Vegetable behavioral tool demonstrates validity with MyPlate vegetable cups and carotenoid and inflammatory biomarkers.

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1 INTRODUCTION reviewer requested edits highlighted in yellow.

2 Eating more vegetables and fruit is a recommendation of the 2015 Dietary Guidelines, as well as the six preceding Guidelines
3 [U.S. Department of Health and Human Services and U.S. Department of Agriculture, 2015]. The reasons are: 1) most vegetables
4 are major contributors of nutrients under-consumed by most Americans; 2) evidence analysis of the literature is rated as
5 providing moderate support for the association of vegetables and fruit with reduced risk of many chronic diseases; 3) most
6 vegetables and fruit, when prepared without added fats or sugars, are low in calories, providing a link to a lower energy dense
7 diet and hence a strategy for obesity prevention.

8 Regarding vegetable and fruit intakes, young children are not meeting vegetable recommendations [U.S. Department of
9 Health and Human Services and U.S. Department of Agriculture, 2015]. While generally meeting fruit recommendations,
10 children, 1-3 years and 4-8 years, are eating 70% and 53% of recommendations for vegetables, 1.0 and 1.5 cup equivalents. Given
11 the above intakes, many nutrition professionals at federal nutrition assistance and education programs target family vegetable
12 behaviors to improve diet quality for children.

13 Our previously published literature review identified no vegetable behavior assessment tools for young children, aged 3-5
14 years, and specifically not for low-income children, aged 3-5 years, to accompany the above nutrition education experience
15 [Townsend et al. 2009]. A fruit and vegetable tool was identified for 8-10 year old multi-income African American children
16 [Cullen et al. 2004] and a tool for 0-3 year old children [Tibbs et al 2001]. Subsequent to that review, Sharafi et al. validated a
17 liking survey to assess diet quality of preschoolers [2015] and short food frequency questionnaires were developed to serve a
18 similar function [Bell et al. 2013]. The closest match in our search was a validation study for a social support tool for fruit and
19 vegetable intake for children 6-12 years using an assessment of household fruit and vegetable availability [Dave et al. 2012].
20 Notable is that the five studies identified here did not employ clinical biomarkers as objective and independent assessments of
21 validity [Bell et al. 2013]. No validation studies were identified focusing on vegetable or vegetable plus fruit behaviors
22 appropriate for low-income families with young children 3-5 years in the U.S.

23 Four federally-funded community programs would benefit from development of such a vegetable tool. These programs, all of
24 which target preschool-aged children, are available in most, if not all, low-income communities (Townsend 2006a). They include:
25 Head Start (U.S. Department of Health and Human Services 2014); Special Supplemental Nutrition Program for Women, Infants,

26 and Children [WIC] (U.S. Department of Agriculture Food and Nutrition Service 2014a); Supplemental Nutrition Assistance
27 Program-Education [SNAP-Ed] (U.S. Department of Agriculture Food and Nutrition Service 2014b) and Expanded Food and
28 Nutrition Education Program [EFNEP] (U.S. Department of Agriculture National Institute for Food and Agriculture 2014)

29 ***Focus on Veggies***

30 In response to this identified need, a tool called *Focus on Veggies* was created with 10 items representing parent and child
31 behaviors in the child's eating environment. The content of this tool is evidenced-based with content validity demonstrated
32 [Townsend et al. 2009] and includes vegetable accessibility [Kratt et al. 2000; Cullen et al. 2003], vegetable availability [Kratt et
33 al. 2000; Cullen et al. 2003], parent's role modelling of vegetable eating [Tibbs et al. 2001], child's picky eating [Galloway et al.
34 2005], child eating vegetables at meals/snacks [Reimer et al. 2004], parent planning meals [Reimer et al. 2004], and child eating
35 beans [Reimer et al. 2004; note: beans count as a vegetable when protein requirement is met, per US Dietary Guidelines 2015].

36 Cognitive interviews (n=77) with ethnically diverse Head Start and EFNEP parents provided contextually rich qualitative data
37 for tool development. Guided by respondents, items were written in familiar one- and two-syllable vocabulary supported by
38 realistic color photographs for text minimization and clarification. To capture the ten behaviors with cooking and eating
39 environments of 'real' parents and children, photographic sessions were conducted in participants' homes. The resulting version
40 provided support for the face validity of the tool with low-income respondents [Townsend et al. 2015]. **The 10-item *Focus on***
41 ***Veggies* is arranged on one sheet, 2 sided, and is shown in Figure 1 [Townsend et al. 2015].**

42 **Purpose**

43 Building on the qualitative work described above, this current paper describes the efforts to further establish the tool's reliability
44 and validity with parents of low socioeconomic status. We hypothesize that *Focus on Veggies* will demonstrate reliability using 4
45 approaches and validity using 5 methods.

46 **MATERIALS and METHODS**

47 **Participants**

48 Parent/child dyads were recruited at 13 Head Start and WIC sites in two counties in California. They were ethnically diverse
49 parents or caregivers, over the age of 18 years, who understood English as a first or second language, and had at least one child
50 between the ages of 3-5 years enrolled in Head Start or WIC. Respondents received \$10 for each interview, \$30 for the

phlebotomy session and \$30 for an optional at-home family photography session. The protocol was approved by the Institutional Review Board of the University of California, Davis.

Biopsychosocial framework

The framework used for the design of this validation research was based on Systems Theory [Engel 1980]. This framework considered the health of the child in the context of the family environment [Figure 2] and in that respect is similar to the Socio-Ecological Model used to guide development of the tool's content. In a hierarchical fashion, the model recognizes that the parent's psychosocial vegetable characteristics are constructs that temporally precede parent vegetable behaviors [Townsend et al. 2003], including the parent's perception of her diet quality, self-efficacy for eating vegetables, and readiness to eat more vegetables [Townsend & Kaiser 2005]. Those parent behaviors [Townsend et al. 2003] create the environment for the young child and this, in turn, drives or influences the child's vegetables behaviors [Townsend et al. 2015]. In turn, the young child's vegetable behaviors subsequently drive his vegetable intake [measured by three 24-hour diet recalls] resulting in increased consumption of vegetables indicated by increased cup equivalents of vegetables leading to improved intakes of micronutrients [vitamins C and A, β -carotene, folate and fiber]. And as specified by this framework, greater intake of vegetables by the child can result in positive changes in blood values and body weight status, all of which are indicators of the child's health status [Figure 2].

Data collection

Parents completed *Focus on Veggies*, a parent food behavior tool, fruit/vegetable psychosocial tool, and three researcher-administered 24-hour diet recalls about the child. The child provided a fasting blood sample, blood pressure, body temperature and height and weight measurements. For the baseline, data was collected over 15 weeks at four unique sessions in person [n=1] and by phone [n=3]. For reliability analysis, additional data was collected in-person two month later. Anthropometric data was collected again at an in-person session approximately one year later. Each session had a maximum duration of 60 minutes.

Reliability

Four approaches to assessing reliability were used: 1) Internal consistency, the proportion of variance in the scale score attributable to the true score [Litwin 1995]; 2) Item difficulty index, the ratio of each item's mean to its maximum score [Litwin 1995]; 3) Item discrimination index, the ability of an item to discriminate between those who do well on the 10-item tool and those who do not [Nunnally 1994]; 4) Temporal stability, within-person correlation with data collected at two time points with no intervention [Litwin 1995].

77 **Validity**

78 Five types of validity were estimated in addition to face and content validity previously assessed.

79 **1) Concurrent validity with parent psychosocial vegetable constructs.** The psychosocial constructs were measured by the
80 University of California Fruit and Vegetable Inventory with validity previously demonstrated [Townsend & Kaiser 2005; Townsend
81 & Kaiser 2007]. These three constructs included the perception of parent's diet quality; self-efficacy for planning, preparing and
82 eating vegetables; and readiness to eat more vegetables [Townsend and Kaiser, 2006] and were given equal weighting.
83 Responses were coded using a maximum of 5 points per item. Our hypothesis was that parent psychosocial constructs would be
84 positively related to scores on Focus on Veggies.

85 **2) Concurrent validity with parent eating behaviors.** The parent's vegetable behaviors were assessed using four vegetable items
86 from the University of California Food Behavior Checklist [Sylvia et al. 2006]. Serum carotenoids from parent blood samples were
87 used to validate these parent vegetable/fruit behaviors [$r=.44$, $p<.001$] [Murphy et al. 2001; Townsend et al. 2003]. The four
88 items included vegetables as snacks, vegetable quantity, vegetables at main meal, and vegetable types. Parent responses were
89 coded using 4-response options per item for a maximum of four points per item. The four items were scaled and summed for a
90 maximum of 16 points. Our hypothesis was that parent behaviors would be positively related to scores on Focus on Veggies.

91 **3) Convergent validity with child 24-hour dietary recalls.** Recalls [$n=3$] were collected by trained researchers in person and by
92 telephone. The USDA 5-step multi-pass method included [Conway JM et al. 2003]: quick list of foods eaten the previous day; time
93 and eating occasion of each food; probes for missing foods; detailed description and amount of each food; and review of the
94 recall. Food recall data was entered into ESHA's Food Processor Nutrition Analysis Software [<http://www.esha.com/>; Salem, OR
95 97306] using the best possible match to the items reported by parents. ESHA's reports were generated to include a child's
96 nutrient intake and MyPlate cup equivalents. Because breakfast, lunch and snacks, were frequently provided by Head Start and
97 daycare and all parents in the dataset provided dinners, the mean from three diet recalls for the dinner meal was used for this
98 analysis. Our hypothesis was that vitamins A and C, folate, beta carotene, fiber, potassium and magnesium would be positively
99 related to scores on Focus on Veggies.

100 **4) Predictive validity with child anthropometry.** The child's height and weight were measured using digital scales and portable
101 stadiometer [Seca Road Rod, Hanover, MD, USA]. With children dressed in light clothing, with shoes, jackets or outer ware

102 removed, two trained research assistants weighed each participant twice to the nearest 0.1 kg. Height was measured twice to
103 the nearest 0.1 centimeter with the child fully erect, feet together without shoes, and head in the Frankfort Plane, and at the end
104 of a deep inhalation. Body Mass Index [BMI] (kg/m^2) was calculated as a continuous variable using the means of the height and
105 weight measurements. BMI-for-age percentile was then derived by using the Centers for Disease Controls BMI Percentile
106 Calculator for Children [Centers for Disease Control]. Measurements were taken at baseline and 12 months later. Our
107 hypothesis was that BMI-for-age percentile would be negatively related to scores on Focus on Veggies.

108 **5) Criterion validity with child blood samples.** The child arrived at his Head Start school with his parent after an overnight fast
109 (≥ 8 hr.) to have up to 10 ml of blood collected by venipuncture into serum and plasma vacutainers by a certified pediatric
110 phlebotomist. The EDTA plasma vacutainer was iced immediately while the two serum vacutainers remained at room
111 temperature for 30-mile transport to the laboratory at the USDA Western Human Nutrition Research Center on the UC Davis
112 campus.

113 Three carotenoids were selected for assessment, α -carotene, β -carotene, and retinol, because of the hypothesized positive
114 relationship with vegetable intake. The blood samples were analyzed for carotenes using high-performance liquid
115 chromatography [HPLC] [Barua AB et al. 1993]. The values for each carotenoid were ranked and then summed for an index with
116 each given equal weighting. We hypothesized that high scores on *Focus on Veggies* would be positively related to scores on this
117 carotenoid index.

118 Five biomarkers of inflammation were selected for assessment because of their hypothesized negative relationship with a diet
119 high in vegetables: interleukin-6 [IL-6], interleukin-8 [IL-8], tumor necrosis factor- α [TNF- α], retinol binding protein [RBP-4], and
120 C-reactive protein [CRP]. Although temporary acute inflammation is protective with an injury, elevated circulating values in the
121 uninjured well-child generally denotes chronic low grade inflammation, a situation not desirable for long term health [Tam et al.
122 2010]. These five cytokines are risk factors for chronic diseases, especially cardiovascular disease, diabetes and metabolic
123 syndrome [Tam et al. 2010] and obesity [Varma et al 2016]. Plasma IL-6, IL-8, TNF α , RBP-4 concentrations were measured by
124 electro-chemiluminescent immunoassay and plasma CRP by turbidimetric assay using commercially available kits [Meso Scale
125 Discovery/MSD, Rockville, MD and Integra 400+, Roche Diagnostics, Indianapolis, IN]. An internal standard added to each plate
126 assessed plate-to-plate variability. For each biomarker, a unique standard curve was generated and used to harmonize the inter-
127 assay variability. Assayed in duplicates, a given biomarker with a coefficient of variability greater than 25% was removed from

the dataset. The mean of duplicates was used for each biomarker. CRP was log transformed. Outliers defined as data greater than 3 X inter-quartile range were assessed case by case for consistency over time and metabolic plausibility, and retained if either criterion was met. An inflammation index was created by combining the rank values of the five cytokines. *Focus on Veggies* scores were hypothesized to be negatively related to the inflammation index.

Other psychometric properties: readability and respondent burden

Two other properties considered important for this audience and setting were assessed. Readability [Klare 1984] of *Focus on Veggies* was assessed by the Flesch-Kincaid Reading Index and Flesch Reading Ease using Microsoft Word software (MS Office for PC, Microsoft, Inc., Seattle, 2003). Respondent burden was assessed by timing respondents completing the tool.

Statistics

Pearson's Chi-squared test with Yates' continuity correction was used to compare demographic data for dropout and final samples with statistical program R [<https://cran.cnr.berkeley.edu>, V. 2014, Vienna, Austria]. Non-parametric Spearman rank order correlation coefficients were calculated to assess the direction and strength of relationship between two quantitative variables. Item analysis and tests for reliability were conducted using SPSS [version 21.0, SPSS Inc., Chicago, IL, 2015]. The item difficulty index from 0 to 1 was assessed by dividing the item score by the maximum item score. The item discrimination index from 0 to 1 was assessed using the correlation for each item to the total scale without that item. Stability reliability was assessed using the Spearman's rank order correlation test, 1-tailed, comparing data collected at two time points six weeks apart. Internal consistency reliability was assessed with Cronbach α .

Relationships between biomarkers and BMI with *Focus on Veggies* scores were non-linear. Consequently, linear regression models fit to the data failed to produce suitable models. Therefore, we discretized *Focus on Veggies* scores using tertiles to define low, medium and high vegetable behavior scores. We then compared BMI-for-age percentile across the vegetable groups using analysis of variance. Tertile medians of BMI for age percentile, biomarker indices and vegetable cup equivalents are presented in Table 4. While there was some skewness in the data and unequal variances across *Focus on Veggies* scores suggesting the use of Kruskal-Wallis [KW] test instead of ANOVA in some instances, presenting medians are appropriate to summarize subgroup results. Both methods assess differences in the mean of a continuous dependent variable across groupings defined by an independent variable (with three or more groups). Continuous BMI-for-age percentile as an outcome variable is regressed on quartiles of *Focus in Veggies* scores to generate R^2 , adjusted R^2 and partial R^2 .

154 Kendall's tau, a non-parametric equivalent to the Pearson product moment correlation, and KW tests were used in analyses
155 with nutrients from recalls. Boxplots of BMI, dietary variables and biomarker indices were created across *Focus on Veggies*
156 scores to display and confirm the direction of the findings [not presented]. Using our previous research with suggested
157 acceptable minimums [Townsend et al. 2003; Murphy et al. 2001; Townsend and Kaiser 2005], the study was powered at 80% to
158 detect a minimum correlation of 12% between Focus on Veggies scores and outcome variable measured as vegetable cup
159 equivalents and 95% to detect a minimum difference of .33 cup vegetables between the lowest and highest consuming
160 categories using Focus on Veggies scores to define categories.

161 RESULTS

162 Demographics

163 Parents self-reported race/ethnicity as: 20% Hispanic, 7% Latino, 35% African American, 20% white, 5% Asian, 0% American
164 Indian, 16% multi-ethnic, 2% other. Families enrolled in the study participate in at least one USDA assistance program, with WIC
165 (79%), Head Start (70%), and SNAP (61%) being reported most frequently. Parents (92% female) on average were 31±8 years with
166 children (42% female) enrolled being 3.3±1.6 years on average living in a household of 4.2 ±1.0 persons. About a third of parents
167 were married (28%) with a majority reporting a high school diploma (33%) or some college (56%). Parents [65%] reported
168 earning a household income of \$20,000 or less annually. Of the 176 potential eligible parent/child pairs, all were confirmed
169 eligible and enrolled in the study on Day 1. Most (n=133) completed baseline data collection. No significant differences were
170 found between the 133 parent/child pairs and the 43 pairs with only partial data. [p=.80-.99].

171 Reliability

172 The internal consistency of the 10-item vegetable scale was very good ($\alpha=0.83$) [Table 1]. Item difficulty indexes ranged from
173 a low of .33 for 'Child eats beans' to a high of .89 for 'Parent buys vegetables'. The interpretation is 33% of parents responded
174 that their child eats beans while almost all parents (89%) reported buying vegetables. All items fell between the desired score of
175 .2 -.8 [Table 1]. Item discrimination indexes ranged from a low of .05 for 'My child eats beans' to a high of .65 for 'My child eats
176 vegetables at his main meal'. Children with high scores on *Focus on Veggies* do not necessarily eat beans, as expected. An item
177 that discriminates as expected would score similarly to the others in the scale as indicated by a desired value of $r \geq .30$. This also
178 demonstrates that preferences for protective vegetable behaviors vary by families. Item stability ranged from $r=.31$ for 'Parent

179 buys vegetables' to $r=.69$ for "Child eats beans' with the sub-sample of parents not participating in an intervention [$n=98$]. The
180 former item did not meet the desirable cut point of $r \geq .40$. This item was the least reliable temporally meaning that there was
181 more variability in responses from the first administration of the tool to the second administration four weeks later. Two items,
182 'child eating beans' and 'child being a picky eater', were the most stable, meaning that these behaviors were perceived by
183 parents as being more consistent over time while their vegetable purchasing habits were less consistent. The 10-item scale
184 produced a test-retest reliability coefficient of $.74$ [$p \leq .01$], more than meeting the desired minimum of $.60$ for this low-income
185 audience [Table 1].

186 **Validation**

187 **Parent vegetable behaviors and mediators of vegetable behaviors.** As hypothesized, a scale score composed of three parent
188 mediators of vegetable behaviors assessed by the Fruit and Vegetable Inventory was positively related to the child's vegetable
189 score on *Focus on Veggies* [$r=.33$, $p<.00001$]. In addition, the parent behaviors [Table 3] assessed by the Food Behavior Checklist
190 were positively related to the child's score [$r=.57$, $p=\leq .00001$].

191 **Dietary.** The child's total score for *Focus on Veggies* was positively related to the volume of vegetables in cup equivalents
192 [$p=.031$]. The more favorable the child's vegetable behavioral score, the greater the intake of vegetables. A nutrient analysis
193 found many micronutrients significantly related to *Focus on Veggies* scores [Table 2]. These included among others vitamin C
194 [$p=.035$] and A [$p=.0008$], β -carotene [$p=.001$], magnesium [$p=.006$] and potassium [$p=.019$], and soluble fiber [$p=.0045$]. The
195 higher the *Focus on Veggies* score, the greater the calculated intakes of these vitamins, minerals and fiber offering support for
196 convergent validity. Non-significant relationships in the expected directions were calculated between dietary energy density and
197 folate, and the 10-item tool.

198 **Body mass index.** With ANCOVA adjusting for BMI-for-age percentile at baseline, there was a significant relationship between
199 BMI-for-age percentile measured 1 year later and the child's *Focus on Veggies* sub-score of six items ($p=.036$) [Table 4]. Boxplots
200 verified the negative direction of the relationship [not presented]. The healthier the child's score, the lower the child's BMI
201 percentile. A linear regression identified a negative coefficient and a category containing the healthiest vegetable behavior
202 scores being significantly different from the other three categories [$p=.030$]. A linear regression of BMI 1 year later on *Focus on*
203 *Veggies* scores controlling for BMI at baseline had an R^2 of $.63$, adjusted R^2 of $.61$ and partial R^2 of $.06$.

Blood samples. The inflammation index composed of a score of the five inflammatory biomarker values was significantly and negatively related to the score from the child's 10-item vegetable tool [$p=.044$] with a higher score on *Focus on Veggies* being associated with lower inflammation. The lower the score on *Focus on Veggies*, i.e. the child had less healthful vegetable behaviors, the greater the inflammation index from the 5 inflammatory biomarkers in the child's blood sample [Table 4]. The carotenoid index composed of a score of the three carotenoid biomarker values was significantly related to a sub-scale of 7 vegetable behavioral items [$p=.050$]. The greater the *Focus on Veggies* sub-scale score, the greater the carotenoid index calculated from retinol and carotenes [Table 4].

Other psychometric properties. The *Flesh-Kincaid Readability Index* produced a score of grade level 1-2. The *Flesh Reading Ease* produced a score of 99 of a possible 100 indicating the easiest reading. For administration of the 10-item *Focus on Veggies* in a group community setting, respondents took 4-8 minutes to complete on average, meeting our criteria for "rapid" assessment. Ten items with one- and two-syllable words and the pictorial format met our criteria for minimal respondent burden for a self-administered tool.

DISCUSSION

Our study provided data to support that a simple questionnaire can be used to render a rapid, reliable and valid assessment of parent-child vegetable behaviors. These findings supported the hypotheses specified in the Biopsychosocial Framework [Figure 2]. The parent psychosocial constructs and parent vegetable behaviors were strongly related to the child's *Focus on Veggies* scores. The child vegetable behaviors drove the intake of vegetables measured by the 24 hour diet recalls supporting the tool's convergent validity. The child's vegetable intake drove the child's nutrient values. Those nutrients drove the biomarker values from the child's blood sample. The negative relationship between the blood markers forming the inflammation index and the child vegetable behaviors offered support for criterion validity of the tool. These results can be interpreted to suggest that children with lower *Focus on Veggies* scores exhibited more chronic low-grade inflammation, a suggested precursor to excess weight gain and chronic disease [Tam et al. 2010]. Thus, this index could be considered an important indicator of the child's health status. Importantly, our tool can serve as a proxy to identify children at greater risk of having an adverse inflammatory index score and serum carotenoid profile.

228 Finally, the higher the child's vegetable score, the lower the child's BMI percentile 12 months later. The significant inverse
229 association with a sub-scale of six vegetable behaviors contributed some support for the predictive validity. The child's vegetable
230 behaviors score controlling for BMI percentile at baseline, was predictive of children who were more likely to have a higher BMI
231 percentile 1 year later with an R^2 of .63. The interpretation was that 63% of variance in BMI one year later is explained by Focus
232 on Veggies sub-scale scores and BMI at baseline. For example, two children with same BMI at baseline and different *Focus on*
233 *Veggies* scores will have significantly different BMI values one year later. The higher scoring child, i.e. more healthful vegetable
234 behaviors, will have a lower BMI. The partial R^2 of .06 means that 6% of the variance in BMI one year later is explained by the
235 *Focus on Veggies* scores. With numerous factors contributing to a child's BMI such as physical activity, screen time, diet, sleep,
236 genetics and epigenetics, 6% for Focus on Veggies is substantial. Nutrition professionals could use this information to target
237 limited resources for nutrition education to families of those identified children. Clinical staff at community medical clinics could
238 also use this information to guide discussions with parents. Parents could use this tool for self-assessment. And USDA nutrition
239 education programs targeting this audience could use this tool for program evaluation.

240 For professionals desiring a shorter tool, data suggests that the bean item is the preferred candidate for removal. Although
241 the item was very stable, it did not discriminate high from low scoring children. At the same time, retaining this item would give
242 more opportunity to capture change from an intervention given its low difficulty index score with this English speaking audience.
243 Thus we recommend that professionals using this tool consider retention of the item for program evaluation and removal for risk
244 assessment.

245

246 **Literacy issues.** Readability is excellent at grade level 1-2, important for the many federal program participants with literacy
247 issues. Employing visual information processing theories, we learned from previous work that a representative color illustrated-
248 text style assessment tool is the preferred choice of respondents in our California samples, facilitates understanding of test items
249 and results in increased readability compared to the traditional black/white text-alone [Townsend et al. 2008]. As noted, this
250 measure for USDA low-income respondents is easy to administer in a group by a paraprofessional educator or a professional
251 dietitian or researcher and has a low respondent burden taking 4-8 minutes to complete. Reduction of cognitive load for
252 parents with literacy issues can be accomplished with readability of grades 1-2, photographs demonstrating the behaviors and
253 substitution of text with visual images [Townsend et al. 2014; Townsend et al. 2008]. Avoiding a lengthy or complicated

assessment that can generate participant frustration is important for adults with literacy issues [Townsend et al. 2014]. In addition, more formal evaluation methods using parent-reported dietary recalls and food frequencies can be burdensome for participants and expensive for federally funded programs in terms of data entry and analysis software costs.

Social desirability bias. Incremental increases or decreases between biomarkers and BMI-for-age with the *Focus on Veggies* scores were not consistently observed. Instead, the categories associated with the least or most healthy behaviors as shown on boxplots differed from the other categories. In terms of a linear regression model, this would indicate a flat slope over the lower or higher range of scores with a change in slope at the other end. For example, we might observe a flat slope over the lower and middle ranges of Focus on Veggies scores and biomarkers with a sharply increasing slope for the higher scores. We can speculate about the reasons. Social desirability bias may play a role resulting in misclassification of children into categories of Focus on Veggies scores. Parents are hesitant to admit that they do not buy, prepare or serve vegetables to their children, however factual that may be. Thus, we expect over-reporting of child vegetable behaviors, and to a lesser extent, parent vegetable behaviors. This bias would have the effect of reducing the level of statistical significance when examining relationships between Focus on Veggies scores and outcome variables.

Comparisons to other studies. No validation studies of vegetable behavior assessment tools for 3-5 year old children were found for comparisons. Dave et al. [2012] validated a tool for the fruit and vegetable social support for older children, 6-12 year olds. Test-retest reliabilities were .56, .60, and .79 for three scales comparing favorably to .74 for Focus on Veggies. Internal consistency of .87 compares to our alpha of .83. No other data, such as diet recalls, blood samples and heights/weights, were collected to validate their tool. Despite the lack of vegetable validation studies for comparison, our clinical biomarker results are consistent with data reported in clinical studies using older audiences. Specifically, higher vegetable/fruit consumption is related to lower markers of inflammation in adolescents [Holt et al. 2009] and adults [Watzl B et al. 2005].

We also compared our results to those of other studies for young children. The studies from our literature search on vegetable questions and scales did not collect necessary data to validate their scales using an inflammation index, carotenoid index, BMI nor vegetable cup equivalents. Cullen et al. [2003] used parent and child-report accessibility and availability questions to validate vegetable accessibility and availability to the child. Kratt et al. [2000] assessed

279 availability of fruit and vegetables to the child with a parent questionnaire about the previous week. Galloway et al.
280 [2005] assessed the child's degree of picky eating using a subscale of the Child Feeding Questionnaire [Birch et al.
281 2001], and collected child's weight and height. Reimer et al. [2004] used content analysis of parent qualitative data to
282 validate vegetables at meals/snacks, meal planning and child eating beans. None of the studies used biomarkers in
283 their research, making this study groundbreaking in its approach.

284 **Sources of error.** It is important to consider the hypothesized nutrients as a 'cluster' of nutrients. Individually each
285 nutrient is weak in its clinical significance, but taken as a cluster of 8 nutrients and vegetable cup equivalents, the
286 evidence for a relationship between the child's vegetable and nutrient intakes and his *Focus on Veggies* score is
287 stronger and of more clinical significance. There are several explanations to justify the relationships and explain
288 some sources of random and systematic error associated with the tool. Focus on Veggies asks parents about usual
289 behavior, not specifics on a particular day. Behavioral items are simple for participants to answer whereas 24-hour
290 diet recalls to capture intakes is tedious and time consuming for the participant, and labor-intensive for nutrition
291 program staff. There are other food sources of these hypothesized nutrients in addition to vegetables. Fruit also
292 contributes to vitamin C. And wholegrains and fruit contribute to the fiber. Therefore a child could have a high
293 vitamin C intake without eating vegetables. In addition, we used intake at dinner to validate behaviors allowing for
294 some source of error.

295 **Limitations and strengths.** The primary strength of this study is use of multiple analytical approaches using the Biopsychosocial
296 Framework to confirm validity of *Focus on Veggies* [Figure 2] and the multiple approaches to inform reliability. The framework
297 for the study incorporates both subjective measures, i.e. parent reports of child's diet, and objective measures, i.e.
298 anthropometric and chemical biomarkers, to determine the validity of the parent responses on the *Focus on Veggies* tool. The
299 main advantage of using biomarkers for validation is their independence from parent report, with its inherent bias, mainly from
300 social desirability as described above. Although biomarkers are not without laboratory-based error, they do not reflect
301 systematic bias of the parent. Measured BMI is also free of parent bias, but is relatively insensitive to the child's diet and physical
302 activity in the near term and subject to clinical error.

303 The external validity of these findings to other limited-resource audiences is unknown. However, it is worth noting that
304 because these findings are consistent with theories of visual information processing (Townsend et al. 2008; Levie and Lentz
305 1982), with efforts to reduce client cognitive load for audiences with minimal literacy skills (Townsend et al. 2014), these results
306 may be applicable beyond this sample of study respondents. Variability of parent responses may be reduced compared to the
307 general limited-resource target population in California, because parents volunteered to participate at the 13 Head Start and 3
308 WIC sites. Consequently, selection bias must be considered as a threat to external validity [Cook and Campbell 1979]. Social
309 desirability bias influencing parent responses on all parent-report tools must be noted as a threat to internal validity. In addition,
310 the Hawthorne Effect from the 24-hour diet recalls may have served as a motivator for better food selection by parents [Cook
311 and Campbell 1979]. A next step involves testing *Focus on Veggies* with a broader sample from other sites in California, and then
312 other states.

313 **CONCLUSION**

314 Validity of *Focus on Veggies* has been demonstrated for these parents and children with vegetable cup equivalents and
315 hypothesized micronutrients calculated from 24-hour diet recalls, parent eating behaviors and psychosocial mediators of
316 vegetable behaviors, and child anthropometric and chemical biomarkers. *Focus on Veggies*, a simple assessment tool, can
317 inform practitioners about the child's health status. A child with a low *Focus on Veggies* score shows a less than healthful profile
318 with a higher inflammation index, lower carotenoid index, higher BMI-for-age percentile and lower vegetable intake. In contrast,
319 a child with a high *Focus on Veggies* score, shows a healthful profile with a lower inflammation index, higher carotenoid index,
320 lower BMI and higher vegetable intake.

321 Because rigorously validated tools for participants of these federal nutrition programs are few and even fewer for low-literate
322 participants (Townsend et al. 2009), other researchers could employ these methods in future validation research. In addition,
323 with prevention key, the tool could be a valuable health promotion opportunity for providing individualized feedback or crafting
324 personalized messages to parents enrolled in SNAP-Ed, EFNEP, WIC, or Head Start. The resulting pictorial assessment with low-
325 respondent burden may be useful in evaluating vegetable behaviors for risk assessment, program needs assessment and
326 evaluation among program participants of federal nutrition assistance programs.

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Table 1

10-item *Focus on Veggies*: Responses and 4 assessments of reliability to include item difficulty index, item discrimination index, test re-test reliability for items and scale, and internal consistency.

	Item content	Item mean ±SD ^a	Item Difficulty Index ^b	Item Discrimination Index ^c	Reliability/ Stability ^d
	α =.83 with 10 items	Minimum=1 Maximum=5			
	Goal		≥.20 ≤.80	≥.30	≥.30
	Child behaviors				
1	Eating vegetables	3.64 ±1.18	.73	.70	.55
2	Eating beans	1.65 ±.98	.33	.12	.69
3	Being picky eater	3.77 ±1.13	.75	.32	.67
4	Snacking on vegetables	3.74 ±1.13	.75	.62	.57
5	Eating vegetables main meal	3.26 ±.98	.65	.44	.47
6	Eating ≥1 kind	2.86 ±1.19	.57	.70	.60
	Parent behaviors				
7	Modelling vegetable eating	3.89 ±1.11	.78	.59	.58
8	Keeping veg ready to eat	3.61 ±1.27	.72	.66	.54
9	Buying vegetables	4.46 ±.81	.89	.55	.31

10	Planning meals	3.77 ±1.19	.75	.43	.48
	10-item <i>Focus on Veggies</i>				.74

^a Item mean ± the standard deviation with a minimum of 1 points and maximum of 5.

^b Item difficulty scores were generated by dividing the mean value of the responses to the item by 5, the maximum value possible on a 5-item scale.

^c Corrected Item-Total Correlation

^d Non-parametric Spearman rank order correlation, 1-tailed, at two time points 4 weeks apart with no intervention, coefficient *r*.

Table 2.

The relationship of 10-item Focus on Veggies with the child's vegetable cup equivalents and hypothesized vitamins and minerals.

		p-value ^a	Tau ^b
24 hour diet recalls [3, mean]			
Vegetables, volume	Cup equivalents	0.031	.17
Energy density	Cals/gm	.098	-.12
Vitamins			
	C [mg]	0.035	.17
	A [RAE]	0.0008	.13
	Folate [DFE mcg]	0.057	.18
Minerals			
	Potassium [mg]	0.019	.15
	Magnesium [mg]	0.0063	.18
Other nutrients			
	Beta-carotene [mcg]	0.001	.16
	Fiber [g]	0.023	.16
	Soluble fiber [g]	0.0045	.23

^a Usual assumptions for analysis of variance not met. Therefore continuous variable was divided into 4 categories with Kruskal-Wallis test.

^b Kendall's tau rank correlation

Table 3

The relationship of 10-item Focus on Veggies with the parent University of California Food Behavior Checklist and parent University of California Fruit and Vegetable Inventory.

	p-value	r
Fruit and Vegetable Inventory [Parent vegetable mediators] ^a		
	<.00001	.33
Food Behavior Checklist [4 parent vegetable items] ^b		
	<.00001	.57

^a Perception of diet quality, vegetable readiness to eat more, vegetable self-efficacy

^b Four vegetable items in the University of California Food Behavior Checklist include vegetables as snacks, vegetables as cups, vegetable variety, and 2 vegetables at main meal.

Table 4

The relationship of *Focus on Veggies* with inflammation index, carotenoid index, body mass index-for-age [BMI] percentile and vegetable cup equivalents.

	Median 1	Median 2	Median 3	p-value
BIOCHEMICAL				
Inflammation index ^a	317.5	246.0	227.5	.04
Carotenoid index ^b	143.5	191.0	224.5	.05
ANTHROMETRIC				
BMI-for-age percentile ^c	87.5	70.0	59.0	.04
DIETARY				
Vegetable cup equivalents ^d	.120	.303	.425	.03

^a Inflammation Index is composed of the sum of five biomarkers [CRP, IL-6, IL-8, TNF- α , and RBP-4]. ANOVA using the 10-item Focus on Veggies scale. Tertile categories for *Focus on Veggies* are 0-28, 28-42, 42-50 for a maximum of 50 points.

^b Carotenoid Index is composed of the sum of three biomarkers [α -carotene, β -carotene, retinol]. ANOVA using 7 vegetable items [child eating ≥ 1 kind, child eating vegetables main meal, child snacking on vegetables, child eating vegetables, child picky eater, child eating beans, parent modelling]. Tertile categories for *Focus on Veggies* are 0-16, 16-29, 29-35 for a maximum of 35 points.

^c ANCOVA, model adjusted for baseline BMI, using 6 vegetable items [Child eating ≥ 1 kind, child eating vegetables main meal, child snacking on vegetables, parent modelling, parent keeping vegetables ready to eat, parent buying vegetables]. Tertile categories for *Focus on Veggies* are 0-15, 15-27, 27-30 for a maximum of 30 points.

^d ANOVA using the 10-item Focus on Veggies scale. Tertile categories for *Focus on Veggies* are 0-28, 28-42, 42-50 for a maximum of 50 points.

Figures

Figure 1

10-item *Focus on Veggies* assessment tool for low-income families with young children.

Focus on Veggies

Name

Date

 / /

My child eats vegetables.

- ☐ rarely
 ☐ some days
 ☐ most days
 ☐ almost every day
 ☐ every day

I buy vegetables.

- ☐ rarely
 ☐ sometimes
 ☐ often
 ☐ very often
 ☐ always

My child eats snack foods like apples, bananas or carrots.

- ☐ rarely
 ☐ some days
 ☐ most days
 ☐ almost every day
 ☐ every day

My child sees me eat vegetables.

- ☐ rarely
 ☐ some days
 ☐ most days
 ☐ almost every day
 ☐ every day

My child eats more than one kind of vegetable a day.

- ☐ rarely
 ☐ some days
 ☐ most days
 ☐ almost every day
 ☐ every day

My child is picky about the food he eats.

- ☐ rarely
 ☐ some days
 ☐ most days
 ☐ almost every day
 ☐ every day

My child eats beans times a week.

- ☐ 0
 ☐ 1
 ☐ 2
 ☐ 3
 ☐ 4
 ☐ 5
 ☐ 6
 ☐ 7

I keep vegetables ready for my child to eat.

- ☐ rarely
 ☐ some days
 ☐ most days
 ☐ almost every day
 ☐ every day

I plan meals.

- ☐ rarely
 ☐ sometimes
 ☐ often
 ☐ very often
 ☐ always

My child eats vegetables at his main meal.

- ☐ 0
 ☐ 1
 ☐ 2
 ☐ 3 or more

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Figure 2

Figure 2. Biopsychosocial framework for validation of *Focus on Veggies* targeting low-income families with preschool-aged children.