored display at the top of the housing unit will initially display the number 5, and then will count down the remaining minutes. **After 5 minutes, the instrument will display a logo. If the Veggie Meter® is in an environment with consistent ambient temperature, the 5-minute warm-up period is sufficient. If the Veggie Meter® is introduced to a new environment, especially one with substantial changes in temperature, lighting, or relative humidity, the device should run for 15-minutes before use to allow for adequate acclimation.**

3.6 Once the logo is displayed, move the laptop curser to the "VeggieMeter Shortcut" icon located on the desktop screen. Double click the icon to activate the program.

3.7 The Veggie Meter® program window should appear. There will be three panels indicating the following:

a. ***Top Panel*** – entry location for optional participant identification number and demographic information

b. ***Right Panel*** – selection of desired measurement mode ("Single Scan or "Average of 3 Scans") and calibration measurements (DARK REFERNCES and WHITE REFERENCE)

c. ***Display Panel*** – displays skin carotenoid score and histogram of general population. The display panel is blank prior to calibration.

Calibration

It is important to note that the expected ranges for calibration of the Veggie Meter® are device specific. Please refer to the operating manual to locate the expected range for dark and light calibration values for your device.

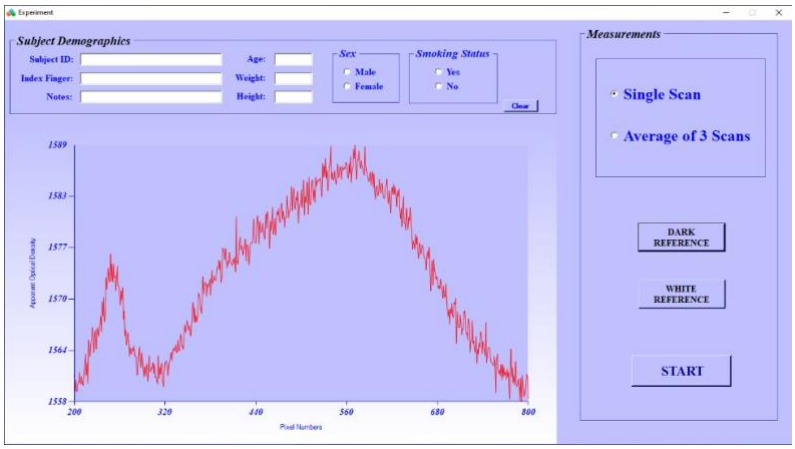
* 3.8 *Dark Calibration*

Gently slide the dark reference stick with the two side rails over the cradle, located on the housing port, such that the rails are pointing down and the black felt at the bottom of the stick is facing downwards, a few millimeters above the lens. The reference stick should push against the two inside plastic pegs while protruding from the housing port. When positioned properly, the instrument's white LED light will illuminate the black felt. Keep felt clean by avoiding touching the felt. Blow off lint or dust if needed.

Dark Reference Stick

Once the dark calibration stick has been properly inserted, click on the DARK REFERNCE button, located in the right panel of the program window. The display panel will now show the reflection spectrum for the dark reference felt material. A jagged low-intensity narrow band should appear on the left and a low-intensity broad band should be represented on the right side of the light reflecting spectrum. The intensity (height) of the spectrum is indicated on the vertical axis and the number of pixels is recorded on the x-axis.

Refer to the reference image below. Exact values may differ somewhat from those presented in this reference image as the calibration values are device specific. If the general image of a low-intensity narrow band followed by a broad band does not correspond to the reference image, repeat the dark calibration. Note that any contamination of the contact lens (from fingerprints, etc.) or the felt material (lint, etc.) may lead to increased intensities. In other words, it is desirable to have clean optics, such that the reflection intensities are as low as possible, with a resulting appearance of a highly jagged low-intensity reflection trace.



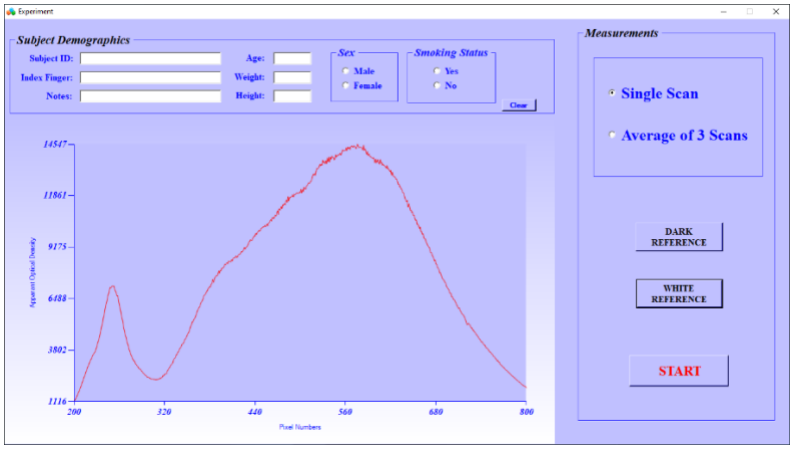
*3.9 *White Calibration*

Open the spring-loaded clip by pressing the back of the lever located on the housing port with one hand. Using the other hand, insert the white reference stick into the finger cradle, such that the indentation on the white plastic material fits snugly over the contact lens. Make sure the white surface of the reference stick is pointing downward.

Once the white calibration stick has been properly inserted, click on the WHITE REFERNCE button, located in the right panel of the program window. The display panel will now show the reflection spectrum for the white reference. A strong, high-intensity peak should appear on the left and a more intense broad band should be represented on the right side of the light reflecting spectrum. The

intensity (height) of the strong peak is indicated on the vertical axis and the number of pixels is recorded on the x-axis. Note that the auto-scaled reflection intensities are much higher now compared to the dark reflection spectrum and as a consequence the trace is much smoother.

Refer to the reference image below. Exact values may slightly differ from those presented in this reference image as the calibration values are device specific. If the general image of a high-intensity narrow peak followed by a broad band does not correspond to the reference image, repeat the white calibration.



Calibration Note: Calibration should be performed every time the instrument is turned on. For improved accuracy in the research setting, it is recommended that re-calibration occur with both black and white reference sticks every **1 hour of operation or if the device is relocated or moved prior to the 1-hour time interval**. You DO NOT have to turn the instrument off and on and restart the software: When ~1 hour interval lapsed, insert the black reference stick and click the “DARK REFERENCE” button, then insert the white reference stick and click the “WHITE REFERNCE” button. The respective intensities will override the previously stored intensities.

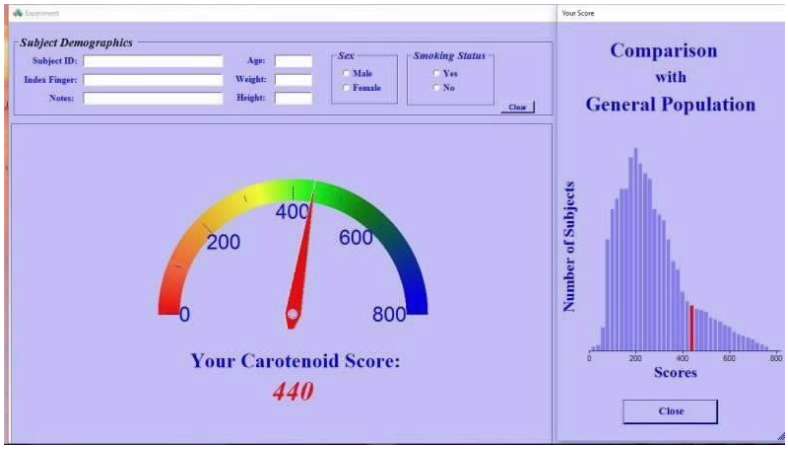
4. Skin Measurements

4.1 Once the device is calibrated, the instrument is ready for skin measurements.

*4.2 Enter the subject identification information. **To ensure participant anonymity, it is recommended to use a coded identification number in lieu of a name. As skin carotenoid scores may be impacted by individual characteristics, it is encouraged to record the following information when possible: age, sex, BMI, smoking status, supplement use, and diagnosed chronic diseases. Although these data are not required for measuring skin carotenoids, individual level data will increase the diversity and generalizability of the universal data repository.** Select the desired measurement mode by selecting either "Single Scan" button or "Average of 3 Scans" button. **For research purposes, the "Average of 3 Scans" mode is recommended for higher accuracy.**

*4.3 Insert the **non-dominant, ring finger** and click on START.

In "Single Scan" mode, the display window will first flash SCANNING IN PROGRESS, display a progress bar, and after finishing the scanning, will show "Your Carotenoid Score" on a scale from 0 to 800. In the display panel on the right, the measured score is compared with a histogram of scores for the general population. The histogram region corresponding to the measured score is shown in red. Click on the "Close" button to return to the Measurements screen and start a new scan.



In the "Average of 3 Scans" mode, follow the prompts in the displays and obtain the final score after 3 consecutive measurements. **Remove the finger after each measurement to allow for reperfusion.** Click on the "Close" button to return to the measurement screen.

*4.4 **To ensure minimal lens interference, the surface of the contact lens should be cleaned using an optical cloth in combination with a glass cleaning fluid after every participant has completed the three scan (triplicate) mode.**

4.5 The measurement data will automatically save into a folder on disk C called ResultsM. A shortcut icon for the results is located on the desktop.

5. Shutting Down

5.1 Once data collected is completed, click on the upper right window (x) in the measurements screen to close the program window. In the following exit window, click the OK button to close. It is important to close the program prior to disconnecting the device, as communication between the Veggie Meter® and laptop computer may be lost.

5.2 Turn off power to house of Veggie Meter® by pressing the lower button on the back of the base (pedestal).

5.3 Disconnect the black power cable from the Veggie Meter® and the USB port from the laptop. If the device contains two cords, after removing the black cord, disconnect the grey cable from the Veggie Meter® and the USB port from the laptop.

5.4 Pull up the main computer menu by sliding up on the mouse pad, click on "Power" and in the following window on the "Shut Down".

6. Notes

*6.1 In order to demonstrate the positioning, a spare cradle is supplied. The **non-dominant ring finger** should be positioned in the cradle, such that the pad of the finger fits snugly into the indentation of the cradle, and the tip of the finger touching the top edge of the cradle indentation. The user should fully feel the full rounded shape of the contact lens under the widest part of the measured finger.

*6.2 The Veggie Meter® is a highly sensitive optoelectronic device. Avoid mechanical shock and exposure to excessive heat or bright light. **For this reason, it is recommended to record environmental conditions, such as temperature and relative humidity when using the device outdoors or under variable environmental conditions where exposures to excessive heat or light may occur.**

6.3 Skin is a highly heterogenous tissue, with the degree of heterogeneity varying between individuals. It is normal that carotenoid scores vary from scan to scan for

the same individual. Any obtained score is usually within 10% of the average score that can be obtained via multiple measurements.

*6.4 Make sure the measured finger is clean. Any contamination needs to be removed prior to measurements. **Washing hands with soap and warm water is the most effective. If hand washing resources are unavailable, use a pre-moistened alcohol prep pad or hand wipes.**

*6.5 On occasion, the communication between the computer and instrument can be lost. This may occur if the shutting-down procedure is not followed. In those cases, corresponding messages will appear in the program window. To re-establish communication, abort or exit the program, and/or unplug and reconnect the instruments USB connections. This will require the standard operating procedure to be repeated to ensure the device is functioning properly. **It is recommended to back up the data on a USB drive and transfer to project-specific password-encrypted file to ensure no data is lost.**

6.6 Do not open the file ResultsM.csv or click on the Results shortcut icon while the instrument software is running. This may result in inaccurate syncing of data.

6.7 To avoid laptop battery discharge, remember to disconnect the black and grey cables between the Veggie Meter® and USB ports of the laptop as part of a normal shutdown procedure.