

Original Article

Predictors of carotenoid status in New Zealand children using carotenoid reflection score: a cross-sectional study

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Background and Objectives: Optically-assessed skin carotenoid status offers an objective measure of vegetable and fruit intake. Data from the Sodium and Potassium Intake (SNaK+) study was used to explore associations of carotenoid reflection scores with body size, annual household income, vegetable and fruit intake and potassium intake, and the relationship between carotenoid reflection score and 24-h urinary potassium in children. **Methods and Study Design:** Seventy-five children (29 girls, 37 boys; 9 not stated) aged 8 to 13 years from five New Zealand primary schools were supported by the parents/caregivers to complete validated questions about their frequency of vegetable and fruit intake. Carotenoid reflection scores were measured using reflection spectroscopy and 24-h urine samples (n = 69) were collected. Differences by gender and age were tested using two-sample t-tests. Linear regression identified determinants of carotenoid reflection score. **Results:** Children's overall mean (standard deviation) carotenoid reflection score was 236 (91), with 51% having low scores (< 250). Younger boys (< 10 years) had higher carotenoid reflection scores than younger girls (mean difference (95% confidence interval); 74 (4, 144). Few (8%, n = 6) children met vegetable intake recommendations. Annual household income and fruit servings per day were predictive correlates of carotenoid reflection scores ($R^2 = 0.17$, $F(2,52) = 6.10$, $p < 0.01$). Twenty-four-hour urinary potassium did not correlate with carotenoid reflection score. **Conclusions:** This study identified low household income and low fruit intake as drivers of low carotenoid status in children, emphasizing the need for targeted dietary interventions. A study is required to further explore the relationship between carotenoid reflection score and potassium excretion.

Key Words: reflection spectroscopy, Veggie Meter, vegetable consumption, potassium, children

INTRODUCTION

Vegetables and fruit are essential for children's growth and development, supplying vital nutrients that optimise immunity,¹ gut function,² cognition,³ mental wellbeing,⁴ and overall health. Children who eat enough vegetables and fruit are more likely to adopt healthy eating habits such as eating a 'rainbow' of fruit and vegetables throughout their lives.^{5, 6, 7} Coloured vegetables and fruit also contain yellow/red/orange pigments known as carotenoids, which are antioxidants.^{8, 9, 10} The most abundant carotenoid is β carotene, a precursor of vitamin A found in apricots, carrots, pumpkin, tomatoes, and dark green leafy vegetables.¹¹ Further, vegetables and fruit are high in potassium, which is important for normal cell function and blood pressure,¹² the latter being particularly important for maintaining low blood pressure and preventing cardiovascular disease later in life.¹³

In Aotearoa New Zealand (NZ), an ethnically diverse country of approximately 5 million people with 17.8% identifying as tangata whenua (indigenous) Māori (n =

887,493) and 8.9% as from Pacific communities (n = 442,632), only 5% of children 2-14 years meet the recommended 2.5 to 5.5 servings of vegetables and 1-2 servings of fruit per day.¹⁴ These rates decline with age, with only 3% of 10-14 year-olds meeting vegetables and fruit recommendations.¹⁵ While suboptimal vegetables and fruit intakes exist similarly for girls, boys, and among different ethnicities, fewer children in the most deprived neighbourhoods eat enough fruit (67%), compared to

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those in the least deprived (76%). In countries like NZ including the UK and Australia, the situation is similar.^{16, 17} Consequently, several strategies have been developed to increase children's vegetables and fruit intakes, including improving access to vegetables and fruit, updating nutrition policies, free provision of school meals, and delivering education (e.g. cooking classes and garden activities).^{18, 19} Programs for NZ schools in low socioeconomic areas include Fruit in Schools (free daily fruit) and Ka Ora, Ka Ako (free healthy lunches started in 2019). These programs have been shown to somewhat improve children's vegetables and fruit intake,²⁰ however, more work is needed to address inequities and ongoing low vegetables and fruit intakes.

Accurate monitoring of children's vegetables and fruit intake is important to set a baseline for measuring the impact of interventions and public health policies. However, the best practice measure of multiple 24-h recalls is burdensome for both participants and researchers, can be difficult for young children, and is subjective.

A 2023 systematic review (n = 11 articles; n = 2,809 total participants) and meta-analysis concluded that assessing reflection spectroscopy-based carotenoid reflection score is a valid method to estimate children's vegetables and fruit consumption.²¹ However, authors acknowledged several methodological limitations, including the low number of studies in the meta-analysis (n = 4; n = 726 children) and limited information by age group. Notably, children aged 8–13 years are underrepresented in carotenoid status research, yet this developmentally distinct period — marked by increasing dietary autonomy and school-based influences — is important to include in validation studies. In addition, there were no studies conducted in New Zealand — an ethnically diverse population — aligning with the review's call to report carotenoid status in more diverse groups. No studies have yet explored the relationship between carotenoid reflection score and potassium intake, a key nutrient linked to vegetable and fruit consumption.

In 2022/2023 the Sodium and Potassium Intake (SNaK⁺) Study used reflection spectroscopy via the Veggie Meter™ to estimate vegetables and fruit intake in 75 New Zealand primary school children (8–13 years) living in Auckland and Dunedin. A gold-standard 24-h urine collection^{22, 23} was used to measure sodium and potassium intake, and validated questions assessed vegetables and fruit consumption.^{24, 25} This manuscript reports a secondary analysis of data from the SNaK⁺ study to explore potential correlates of carotenoid reflection score in New Zealand children and provide new evidence for the relationship between carotenoid reflection score and 24-h urinary potassium excretion.

METHODS

Setting

This study was conducted according to the guidelines laid down in the Declaration of Helsinki and all procedures involving human subjects were approved by the Central Health and Disability Ethics Committee (20/CEN/85). Written informed consent was obtained from all subjects. A study protocol for the SNaK⁺ study and other study-related documents are available on the open-source web-

site Figshare.²⁶ This is a secondary analysis and not powered in the main study protocol. However, for a prediction equation with up to three predictors (i.e., body size, household income, vegetable and fruit intake or potassium intake) and based on a moderate effect size of 0.15, a power of 0.80, and a probability level of ≤ 0.05 ,²⁷ it was determined retrospectively that the minimum sample size of 76 would be sufficient to detect an association with the outcome of interest, the carotenoid reflection score.

Participants

Five schools agreed to participate. Inclusion criteria for children taking part included: that they were in Years 3 to 6 (ages ~8 to 13 yrs); able to give written informed assent alongside parent/caregiver consent, and could speak and understand English. Participating children had to attend a NZ primary or intermediate school that had provided written informed consent (either paper form or electronic) from a member of the Board of Trustees, principal, and participating teacher. A total of 75 children had their carotenoid reflection measurements determined by trained researchers with expertise in nutrition. Demographic and dietary habits data including child age, gender, self-identified ethnic group, fruit and vegetable intake, discretionary food intake (fast food and sugary drinks), use of dietary supplements, and discretionary salt use were collected using an online questionnaire administered via REDCap software²⁸ and completed by parents/caregivers before study start (i.e. the baseline questionnaire).²⁶

Measurement of carotenoid-containing food intake

Total daily servings of fruit and vegetable intake were assessed using two items adapted from the 2020/21 New Zealand Health Survey (Children's Questionnaire), featuring show cards and standard serving size definitions (e.g., 1 medium fruit or ½ cup cooked vegetables = 1 serving). Parents/caregivers reported their child's usual daily intake of fresh, frozen, stewed, or canned forms, excluding juice and dried products.²⁵ Additionally, three targeted questions assessed intake of key carotenoid-rich vegetables — carrots/pumpkin, dark green leafy vegetables (e.g., silverbeet, spinach), and tomatoes — selected for their availability in New Zealand and importance as carotenoid sources.²⁹ These questions used the same serving size definitions and response scale, with tomato intake also including sauce (e.g., 1 tablespoon of tomato sauce = 1 serving). A sum of self-reported servings was then calculated.

Measurement of skin carotenoid concentration

The Veggie Meter™ (Longevity Link Corporation, Salt Lake City, UT, USA) was used to determine pressure-mediated carotenoid reflection score. This is a small, portable, non-invasive device that uses reflection spectroscopy to objectively measure skin carotenoids (Figure 1). The device provides a skin carotenoid reflection score on a scale from 0 to 800, with a higher score indicating more vegetables and fruit have been eaten in the previous two months and repeat measures detecting changes in as little as two weeks.³⁰ Adhering to the manufacturer's guidelines, children were asked to wash (with soap and water) and dry their hands. The right-hand index finger



Figure 1. Veggie Meter™ used to assess children's vegetable intake

was inserted into the designated port, pressing its fingertip against the contact lens. A spring-loaded clip gently ensured the finger remained in place. For consistency, three successive and separate carotenoid reflection score measurements were recorded for each participant, with the average of three being recorded for analysis. Between readings, the finger was briefly removed for reperfusion. Any notable inconsistencies such as an out-of-range measure led to a repeated measurement. The contact lens surface was cleaned with an optical cloth before the first measurement for each child. Before each measurement and every two hours thereafter the instrument underwent calibration using the manufacturer-provided dark and white reference wedges. The carotenoid reflection score was interpreted as low (< 250), moderate (251 to 529), or high (530 to 800);^{31, 32} these cut points were determined from the equivalence of carotenoid reflection scores with measurements of plasma carotenoid concentrations measured by the Canterbury Health Laboratories who state that the "normal" plasma carotenoid concentration is 1.5 to 3.0 $\mu\text{mol/L}$.^{32, 33} Canterbury Health Laboratories is an accredited diagnostic service provider in New Zealand, and this reference range reflects both published data and their validated internal clinical standards for children.

Twenty-four hour urine collection

Measurement of children's potassium intake was carried out by gold standard 24-h urine collection and analysis,³⁴ following the study protocol.²⁶ Briefly, children started their 24-h urine collection at school or home and finished it at home; they were instructed to discard their initial urine, record the time, and collect all voided urine over the next 24 h using provided bottles and jugs, returning them to school. Children and parents/caregivers were asked to report collection hours and any issues encountered on a provided form. After a researcher returned urine samples to the University of Auckland, the total volume of each collection was recorded and aliquots were prepared, labelled, and frozen at -4°C for laboratory analyses.

Samples were analysed for urinary potassium using a Hitachi Cobas C311 analyser, using an Ion selective electrode. Urinary creatinine was analysed on the Cobas C311 using a colorimetric assay based on the Jaffe method.³³ The molecular weight of potassium was used to convert mmol to mg of potassium.³⁵ Samples not exactly covering 24-h were normalised. Samples were considered incomplete if they met all the following criteria: collection was < 300 mL urine (total volume) and/or > 1 missed collection, collection time was < 20 h or > 28 h, and creatinine was < 0.1 mmol/kg body weight.³⁶

Statistical analysis

Demographic information was summarised using percentages for categorical variables. Continuous variables were checked for normality and summarised as the mean (SD). Two sample t-tests were used to assess statistical significance between outcomes for girls and boys, and older (> 10 years) and younger (≤ 10 years) children. Comparisons by ethnicity and annual household income were not possible due to small subgroup sample sizes. Few children were taking supplements, most of which were non-carotenoid, so further analysis was not conducted. Statistical tests were two-sided at a 5% significance level. Visual inspection of scatter plots and Pearson r for linear relationships or Spearman's rho r for non-linear relationships were used to determine correlation. Determinants of the carotenoid reflection score were entered stepwise into multiple linear regression to identify the best combination of predictors. Variance Inflation Factors < 2 confirmed the absence of collinearity. A fitted regression model was derived where carotenoid reflection score = $\beta_1 \times \text{predictor1} + \beta_2 \times \text{predictor2}$. The degree of change of carotenoid reflection score for one unit of change for the variable is shown by the β coefficient. The change in R^2 was used to report how much variance in the carotenoid reflection score was accounted for with the addition of each predictor. Analyses were performed using IBM SPSS statistics software (version 25.0, IBM Corp., Armonk, New York, NY, USA).

RESULTS

Participant characteristics

The mean (SD) age of children was 9.4 (2.1) years. Most participants were NZ European ($n = 47$ or 63%) and few identified as Māori ($n = 9$ or 12%) and Pacific ($n = 5$ or 7%). Most were living in a household with an annual income of \$NZ 100,000 or more ($n = 44$ or 65%), which is higher than the NZ average household equivalised disposable annual income of \$44,142 after housing costs in 2023.³⁷ Characteristics of children and caregivers/parents are described in Table 1 and Supplementary Table 1, respectively.

Carotenoid reflection scores

Seventy-five children had their carotenoid reflection scores measured, of which 29 (39%) were girls, 37 (49%) were boys and 9 (12%) did not state their gender. The mean (SD) carotenoid reflection scores overall and by gender (girls and boys) were 236 (91), 219 (106), and 247 (85), respectively (Table 2). Fifty-one percent of children had a low carotenoid reflection score (< 250), with 16% scoring between 250 and < 279 , and 33.3% scoring < 530 . No child scored higher than 530 (Figure 2). Younger (< 10 yrs) boys had higher carotenoid reflection scores than younger girls; the mean difference was 74 units (95% CI, 4, 144) (Table 2). No other significant differences were observed by gender or age group.

Intake of carotenoid-containing foods

As reported by parents, 8% ($n = 6$) of children consumed three or more servings of vegetables every day as recommended by national guidelines at the time of data collection (mean (SD), 1.9 (1.1) servings per day) and 72% ($n = 54$) were eating the recommended two servings of fruit (mean (SD), 2.1 (1.1) servings per day) (Figure 3, Supplementary Table 2 and 3). There was no significant difference between girls and boys (Supplementary Table 3). Approximately half of the children were eating none or less than one serving per day of dark green leafy vegetables (54%), carrots/pumpkin (56%), and tomatoes (58%) (Figure 2, Supplementary Table 2). The mean (SD) number of servings from the aforementioned carotenoid-containing vegetables for all children was less than one serving per day: dark green leafy vegetables = 0.3 (0.6), carrots/pumpkin = 0.5 (0.6), tomatoes = 0.4 (0.7). Differences by gender and age group were not significant (Supplementary Table 3 and 4, respectively).

Children who met daily fruit recommendations had a significantly higher carotenoid reflection score than those who did not (mean (SD): 252 (88) vs. 167 (86), respectively, $p = 0.002$). The mean difference (95% CI) carotenoid reflection score was higher for children who ate at least one serving of dark green leafy vegetables per day compared to those who ate less than one serve or none (41 (-8, 89)) (Supplementary Table 5).

Ten children (13%) were reported to be taking supplements, including multivitamins and other non-carotenoid-specific nutrients (Table 1).

Twenty-four-hour urinary potassium excretion

Sixty-nine (92%) children returned 24-h urine samples for assessment of urinary potassium. Of these, fifty-nine

(86%) met the criteria for completion (Table 1; Supplementary Table 6). The overall mean (SD) urinary potassium was (1567 (773) mg), for girls was (1438 (677) mg) and for boys was (1667 (867) mg).

Associations between carotenoid reflection score and determinants

As shown in Table 3, there was a weak negative correlation between body weight and carotenoid reflection score ($r = -0.24$, $p < 0.05$) and waist circumference and carotenoid reflection score ($r = -0.23$, $p < 0.05$) for all children, but not for girls and boys separately. Weak positive correlations were also observed between fruit servings per day and carotenoid reflection score ($r = 0.3$, $p < 0.05$), and annual household income and carotenoid reflection score ($r = 0.26$, $p < 0.05$) for all children.

Predictors of carotenoid reflection score

Stepwise regression analysis was performed to examine predictors of carotenoid reflection score. Height, weight, and BMI were excluded from the model due to collinearity. Variables specific to carotenoid vegetables were also excluded due to interaction with vegetable servings per day. Age, waist circumference, gender, ethnicity, vegetable intake per day, fruit intake per day, urinary potassium, and annual household income were entered into the model. The final model showed that two predictors explained a total of 17% of the variation: carotenoid reflection score = $193.31 - 88.33 \times (\text{household income}) + 24.80 \times (\text{fruit servings per day})$ ($R^2 = 0.17$, $F(2, 52) = 6.10$, $p < 0.01$) (Table 4; Supplementary Figure 1). Nonetheless, an annual household income of \$NZ 50,000 or less explained a total of 10% of the variation with each unit of decrease in annual household income associated with a decrease in the carotenoid reflection score of 100 units. Fruit intake per day explained an additional 7% of the variation with 1 serving a day adding 25 units to the carotenoid reflection score (Table 4).

DISCUSSION

This is the first study to use reflection spectroscopy via the Veggie Meter™ to measure carotenoid status in NZ primary school children; it also provides new insights for understudied correlates including body size, income, and potassium intake. Annual household income and daily fruit servings emerged as the strongest determinants of carotenoid reflection score, and urinary potassium excretion appeared unrelated. We also revealed a high prevalence of low carotenoid status, particularly among younger girls compared to boys, alongside overall suboptimal vegetable and potassium intake.

The positive association between annual household income and carotenoid reflection score likely reflects socioeconomic disparities that often limit vegetables and fruit access and consumption in low-income families.^{38, 39, 40} In NZ, children with food insecurity (i.e. limited or uncertain availability for nutritionally adequate, safe, and acceptable foods) have lower vegetables and fruit intake than those in food-secure households, with mothers of 8 and 12-year-old children reporting a limited variety of vegetables and fruit in the house because of a lack of money.⁴¹ Few other studies have examined household

Table 1. Characteristics of the 75 children who participated in the study

	Girls		Boys		Gender not stated		Total	
	Number	Mean (SD)	Number	Mean (SD)	Number	Mean (SD)	Number	Mean (SD)
Age (years)	29	9.6 (1.4)	37	9.5 (2.1)	9	8.9 (3.4)	75	9.4 (2.1)
Height (cm)	29	140.6 (10.4)	37	141.4 (8.3)	9	144.9 (5.5)	75	141.5 (8.9)
Weight (kg)	29	37.8 (14.0)	37	36.3 (8.9)	8	37.6 (10.1)	75	37.0 (11.2)
Waist circumference (cm)	29	64.0 (12.6)	37	63.9 (9.4)	9	63.4 (11.1)	75	63.9 (3.8)
BMI (kg/m ²)	29	18.0 (4.1)	37	18.3 (3.4)	9	19.0 (4.7)	75	18.3
Ethnicity (n=75)	N	%	N	%	N	%	N	%
Māori	3	10	6	16	-	-	9	12
Pacific	4	14	5	14	-	-	7	9
European	21	72	26	70	-	-	47	63
Asian	3	10	1	3	-	-	4	5
South Asian	3	10	8	22	-	-	11	15
Other	3	10	2	5	-	-	5	7
Not stated	-	-	-	-	-	-	7	9
Urine sample collection day								
Weekday (at school)	5	17	8	22	-	-	13	17
Weekend day (at home)	24	83	29	78	2	22	55	73
Not stated	-	-	-	-	7	78	7	9
Child health (n=68)								
Currently has a health or medical condition	6	22	9	24	1	11	16	24
Currently taking prescription medication	5	17	5	14	0	0	10	15
Currently taking a dietary supplement [†]	4	14	6	16	0	0	10	15
Household income (n=68)								
\$50,000 or less	5	17	3	8	0	0	8	12
\$50,001 - \$100,000	3	10	5	17	0	0	8	12
\$100,001 or more	17	59	26	70	1	11	44	65
Prefer not to say	4	14	7	19	1	11	12	18

[†]Reported supplements were multivitamins, vitamin C, iron, vitamin D, zinc, melatonin and/or magnesium.

Table 2. Carotenoid reflection scores and complete twenty-four-hour urinary potassium excretion overall and by age and gender

	Girls	Boys	Gender not stated	Total	Difference by gender	
	Mean (SD)	Mean (SD)	Mean (SD)	Mean (SD)	Mean difference Girls - Boys (95% CI) [†]	p-value
Carotenoid reflection scores	n=29	n=37	n=9	N=75		
All	219 (106)	247 (85)	247 (60)	236 (91)	29 (-18, 76)	0.23
<10 years (n=33) [‡]	203 (104)	277 (87)	245 (17)	244 (97)	74 (4, 144)	0.04
=>10 years (n=42) [§]	233 (110)	222 (76)	248 (26)	233 (87)	-11 (-75, 53)	0.73
24-hour urinary potassium excretion	n=21	n=33	n=5	N=59		
Potassium (mg/24 hrs)	1438 (677)	1667 (867)	1444 (423)	1567 (773)	-229 (-676 to 219)	0.31
24-hr volume (mL)	839 (451)	864 (357)	948 (488)	862 (397)	n/a	n/a
25-hr creatinine (mg)	629 (281)	689 (243)	721 (228)	670 (254)	n/a	n/a

[†]Two sample t-tests were used to assess statistical significance between values for girls and boys. Pooled methods were used due to no evidence of unequal variance. Bold font indicates statistical significance p<0.05.

[‡]girls, n=14; boys, n=17. [§]girls, n=15; boys, n=20. At the time of the study, adequate Intake (AI) for New Zealand boys (3000 mg/day) and girls (2500 mg/day) aged 9 to 13 years, which is also below the WHO recommendation of 3510mg/day.⁷⁷

Table 3. Correlation of carotenoid reflection scores with participant characteristics, diet and 24-hour urinary potassium

Determinant	Pearson Correlation Coefficient, r (95% CI) [†]		
	All	Girls	Boys
Age	-0.08 (-0.31, 0.15)	0.01 (-0.41, 0.44)	-0.22 (-0.51, 0.12)
Weight	-.24* (0.47, -0.01)	-0.25 (-0.59, 0.13)	-0.14 (-0.57, 0.23)
Height	-0.17 (-0.40, 0.06)	-0.21 (-0.59, 0.18)	-0.13 (-0.47, 0.22)
Waist circumference	-0.23* (-0.49, -0.05)	-0.28 (-0.64, 0.11)	-0.19 (-0.56, 0.16)
BMI	-0.09 (-0.32, 0.14)	-0.04 (-0.46, 0.39)	-0.12 (-0.47, 0.23)
Ethnicity [‡]	0.13 (-0.15, 0.31)	0.30 (-0.33, 1.24)	0.18 (-0.26, 0.82)
Vegetables/day	0.23 (-0.01, 0.49)	0.28 (-0.10, 0.66)	0.22 (-0.12, 0.58)
Fruit/day	0.30* (0.07, 0.56)	0.49** (0.18, 0.99)	0.11 (-0.21, 0.40)
Dark green leafy veg/day	0.03 (-0.23, 0.28)	0.13 (-0.27, 0.53)	-0.07 (-0.42, 0.27)
Carrots/pumpkin/day	0.1 (-0.15, 0.35)	-0.01 (-0.77, 0.46)	0.16 (-0.15, 0.40)
Tomatoes/day	0.21 (-0.03, 0.47)	0.31 (-0.06, 0.62)	0.24 (-0.13, 0.79)
24-hour urinary potassium	-0.078 (-0.36, 0.18)	0.11 (-0.53, 0.56)	-0.23 (-0.56, 0.13)
Household income	0.26* (0.02, 0.51)	0.40* (-0.07, 0.74)	0.23 (-0.08, 0.62)

[†]Spearman's rho reported for ethnicity and household income only.

[‡]Ethnicity coded as European = 1, Non-European = 0. Note. N = 75

Value of r < 0.3=none or very weak; r = 0.3 to < 0.5 = weak; r = 0.5 to 0.7 = moderate; r > 0.7 = strong.

*p < 0.05; **p < 0.01.

Table 4. Multiple regression analysis with skin carotenoid reflection score as the independent variable

Independent variable	Regression coefficient, β	SE, β	$^{\dagger}R^2$ (p)
Constant	193.31	26.33	(<0.001)
Household income \$50,000 or less	-88.33	35.13	0.091 (0.015)
Fruit servings per day	24.80	10.81	0.159 (0.026)

† Adjusted for number of predictors (age, gender, waist circumference, ethnicity, vegetable intake per day, fruit intake per day, urinary potassium and household income). Stepwise linear regression was used to identify predictors of carotenoid reflection score

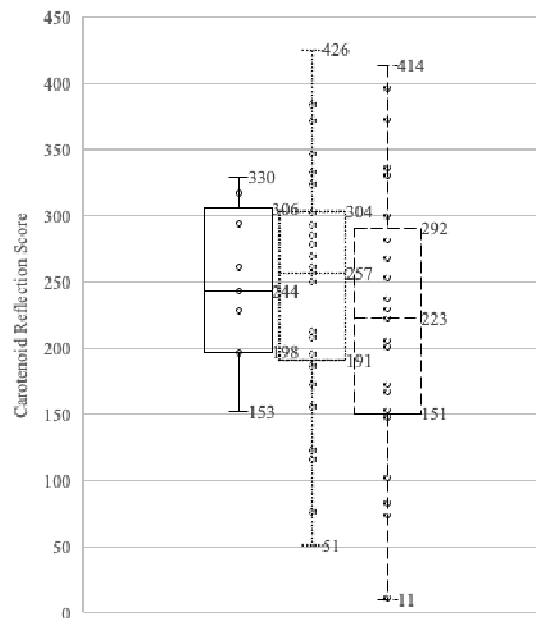


Figure 2. Box and whisker plot of carotenoid reflection scores for all children by gender. *Legend:* solid line = not stated; dotted line = boys; dashed line = girls.

income and carotenoid reflection score in children. One US study by Martinelli et al. (2021), involving 143 elementary school children (9–11 years) found students in higher-income schools had lower carotenoid status compared to those in lower-income schools (mean carotenoid reflection score = 201 and 221, respectively). High participation rates in free vegetables and fruit programs among low-income schools in the US may account for these findings. Only two of the five schools in the SNaK⁺ study received such programs. Other cross-sectional data in low-income population groups also present low carotenoid reflection scores, including a US after-school garden and nutrition program where mean carotenoid reflection scores of 174 and 192 were observed pre- and post-intervention, respectively ($p = 0.66$).^{42, 43} These findings support advocacy to provide targeted interventions for improving children's vegetables and fruit intake and thus carotenoid status while considering socioeconomic circumstances.^{31, 44}

Our study showed that one serving of fruit per day raises carotenoid reflection score by 25 points, which is unsurprising as most (71%) children met fruit recommendations and fruit contains carotenoids. Other research observes similar findings,^{45, 46, 47} for example, in 321 school children (aged 10 years) in Japan, higher fruit and green-yellow vegetable consumption were independently associated with higher carotenoid reflection scores ($r = 12.2$ and $p = 0.04$; $r = 20.8$, $p = 0.01$, respectively).⁴⁷

Likewise, Obana et al. (2022) reported higher carotenoid reflection scores amongst Japanese children (7–14 years; $n = 261$) consuming more green and yellow vegetables, vegetable/tomato juice, and fruits. However, the evidence is not always consistent as Martinelli et al. (2021) found a weak positive correlation between daily vegetable intake and carotenoid reflection score ($r = 0.174$, $p = 0.042$) and no significant relationship between orange/green vegetables and CRS. Our regression analysis similarly did not find servings of vegetables or carotenoid-rich vegetables including dark green leafy vegetables, tomatoes, or pumpkin/carrots as predictors of carotenoid reflection score. However, the very low intake of these vegetables in the SNaK⁺ study population, especially dark green leafy vegetables, may explain why no significant association was found. Alternatively, inconclusive findings highlight the complexities inherent in assessing the impact of the human diet on nutrient biomarkers, including nuanced effects of individual foods versus whole dietary patterns.⁴⁸ Regardless, concurrent low levels of vegetable intake and carotenoid reflection score underscore the broader and potentially more urgent issue of inadequate vegetable consumption for optimal health among children.

The mean carotenoid reflection score for children in our sample indicated a high prevalence of low carotenoid status. This is supported by suboptimal parent-reported vegetables and fruit intake, particularly as only 31% met

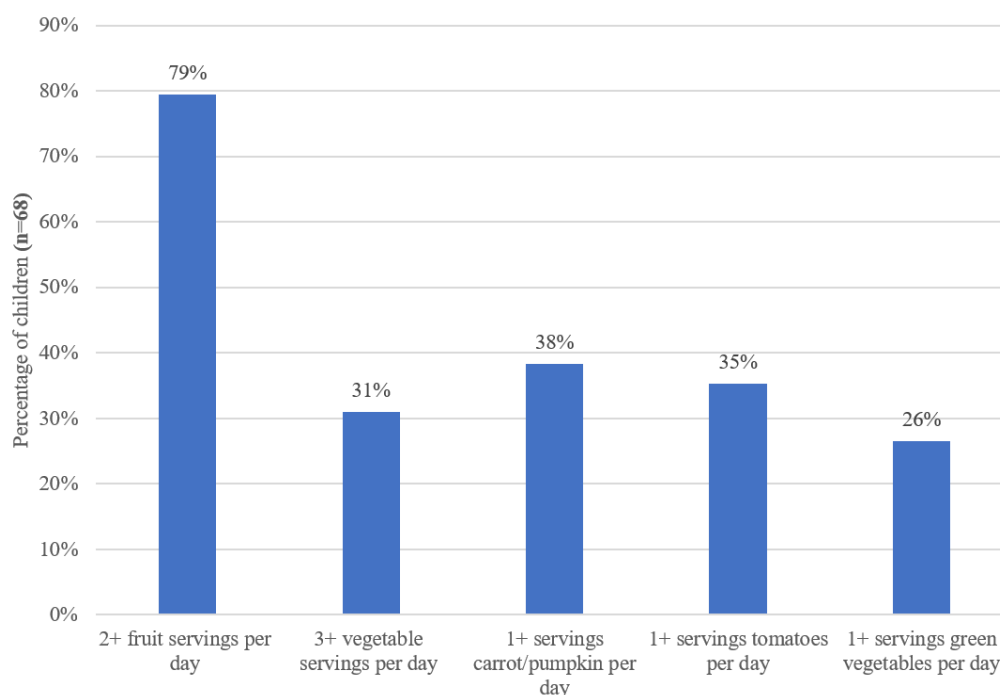


Figure 3. Percentage of children with optimal dietary behaviours for fruit and vegetables, and carotenoid foods as reported by parents/caregivers. Note. Analysis is based on the recommended dietary guidelines that were in place at the time of data collection (i.e. children consume at least 2 fruit servings and 3 vegetable servings per day); these have since been updated but not applied to the current analysis⁷⁸

vegetable recommendations, and is consistent with national statistics.⁴⁹ This is concerning as low carotenoid reflection scores and low vegetable consumption in childhood could track into adulthood, and increase the risk of chronic diseases.⁵⁰ Even lower carotenoid reflection scores are reported in US children, with mean scores ranging from 156 to 174.^{42, 46, 51, 52} The reason for our higher scores is unclear, especially as comparative studies lack comprehensive information on vegetables and fruit intake and data from the US National Health and Nutrition Examination Survey (2015–2018) found most (81%) of 6–11-year-olds ate fruit on a given day (albeit quantities unknown).⁵³ Conversely, higher carotenoid status has been observed among primary school children in Japan of 34947 and 448.⁴⁵ These differences in carotenoid reflection score by geographic location or ethnicity may be subject to the School Lunch Program Act^{54, 55} and cultural food preferences, especially as the Japanese-style diet emphasises grains, protein-rich foods (e.g., fish and meat), and vegetables.^{56, 57, 58} However, vegetables and fruit consumption is declining in Japan, with fruit being consumed less frequently compared to Western countries.^{56, 58}

In New Zealand, where adult carotenoid reflection scores³¹ and vegetable intake are also low,⁴⁹ and where adults strongly influence children's eating habits through role modelling and food availability,⁵⁹ it is plausible that the low carotenoid reflection scores observed in our study was influenced by broader family dietary patterns.⁶⁰ These patterns are further shaped by cultural and geographic differences in food access and preferences. Aotearoa New Zealand is home to a diverse population, including Māori, Pacific, and Asian communities, whose traditional food practices, access to land, and affordability

of culturally preferred vegetables and fruit may affect intake.^{61, 62} Regional access to fresh produce, particularly in rural or low-income areas, can also influence availability.⁶³ Although our limited sample size precluded analysis by ethnicity or region, future research could explore links between carotenoid reflection score, dietary patterns, and sociocultural backgrounds of family members to better inform culturally appropriate and equitable interventions.⁶⁴

The reason for higher carotenoid reflection scores in younger boys compared to girls of the same age, although still below the threshold of 250, is unclear but was also reported by May et al., 2020 in the US (i.e. higher carotenoid reflection scores in preschool boys than girls (283 ± 75 and 243 ± 89 , respectively; $p = 0.016$)). However, another study in US elementary school children found no gender differences.⁵¹ Variability in findings may be due to early life nutrition (i.e. duration of breastfeeding), genetics, age, body size, fat distribution, the food matrix, or other influencing factors.^{65, 66} For example, larger children may need to eat more carotenoid-rich vegetables and fruit to achieve the same tissue carotenoid concentration as smaller children due to the fat-soluble nature of carotenoids.^{66, 67} This factor could explain why girls had lower carotenoid reflection scores in our study since girls have been shown to have a higher body fat percentage than boys in New Zealand.⁶⁸ Aligning with Obana et al. (2022), BMI was not a predictor of carotenoid reflection score in our sample, but the prevalence of obesity in both studies was low and BMI lacks specificity.⁶⁹ Additionally, the narrow age ranges of the children measured could explain why no association between age and carotenoid reflection score was found. Moreover, as children's body size and eating habits evolve with age, it is premature to

rule out the influence of age and BMI on carotenoid reflection score, particularly given global trends of lower vegetables and fruit intake among older children.^{59, 70} Thus, further research comparing carotenoid reflection scores in children of varying ages, genders, and body size is warranted.

The absence of a relationship between urinary potassium excretion and carotenoid reflection score was unexpected, as it is reasonable to assume that both would reflect dietary fruit and vegetable intake. Our results may reflect the secondary nature of the analysis, with the SNaK⁺ study primarily focused on measuring sodium and potassium intake concerning cardiovascular health. Other constraints, such as a small sample size and participant preference to collect urine over the weekend when overall dietary quality is typically lower, rather than on a school day⁷¹ could have skewed findings. To our knowledge, no other studies have compared gold standard urinary potassium excretion with carotenoid reflection score,⁴⁸ but instead, examine the relationship between potassium intake and vegetables and fruit intake^{72, 73, 74} or dietary sources of potassium.^{34, 75} For instance, a cross-sectional study among 8-year-old children ($n = 330$) in Australia found overnight urinary potassium correlated significantly with self-reported vegetables and fruit intake ($r = 0.12$, $p = 0.03$).⁷² It is speculative but possible that a similar correlation with carotenoid reflection score might have been found if it had been included in their study design. More work is needed to clarify findings, though addressing ethical issues, significant participant burden, and cultural concerns related to 24-h urine collection in children is essential.^{34, 76, 77}

A notable strength of this study is that it is the first to assess carotenoid status among NZ primary school children, offering comparative evidence with other countries. Our data responds to research gaps identified in a 2023 systematic review,²¹ reporting on less-studied factors influencing carotenoid reflection score, such as BMI and socioeconomic variables, and introducing novel urinary potassium data. Our findings reinforce the Veggie Meter'sTM feasibility for assessing children's vegetables and fruit intake in primary school settings, yet there are limitations. The small, non-representative sample was impacted by COVID-19 disruptions and local flooding, thus limiting generalisability. In particular, limited sample diversity and size precluded comparisons by ethnicity and resulted in broad coding of ethnicity that oversimplified diversity and our analysis. Despite conducting collinearity diagnostics, it is possible that multicollinearity of closely related variables (e.g., potassium excretion vs vegetable intake, weight vs BMI) was present, decreasing the precision and power. Furthermore, although an objective measure of urinary potassium was undertaken using a gold-standard 24-h urine collection alongside comprehensive participant support and sensitivity analysis, findings may have been clearer with multiple 24-h collections evenly distributed over the week. There is also evidence that 24-h urine may underestimate potassium intake.⁷⁶ The reliance on cross-sectional, self-reported vegetables and fruit intake, albeit validated through a food frequency questionnaire, introduces inherent biases. In addition, we did not assess other carotenoid-rich foods (e.g. eggs,

dairy, fish), collect specific data on types of vegetables and fruit (e.g., intake of banana may have skewed potassium results), or control for several factors affecting carotenoid reflection score, such as genetic variations, presence of melanin, and adiposity. Supplement use was infrequent and largely non-carotenoid, suggesting minimal impact on findings. Finally, more information on the family diet and household food insecurity, including variables known to impact vegetables and fruit consumption in NZ (e.g. receiving financial assistance, and food bank participation) could have guided more targeted recommendations.

Conclusion

Although this study was small, results suggest low carotenoid status among NZ children, primarily influenced by household income and daily fruit servings. Our findings corroborate with national vegetables and fruit intake data^{24, 49} and highlight the need for interventions and policies to increase vegetable intakes within budget constraints. Leveraging NZ's strong horticulture sector, strategies like removing goods and service tax from vegetables and fruit, increasing government assistance to low-income families, nationwide free vegetables and fruit in schools, and emphasising vegetables and fruit diversity including carotenoid-rich vegetables within food-based dietary guidelines could also be implemented. Future research aiming to elucidate factors influencing children's carotenoid reflection score should integrate targeted interventions and policies for low-income households and involve larger, diverse samples. Understanding the relationships between carotenoid reflection score and other potential vegetables and fruit intake markers, like potassium urinary excretion, is essential for broader use of reflection spectroscopy via the Veggie MeterTM in monitoring and assessing the impact of new interventions and policies. Together, these insights will guide researchers, health professionals, and policymakers in effectively customising and evaluating interventions and dietary advice for children and families. Finally, we have underscored the urgency of improving children's dietary habits.

SUPPLEMENTARY MATERIALS

All supplementary materials are available upon request to the editorial office.

DATA AVAILABILITY STATEMENT

The study protocol and dissemination documents are freely available on the University of Auckland data sharing website Figshare. However, the participant data associated with our study is not available, in line with our ethics approval process.

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