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### **Abstract**

*Objective:* To examine the associations of dietary diversity with anaemia and iron status among primary school-aged children in South Africa.

*Design:* An analysis was conducted with pooled individual data from the baseline surveys from three previously conducted independent intervention studies. Two different dietary diversity scores (DDS) were calculated based on data from 1-day (1-d) and 3-day (3-d) dietary recall periods, respectively. Logistic regression analysis was performed to examine the associations of dietary diversity with anaemia and iron status.

Setting: KwaZulu-Natal and North West provinces, South Africa.

Participants: Children (n 578) 5- to 12-year-old.

Results: A DDS  $\leq$  4 was associated with higher odds of being anaemic (1-d P=0.001; 3-d P=0.006) and being iron deficient (ID) (3-d P<0.001). For both recall periods, consumption of 'vegetables and fruits other than vitamin A-rich' and 'animal-source foods (ASF)' was associated with lower odds of being anaemic (both P=0.002), and 'organ meats' with lower odds of being ID (1-d P=0.045; 3-d P<0.001). Consumption of 'meat, chicken and fish' was associated with lower odds of being anaemic (P=0.045), and 'vegetables and fruits other than vitamin A-rich', 'legumes, nuts and seeds' and 'ASF' with lower odds of being ID for the 3-d recall period only (P=0.038, P=0.020 and P=0.003, respectively).

*Conclusion:* In order to improve anaemia and iron status among primary schoolaged children, dietary diversification, with emphasis on consumption of vegetables, fruits and ASF (including organ meats), should be promoted.

Keywords
Dietary diversity
Anaemia
Iron deficiency
School-aged children
South Africa

The global prevalence of anaemia is about 34% affecting approximately 2·36 billion people, including 600 million children<sup>(1,2)</sup>. Nutritional anaemia is a common condition in primary school-aged children that results from a lack of certain vitamins and minerals, including iron<sup>(2)</sup>. The potential consequences of nutritional anaemia in children include loss of appetite, growth retardation, reduced cognitive development and ability to concentrate, which ultimately has a negative impact on schoolwork performance<sup>(3)</sup>.

Food consumption is one of the most important factors in the aetiology of nutritional anaemia and iron deficiency (ID) in school-aged children<sup>(4,5)</sup>. The intake of a great variety of foods across and within the various food groups is needed to meet nutrient requirements and may help to

alleviate multiple micronutrient deficiencies and reduce the risk of nutritional anaemia<sup>(6–8)</sup>. In low- and middle-income countries, including South Africa, the diets of the majority of children lack variety and consist mainly of plant-based foods (predominantly cereals, roots and tubers), with limited consumption of animal-source foods (ASF), fruits and vegetables<sup>(6,9–11)</sup>. Individuals who consume mostly cereal-based diets are at greater risk of developing ID due to the high content of iron inhibitors, such as phytic acid, in the diets<sup>(12,13)</sup>. In contrast, increasing the intake of ASF, and fruits and vegetables rich in vitamin C and carotenoids may increase the absorption of iron from cereal-based diets<sup>(14)</sup>. In addition, consumption of ASF contributes to dietary intake of nutrients such as highly

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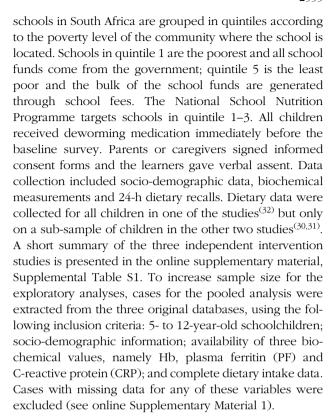
bioavailable haem iron, vitamin B<sub>12</sub> and vitamin A and plays an important role in reducing the risk of developing anaemia(15-17). While some studies have reported an association between dietary diversity and biomarkers of iron status in different age groups<sup>(18–20)</sup>, other studies showed no association between dietary diversity and anaemia<sup>(21,22)</sup>. Furthermore, in pre-schoolchildren in Ethiopia, a low prevalence of anaemia was observed even though children consumed a diet with low food variety<sup>(23)</sup>. Evidence on the relationship of dietary diversity with iron status and anaemia is therefore inconclusive.

Dietary diversity can be evaluated using simple tools such as dietary diversity scores (DDS), that is, the number of food groups consumed over a reference period<sup>(24)</sup>. Several studies have shown that these scores are good proxies of overall dietary quality<sup>(25-27)</sup>. However, the cutoff point for determining low DDS and the reference period on which the DDS is based may differ between studies. A DDS that was calculated based on a 1-day (1-d) reference period did not show an association with anaemia in pregnant women resident in rural areas of Northern Ghana<sup>(22)</sup>. Furthermore, a study in South African schoolchildren found no association between dietary diversity and anaemia using a 1-d reference period, but when a longer 7-day (7-d) reference period was used, low dietary diversity was significantly associated with anaemia risk<sup>(19)</sup>. Thus, although dietary diversity may be important in preventing nutritional anaemia and ID, there is limited knowledge on how the diversity of diets as a whole and the consumption of specific food groups are related to anaemia and ID in school-aged children.

The FAO uses a single 24-h recall period to calculate DDS because of less recall error compared with longer recall periods<sup>(28)</sup>. Collecting dietary data using 24-h recalls can be repeated on several non-consecutive days, with 3- to 7-d being the most widely used period for the purpose of an appropriate estimation of an individual's usual intake<sup>(29)</sup>. The aim of this paper is to describe the association of dietary diversity based on two different reference periods (a single 24-h recall v. three 24-h recalls) with anaemia and ID among primary school-aged children in South Africa.

# Methods

An analysis was conducted with pooled individual data from the baseline surveys from three independent intervention studies in schoolchildren at different sites in South Africa, which have been previously reported<sup>(30–32)</sup>. The original studies were conducted between April 2009 and June 2012 at primary schools in low socio-economic areas in KwaZulu-Natal (KZN) and North West (NW) provinces. These areas were malaria-free. At the time of data collection, all schools were participating in the National School Nutrition Programme of South Africa, which provided children with a daily meal on school days. Public



### **Biochemical measurements**

In the original studies, blood samples collected from the children were used to analyse biochemical indicators. For the pooled data analysis, the Hb, PF and CRP concentrations were used. In the study in KZN, the Hb concentrations were measured in whole blood through the direct cyanmethemoglobin method (Bio Rad Laboratories [PTY] Ltd) by using Drabkin's solution and a standard miniphotometer<sup>(30)</sup>, while in the two studies in NW, a haematology analyser (Coulter® Ac·TTM 5diff CP; Beckman Coulter) was used<sup>(31,32)</sup>. The PF and CRP concentrations were measured using an automated chemiluminescent immunoassay system (CLIA, IMMULITE) in the KZN study<sup>(30)</sup>, while in the two studies in NW, a ferritin ELISA kit (Ramco Laboratories Inc.) was used for PF and an immunoturbidimetric method (Technicon RA-1000 auto analyser) for CRP<sup>(31,32)</sup>. While the direct cyanmethemoglobin method and chemiluminescent immunoassay system used in the KZN study are the reference methods for Hb and PF (and CRP) measurements, respectively, the methods used in the two studies in NW are reliable and have comparable accuracy (33,34). The PF values of children with a CRP concentration ≥5 mg/l were adjusted for inflammation by multiplying PF values with a correction factor of 0.65<sup>(35)</sup>. Anaemia was defined as Hb < 115 g/ and ID as adjusted PF < 15  $\mu$ g/l<sup>(36,37)</sup>.

# Dietary diversity assessment

In all three intervention studies, dietary data were collected using the multiple-pass 24-h dietary recall method. In all studies, the 24-h dietary recalls were done approximately





1 week apart on three non-consecutive days, consisting of two weekdays and one weekend day, and thus were done over a period of 2-3 weeks. Children and their parents or caregivers were interviewed by trained fieldworkers and were asked to recall all foods and beverages items consumed the previous day. Similar dietary assessment aids to estimate portion sizes were used in the three studies which included plastic food models, household utensils, food packaging materials and 'dish-up and measure' (the participant used dry oats or rice to indicate the portion size for cooked food which was then quantified by the fieldworker using a measuring cup). Amounts reported in household measures or volume were converted to grams using the Medical Research Council Food Quantities Manual<sup>(38)</sup>. The energy and nutrient intakes for the pooled group and each of the study sites have been reported elsewhere<sup>(39)</sup>.

For the purpose of this study, foods reported during the 24-h recalls were categorised into the following groups: (i) starchy staples (combination of cereals, white roots/ tubers), (ii) dark green leafy vegetables, (iii) other vitamin A-rich fruits and vegetables (including vitamin A-rich vegetables, tubers and fruits), (iv) other fruits and vegetables, (v) organ meat, (vi) meat, chicken and fish, (vii) eggs, (viii) legumes, nuts and seeds and (ix) milk and dairy products<sup>(28)</sup>. A score of '1' was assigned to each food group if at least one food item within the specific food group was consumed during the reference period. A score of '0' was assigned if the child did not consume any food item from a given food group. The DDS for each child was calculated as the sum of the scores, with a maximum possible score of '9'. According to the guidelines, the mean DDS value can be used as cut-off point to indicate low dietary diversity<sup>(28)</sup>. Based on the mean DDS for the pooled group, low dietary diversity was therefore defined as DDS ≤ 4. In addition, an animal-source foods score (ASFS) was calculated based on five ASF, namely: (i) organ meat, (ii) red meat (beef mince, beef and pork sausages, processed cold meat), (iii) white meat (chicken meat), (iv) fish and (v) eggs. A score of '1' was assigned to each of these groups if at least one food item within the specific food group was consumed during the reference period. The ASFS for each child was calculated as the sum of the scores given, with a maximum possible score of '5'(28). Two different DDS were calculated: one for a 1-d recall period (1-d DDS) and one for a 3-day (3-d DDS) recall period. The 1-d DDS was calculated as the number of different food groups consumed during the first 24-h recall period. The 3-d DDS was calculated as the number of different food groups consumed during the three 24-h recall days.

# Statistical analyses

The statistical package SPSS version 25 (Inc.) was used for all statistical analyses. Pearson's  $\chi^2$  test was used to compare the proportions of children consuming specific food

groups, including the ASF group, and the percentage of children below the cut-off values for the DDS and ASFS. The paired sample t test was used to compare the mean DDS and ASFS between the two reference periods. The t test was used to compare the proportions of children who consumed specific food groups according to anaemia and iron status. Logistic regression analysis was done with the binary outcome (anaemic v. non-anaemic and ID v. non-ID) as the dependent variables and DDS and foods groups as independent variables. The results are presented as OR and 95% CI only for food groups that showed statistically significant results in at least one of the reference periods. The potential confounders (age, gender and study site) were included in the model. Statistical significance was set at P < 0.05 and a trend as P < 0.1.

### Results

# Characteristics and anaemia and iron status of children in the pooled group

The mean age of the children (n 578) was 8-7 (sp 1-3) years, the gender distribution was 51-0% boys and 49-0% girls, and 13-8% of the children were anaemic, 27-7% were ID and 19-8% presented with elevated CRP levels. Results on the biochemical indicators of anaemia and iron status for each of the study sites have been reported elsewhere<sup>(39)</sup>. In short, differences across study sites were observed. Compared with the other two study sites, the KZN study site had a significantly higher proportion of anaemic children ( $26.5 \ v.\ 14.0 \ and 10.4 \ %$ , respectively; P < 0.001) and ID children ( $42.2 \ v.\ 18.8 \ and <math>26.3 \ %$ , respectively; P < 0.001). The proportion of children with raised CRP levels was higher in NW2 ( $22.8 \ \%$ ) than in the other two study sites ( $19.6 \ and \ 8.0 \ \%$ , respectively; P < 0.01).

For the pooled group, the mean Hb concentrations for anaemic (n 80) v. non-anaemic (n 498) children were 111 (95 % CI 108, 113) and 128 (95 % CI 127, 129) g/l, respectively, and adjusted PF concentrations for ID (n 160) v. non-ID (n 418) children were 9·8 (95 % CI 7·8, 10·3) and 31·2 (95 % CI 27·9, 34·8)  $\mu$ g/l, respectively.

# The proportions of children who consumed specific food groups, the mean DDS and ASFS as calculated for two reference periods

Table 1 presents the results of the proportions of children who consumed specific food groups, and the mean DDS and ASFS as calculated for two reference periods. All children consumed foods from the 'starchy staples' group for both recall periods, but a significantly higher proportion of children consumed foods from certain groups, in particular, 'vegetables and fruits' (other than vitamin A-rich), 'organ meat', 'eggs' and ASF groups, according



**Table 1** Proportions (%) of 5- to 12-year-old schoolchildren who consumed foods from specific food groups, according to the 1-d and 3-d 24-h recall periods\*

	1-d 2	24-h ( <i>n</i> 57	8)	3-d 24-h ( <i>n</i> 578)†			
Food groups and scores	Mean	n	SD	Mean	n	SD	Р
Starchy staples (cereals/white roots)		100.0			100.0		1.000
Dark green leafy vegetables		26.1			27.5		0.631
Vitamin A-rich vegetables and fruits		13.2			13.8		0.709
Vegetables and fruits other than vitamin A-rich		66.2			68.2		0.048
Meat, chicken and fish		73.0			73.4		0.823
Organ meat		9.5			10.6		0.022
Eggs		20.9			30.7		0.010
Legumes, nuts and seeds		51.9			58.1		0.042
Milk and dairy products		30.3			31.8		0.358
ASF		78-1			82.5		0.035
DDS ≤ 4		43.3			37.5		0.012
DDS	4.69		1.2	4.84		1.1	<0.001
ASFS	2.42		1.4	2.91		1.2	<0.001

<sup>1-</sup>d, 1-day; 3-d, 3-day; DDS, dietary diversity score; ASF, animal-source foods; ASFS, animal-source foods score.

to the 3-d recall period compared with the 1-d recall period (all P < 0.05). The mean DDS and mean ASFS were significantly higher for the 3-d recall period compared with the 1-d recall period, and the proportion of children with DDS  $\leq 4$  was significantly lower for the 3-d period compared with the 1-d period.

# The proportions of children who consumed specific food groups over the two reference periods according to anaemia and iron status

Children were stratified according to their anaemia and iron status (Table 2), and the proportion of children who consumed different food groups and the means of the DDS and ASFS for the two reference periods were compared between these sub-groups. A significantly lower proportion of anaemic (v. non-anaemic) children consumed foods from the 'vegetables and fruits (other than vitamin A-rich)', 'organ meat' and the 'ASF' groups for both reference periods. Furthermore, for the 3-d reference period, it was found that a significantly lower proportion of anaemic (v. nonanaemic) children consumed foods from the 'meat, fish, and seafood' and 'eggs' groups, and a significantly lower proportion of ID (v. non-ID) children consumed foods from the 'legumes, nuts and seeds' group. Food groups consumed by the lowest proportion of children were 'vitamin A-rich vegetables and fruits' and 'organ meat'. The proportion of children with DDS  $\leq 4$  was significantly higher in the anaemic sub-group (v. non-anaemic) for both recall periods. The means of the DDS were significantly lower in the anaemic and ID sub-groups (v non-anaemic and non-ID, respectively), according to both reference periods, but the mean ASFS was significantly lower for the 3-d reference period only.

# OR and 95% CI for baving anaemia and ID for children according to the food groups consumed and DDS cut-offs

Results from binary logistic regression models with anaemia and ID as dependent variables and food groups and DDS categories calculated from both reference periods as independent variables are shown in Table 3. Consumption of 'vegetables and fruits other than vitamin A-rich' and 'ASF' was associated with lower odds of being anaemic, and consumption of 'organ meats' was associated with lower odds of being ID for both reference periods (all P < 0.05). Consumption of 'meat, chicken and fish' was associated with lower odds of being anaemic, and 'vegetables and fruits other than vitamin A-rich', 'legumes, nuts and seeds' and 'ASF' with lower odds of being ID for the 3-d reference period only (all P < 0.05). A DDS  $\leq 4$  was associated with higher odds of being anaemic for both reference periods (1-d P = 0.001 and 3-d P = 0.006) and higher odds of being ID for the 3-d recall period only (P < 0.001).

# Discussion

Associations of dietary diversity and consumption of different food groups with anaemia and ID were investigated in a pooled sample of primary school-aged children in South Africa. Results showed that children who consumed a diet with low variety (DDS  $\leq$  4) were more likely to be anaemic and ID. These results are similar to other studies that showed a positive association between dietary diversity and haematological indices (18,20), and that a low dietary diversity was associated with anaemia (19). Furthermore, children who consumed foods from the 'vegetables and



<sup>\*</sup>Continuous data reported as mean and sp, categorical data presented as percentages. Paired t test was used to compare the means;  $\chi^2$  test was used to compare the proportions. Statistical significance was set at P < 0.05 and a trend as P < 0.1. †Three non-consecutive days.



Table 2 Proportions (%) of 5- to 12-year-old schoolchildren who consumed specific food groups over the two reference periods according to anaemia and iron status\*

	Sub-groups of children (n 578)													
	Anaemic (n 80)		Non-anaemic (n 498)			ID ( <i>n</i> 160)			Non-ID ( <i>n</i> 418)					
Food groups and scores	Mean	n	SD	Mean	n	SD	P	Mean	n	SD	Mean	n	SD	Р
Starchy staples (cereals/v	hite roots	s)												
1-d 24-h		100.0			100.0		1.000		100.0			100.0		1.000
3-d 24-h†		100.0			100.0		1.000		100.0			100.0		1.000
Dark green leafy vegetabl	es													
1-d 24-h		20.0			27.4		0.217		26.9			25.8		0.830
3-d 24-h		29.9			27.4		0.190		31.7			25.8		0.063
Vitamin A-rich vegetables	and fruits	3												
1-d 24-h		5.0			14.5		0.019		11.9			13.6		0.681
3-d 24-h		11.3			14.3		0.304		13.8			13.9		1.000
Vegetables and fruits other	er than vit	amin Ā-ı	rich		_									
1-d 24-h	,	33.8			71.5		0.001		66.9			66.0		0.833
3-d 24-h		30.0			74.3		0.001		59.4			71.5		0.007
Meat, chicken and fish		000												0 00.
1-d 24-h		65.0			74.3		0.057		75.6			72.0		0.408
3-d 24-h		60.0			75·5		0.006		73·1			73.4		1.000
Organ meat		00 0			700		0 000		701			70 4		1 000
1-d 24-h		0.0			11.2		_		5.0			11.5		0.018
3-d 24-h		0.0			12.2		_		8.1			11.5		0.016
Eggs		0.0			12.2				0.1			11.0		0.010
_ggs 1-d 24-h		13.8			22.1		0.058		20.0			21.3		0.418
3-d 24-h		20.0			32.3		0.038		31.9			30.1		0.418
Legumes, nuts and seeds		20.0			32.3		0.027		31.9			30.1		0.009
	•	42.5			53.4		0.072		48.9			53.9		0.050
1-d 24-h 3-d 24-h		52·5			59·4		0.072		51.9			60.6		0.030
		52.5			59.4		0.103		51.9			00.0		0.032
Milk and dairy products		25.0			01.1		0.209		34.4			00.7		0.190
1-d 24-h 3-d 24-h					31.1							28.7		
		25.0			32.9		0.196		30.6			32.3		0.715
ASF		00.5			70.7		0.005		70.0			04.0		0.040
1-d 24-h		68.5			79.7		0.005		73.3			81.6		0.018
3-d 24-h		73.8			83.9		0.001		74.4			85⋅6		0.002
DDS ≤ 4														
1-d 24-h		65.0			39.8		0.001		46.9			41.9		0.060
3-d 24-h		58.8			34.1		0.001		40.6			36-6		0.051
DDS														
1-d 24-h	3.5		1.2	4.7		1.2	0.003	4.2		1.2	4.9		1.1	0.035
3-d 24-h	3⋅8		1.2	4.9		1.1	0.001	4.2		1.1	4.8		1.1	0.035
ASFS														
1-d 24-h	1.4		0.9	2.1		1.1	0.062	1.7		1.0	2.4		1.2	0.070
3-d 24-h	1⋅8		1.0	3.5		1.2	0.004	1⋅8		1.1	3.4		1.0	0.001

1-d, 1-day; 3-d, 3-day; ID, iron deficiency; DDS, dietary diversity score; ASF, animal-source foods; ASFS, animal-source foods score.

\*Continuous data reported as mean and so, categorical data presented as percentages. The t test was used to compare the mean and so values;  $\chi^2$  test was used to compare the proportions. Statistical significance was set at P < 0.05 and a trend as P < 0.1.

†Three non-consecutive days.

fruits other than vitamin A-rich' group and various ASF during the reference period were less likely to be anaemic and ID compared with non-consumers.

Comparisons between anaemic v. non-anaemic and ID v. non-ID children showed that significantly fewer anaemic and ID children consumed 'vegetables and fruits other than vitamin A-rich'; however, for ID children, this was only the case for the 3-d reference period. The low consumption of 'vegetables and fruits other than vitamin A-rich' by anaemic children corresponds to our findings on the nutrient intake of children (39), which showed that vitamin C

intake was lower in anaemic v. non-anaemic and ID v. non-ID children. Available evidence indicates that fruit and vegetable intake is associated with a reduced risk of many nutrition-related diseases in South Africa<sup>(40)</sup>. In low- and middle-income countries, low consumption of fruits and vegetables was shown to be a predictor of poor iron status, which may lead to nutritional anaemia<sup>(41,42)</sup>. A study of Brazilian children aged 4–10 years showed that children who did not consume fruits and vegetables had a higher prevalence of anaemia compared with consumers<sup>(41)</sup>. Similarly, results of a recent study conducted in adult





**Table 3** OR and 95 % CI for having anaemia and iron deficiency (ID) for 5- to 12-year-old children according to the food groups consumed and dietary diversity scores (DDS) cut-off values\*

		1-d reference perio	od	3-d reference period			
Food groups consumed	OR†	95 % CI	P	OR†	95 % CI	Р	
Anaemia							
Vegetables and fruits other than vitamin A-rich	0.67	0.40, 0.71	0.002	0.38	0.23, 0.62	0.002	
Meat, chicken and fish	0.84	0.57, 1.10	0.084	0.78	0.57, 0.96	0.045	
ASF	0.66	0.52, 0.85	0.002	0.67	0.51, 0.87	0.003	
DDS ≤ 4	1.71	1.30, 2.21	0.001	1.56	1.45, 2.67	0.006	
DDS>4	1.0			1.0			
ID							
Vegetables and fruits other than vitamin A-rich	0.83	0.79, 1.12	0.241	0.80	0.65, 0.82	0.038	
Organ meat	0.86	0.69, 1.02	0.045	0.54	0.42, 0.80	<0.001	
Legumes, nuts and seeds	0.53	0.25, 1.12	0.095	0.73	0.67, 0.93	0.020	
ASF	0.74	0.42, 1.31	0.219	0.78	0.46, 0.92	0.003	
DDS ≤ 4	1.30	0.91, 1.84	0.060	2.79	1.77, 4.48	<0.001	
DDS>4	1.0	·		1.0			

<sup>1-</sup>d, 1-day; 3-d, 3-day; ASF, animal-source foods.

Ghanaian women showed that non-consumers of fruits and vegetables had a higher chance of being anaemic<sup>(42)</sup>. The inverse relationship between the consumption of fruits and vegetables, other than vitamin A-rich, to anaemia and ID can, at least partly, be explained by the presence of nutrients that facilitate absorption of iron from the diet. The high vitamin C content in most fruits and vegetables may play an important role in enhancing the bioavailability of iron from plant-based foods, thus preventing nutritional anaemia<sup>(43)</sup>.

The South African population generally has a low intake of fruits and vegetables<sup>(6)</sup>. Low consumption has also been reported for schoolchildren<sup>(9)</sup>. To thrive, a more frequent and regular intake of a variety of fruits and vegetables should be promoted. The South African Food-Based Dietary Guidelines recommend eating 'plenty of fruit and vegetables every day' as well as various types and colours (44) in order to increase nutrient intake. Although consumption of vitamin A-rich fruits and vegetables was not associated with anaemia and ID in our study, these fruits and vegetables are rich in carotenoids, which are precursors of vitamin A, and consumption may increase the absorption of iron from cereal-based diets<sup>(14)</sup>. The number of children who reported intake of vitamin A-rich fruits and vegetables during the recall period was low (<14%). Strategies to increase intake of fruits and vegetables should thus be explored, as is recommended by the Centres for Disease Control and Prevention<sup>(45)</sup>. For example, home gardening in rural low socio-economic areas could be promoted to improve the intake of fruits and vegetables, particularly those rich in vitamin A<sup>(46)</sup>. Also, the importance of fruit and vegetable consumption could be included in the school curriculum.

Consumption of various ASF also showed significant association with anaemia in primary schoolchildren. More than  $70\,\%$  of the children in our study ate foods from the

'ASF' group, regardless of the reference period. However, significantly fewer anaemic v. non-anaemic and ID v. non-ID children consumed foods from the 'ASF' group. A number of studies in different age groups have shown a positive association between consumption of various ASF and haematological indices<sup>(47–51)</sup>, including a study in school-aged children in Kenya<sup>(48)</sup>. ASF, particularly organ meat (e.g., liver, kidney), followed by red and white meat, and fish, are rich in highly bioavailable nutrients such as iron, vitamins A and B<sub>12</sub>, which are important and effective for iron mobilisation and Hb synthesis and play an important role in managing nutritional anaemia (52-54). Moreover, foods from ASF groups are effective enhancers of non-haem iron absorption when consumed in the same meal as plant-based foods and may increase iron absorption by 2- to 3-fold<sup>(13)</sup>. The South African Food-Based Dietary Guidelines recommend that foods from the ASF group can be eaten daily, but to maintain a healthy balanced diet, consumption of a variety of foods from other food groups is recommended<sup>(44)</sup>.

Significantly fewer ID than non-ID children in our study consumed legumes during the 3-d period, and the logistic regression analysis showed an inverse association between consumption from the 'legumes' group and ID for the 3-d reference period. Adding foods from the legumes group (i.e., dry beans, split peas, lentils and soya) to the diet will increase the nutrient content of meals, and the South African Food-Based Dietary Guidelines recommend that such foods are eaten regularly (55). A study showed that adding legumes can slightly improve the iron content of plantbased diets, although the bioavailability of non-haem iron from these foods is low<sup>(56)</sup>. Legumes are generally rich in iron absorption inhibitors (i.e., phytate), which may affect the absorption of iron derived from both animal and plant sources<sup>(13)</sup>. Other studies, however, could not find an association between the intake of legumes and ID(50,57). In



<sup>\*</sup>Statistical significance was set at P < 0.05 and a trend as P < 0.1. Food groups not indicated in the table showed no statistically significant results for either reference period. †Adjusted for age, gender and study site.



meals containing legumes, non-haem iron bioavailability may be enhanced by adding sufficient amounts of foods rich in vitamin C, which has been shown to reverse the inhibitory effects of phytate<sup>(58)</sup>.

Our results indicate that estimated consumption of specific food groups may vary depending on the reference period. In dietary assessment studies, associations between less frequently consumed food groups and anaemia and ID as outcome might be missed if only a 1-d reference period is used. For instance, no association was found between intake of foods from the 'meat, chicken and fish group' and anaemia, and foods from the 'vegetables and fruits other than vitamin A-rich', 'ASF' and 'legumes' groups and ID in the 1-d reference period. However, the associations between intake of these foods and anaemia and ID, respectively, were found when the 3d reference period was used. Nevertheless, for frequently consumed foods, there was an association regardless of the reference period. When using the DDS to describe dietary variety, there was a statistically significant difference in the mean DDS between the two reference periods, although the difference in the score values was relatively small.

This study contributes to the knowledge about dietary diversity and aetiology of nutritional anaemia in South African primary school-aged children. Considering that South Africa has a National Food Fortification Programme in place, which also aims to address nutritional anaemia and ID, and that all children in this study consumed starchy staples, of which maize meal and bread are fortified, our finding strengthens the point that a diverse diet should be promoted despite or in addition to fortification efforts.

For this study, baseline data from three independent intervention studies were pooled. The study sample is therefore not representative of the study population, and the prevalence figures for anaemia and ID cannot be generalised. It should however be noted that the focus of this paper is on associations of dietary diversity with anaemia and iron status, rather than anaemia and iron status *per se*. Although a limitation of the pooled data is the different tests that were used for the biochemical indicators, there is evidence that these different methods are comparable (33,34). The data are cross-sectional, and the results therefore reflect associations, but causality cannot be established.

## Conclusion

Low dietary diversity was associated with higher odds of being anaemic and ID in South African school-aged children. Our results emphasise the importance of obtaining consumption data for multiple days when studying associations of specific foods groups with anaemia and ID, particularly for foods that are not frequently consumed by the study population. The promotion of dietary diversification, with a focus on greater consumption of vegetables, fruits and ASF, including organ meats, may have a positive effect on improving anaemia and iron status in children of primary school age.

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## Supplementary material

For supplementary material accompanying this paper visit https://doi.org/10.1017/S1368980020000543

#### References

- Vos T, Allen C, Arora M et al. (2016) Global, regional, and national incidence, prevalence, and years lived with disability for 310 diseases and injuries, 1990–2015: a systematic analysis for the Global Burden of Disease Study 2015. Lancet 388, 1545–1602.
- World Health Organization (2015) The Global Prevalence of Anaemia in 2011. Geneva: WHO.
- Zimmermann MB & Hurrell RF (2007) Nutritional iron deficiency. *Lancet* 370, 511–520.
- Ahmed T, Hossain M & Sanin KI (2012) Global burden of maternal and child undernutrition and micronutrient deficiencies. Ann Nutr Metab 61, 8–17.



- Mwaniki EW & Makokha AN (2013) Nutrition status and associated factors among children in public primary schools in Dagoretti, Nairobi, Kenya. Afr Health Sci 13, 38–46.
- Labadarios D, Steyn NP & Nel J (2011) How diverse is the diet of adult South Africans? Nutr J 10, 33–39.
- Kabahenda MK, Andress EL, Nickols SY et al. (2014) Promoting dietary diversity to improve child growth in lessresourced rural settings in Uganda. J Hum Nutr Diet 27, 143–151.
- Korkalo L, Erkkola M, Heinonen AE et al. (2017) Associations of dietary diversity scores and micronutrient status in adolescent Mozambican girls. Eur J Nutr 56, 1179–1189.
- Faber M, Laubscher R & Laurie S (2013) Availability of, access to and consumption of fruits and vegetables in a peri-urban area in KwaZulu-Natal, South Africa. *Matern Child Nutr* 9, 409–424.
- Barugahara EI, Kikafunda J & Gakenia WM (2013) Prevalence and risk factors of nutritional anaemia among female school children in Masindi District, Western Uganda. Afr J Food Agric Nutr Dev 13, 7679–7692.
- Gewa CA, Murphy SP, Weiss RE et al. (2014) Determining minimum food intake amounts for diet diversity scores to maximize associations with nutrient adequacy: an analysis of schoolchildren's diets in rural Kenya. Public Health Nutr 17, 2667–2673.
- Heath ALM & Fairweather-Tait SJ (2002) Clinical implications of changes in the modern diet: iron intake, absorption and status. Best Pract Res Clin Haematol 15, 225–241.
- Hurrell R & Egli I (2010) Iron bioavailability and dietary reference values. Am J Clin Nutr 91, 1461–1467.
- García-Casal MN (2006) Carotenoids increase iron absorption from cereal-based food in the human. Nutr Res 26, 340–344.
- Balarajan Y, Ramakrishnan U, Özaltin E et al. (2011) Anaemia in low-income and middle-income countries. Lancet 378, 2123–2135.
- Schönfeldt HC, Pretorius B & Hall N (2013) The impact of animal source food products on human nutrition and health. S Afr J Anim Sci 43, 394–412.
- Herrador Z, Sordo L, Gadisa E et al. (2014) Micronutrient deficiencies and related factors in school-aged children in Ethiopia: a cross-sectional study in Libo Kemkem and Fogera districts, Amhara Regional State. PLoS One 9, 112858.
- Acham H, Tumuhimbise GA & Kikafunda J (2013) Simple food group diversity as a proxy indicator for iron and vitamin a status of rural primary school children in Uganda. Food Nutr Sci 4, 1271.
- Gwetu TP, Chhagan MK, Martin CJ et al. (2016) Anaemia, iron deficiency and diet independently influence growth patterns of school aged children in South Africa. Acad J Ped Neonatol 1, 555565.
- Saaka M & Rauf A (2015) Role of dietary diversity in ensuring adequate haematological status during pregnancy. *Int J Med Res Health Sci* 4, 749–755.
- Ali I, Thaver A, & Khan L (2014) Assessment of dietary diversity and nutritional status of pregnant women in Islamabad, Pakistan. J Ayub Medical Coll 26, 506–509.
- Saaka M, Oladele J, Larbi A et al. (2017) Dietary diversity is not associated with haematological status of pregnant women resident in rural areas of Northern Ghana. J Nutr Metah 2017, 8497892.
- Gashu D, Stoecker BJ, Adish A et al. (2016) Ethiopian preschool children consuming a predominantly unrefined plant-based diet have low prevalence of iron-deficiency anaemia. Public Health Nutr 19, 1834–1841.
- Steyn NP, Nel JH, Nantel G et al. (2006) Food variety and dietary diversity scores in children: are they good indicators of dietary adequacy? Public Health Nutr 9, 644–650.
- Arimond M, Wiesmann D, Becquey E et al. (2011) Dietary Diversity as a Measure of the Micronutrient Adequacy of

- Women's Diets in Resource-Poor Areas: Summary of Results from Five Sites. Washington, DC: FANTA-2 Bridge, FHI 360.
- Zhao W, Yu K, Tan S et al. (2017) Dietary diversity scores: an indicator of micronutrient inadequacy instead of obesity for Chinese children. BMC Public Health 17, 440.
- Meng L, Wang Y, Li T et al. (2018) Dietary diversity and food variety in Chinese children aged 3–17 years: are they negatively associated with dietary micronutrient inadequacy? Nutrients 10, 1674.
- Kennedy G, Ballard T & Dop MC (2015) Guidelines for Measuring Household and Individual Dietary Diversity.
   Rome, Italy: Nutrition and Consumer Protection Division, Food and Agriculture Organization of the United Nations http://www.fao.org/docrep/014/i1983e/i1983e00.pdf (accessed November 2017).
- FAO (2018) Dietary Assessment: A Resource Guide to Method Selection and Application in Low Resource Settings. Rome: Food and Agriculture Organization of the United Nations.
- Baumgartner J, Smuts CM, Malan L et al. (2012) Effects of iron and n-3 fatty acid supplementation, alone and in combination, on cognition in school children: a randomized, double-blind, placebo-controlled intervention in South Africa. Am J Clin Nutr 96, 1327–1338.
- Van der Hoeven M, Faber M, Osei J et al. (2016) Effect of African leafy vegetables on the micronutrient status of mildly deficient farm-school children in South Africa: a randomized controlled study. Public Health Nutr 19, 935–945.
- Taljaard C, Covic NM, Van Graan AE et al. (2013) Effects of a multi-micronutrient-fortified beverage, with and without sugar, on growth and cognition in South African schoolchildren: a randomised, double-blind, controlled intervention. Br J Nutr 110, 2271–2284.
- Karakochuk C, Hess S, Moorthy D et al. (2019) Measurement and interpretation of hemoglobin concentration in clinical and field settings: a narrative review. Ann NY Acad Sci 1450, 126–146.
- Garcia-Casal MN, Peña-Rosas JP, Urrechaga E et al. (2018) Performance and comparability of laboratory methods for measuring ferritin concentrations in human serum or plasma: a systematic review and meta-analysis. PLoS One 13. e0196576.
- Thurnham DI, McCabe LD, Haldar S et al. (2010) Adjusting plasma ferritin concentrations to remove the effects of subclinical inflammation in the assessment of iron deficiency: a meta-analysis. Am J Clin Nutr 92, 546–555.
- World Health Organization (2011) Haemoglobin Concentrations for the Diagnosis of Anaemia and Assessment of Severity. Geneva: WHO.
- 37. World Health Organization (2011) Serum Ferritin Concentrations for the Assessment of Iron Status and Iron Deficiency in Populations. Geneva: WHO.
- 38. Langenhoven ML, Conradie PJ, Wolmarans P *et al.* (1991) *MRC Food Quantities Manual*. Parowvallei: Medical Research Council
- Visser M, Van Zyl T, Hanekom SM et al. (2019) Nutrient patterns and its relation to anaemia and iron status in 5–12-year-old children in South Africa. Nutrition 62, 194–200.
- Naudé CE (2013) "Eat plenty of vegetables and fruit every day": a food-based dietary guideline for South Africa. S Afr J Clin Nutr 26, Suppl. 2, S46–S56.
- Augusto RA, Cobayashi F & Cardoso MA (2015) Associations between low consumption of fruits and vegetables and nutritional deficiencies in Brazilian schoolchildren. *Public Health Nutr* 18, 927–935.
- 42. Ghose B & Yaya S (2018) Fruit and vegetable consumption and anaemia among adult non-pregnant women: Ghana Demographic and Health Survey. *PeerJ* **6**, e4414.
- 43. Péneau S, Dauchet L, Vergnaud AC *et al.* (2008) Relationship between iron status and dietary fruit and vegetables based





- on their vitamin C and fiber content. Am J Clin Nutr 87, 1298–1305.
- Vorster HH, Badham JB & Venter CS (2013) An introduction to the revised food-based dietary guidelines for South Africa. S Afr J Clin Nutr 26, Suppl. 3, S5–S12.
- Centers for Disease Control and Prevention (2011) Strategies to Prevent Obesity and Other Chronic Diseases: The CDC Guide to Strategies to Increase the Consumption of Fruits and Vegetables. Atlanta: US Department of Health and Human Services.
- 46. Faber M, Venter SL & Benadé AJS (2002) Increased vitamin A intake in children aged 2–5 years through targeted homegardens in a rural South African community. *Public Health Nutr* 5, 11–16.
- Wolmarans P, Dhansay MA, Mansvelt EP et al. (2003) Iron status of South African women working in a fruit-packing factory. Public Health Nutr 6, 439–445.
- Grillenberger M, Neumann C, Bwibo N et al. (2007) The potential of increased meat intake to improve iron nutrition in rural Kenyan schoolchildren. Int J Vitam Nutr Res 77, 193–198.
- 49. Obse N, Mossie A & Gobena T (2013) Magnitude and anemia and associated risk factors among pregnant women attending antenatal care in Shalla Woreda, West Arsi Zone, Oromia region, Ethiopia. Ethiop J Health Sci 23, 165–173.

- Leonard AJ, Chalmers KA, Collins CE et al. (2014) The effect of nutrition knowledge and dietary iron intake on iron status in young women. Appetite 81, 225–231.
- 51. Tahir MH, Bukhari MH, Gillani Z *et al.* (2017) The association of dietary intake and iron deficiency anaemia with the socioeconomic status in general population of Faisalabad. *Ann Punjab Med Coll* **11**, 63–67.
- Fishman SM, Christian P & West KP (2000) The role of vitamins in the prevention and control of anaemia. *Public Health Nutr* 3, 125–150.
- Murphy SP & Allen LH (2003) Nutritional importance of animal source foods. J Nutr 133, Suppl. 2, s3932–s3935.
- Pettit K, Rowley J & Brown N (2011) Iron deficiency. *Paediatr Child Health* 21, 339–343.
- Venter CS, Ochse R & Swart R (2013) "Eat dry beans, split peas, lentils and soya regularly": a food-based dietary guideline. S Afr J Clin Nutr 26, Suppl. 3, s36–s45.
- 56. Saunders AV, Craig WJ, Baines SK *et al.* (2013) Iron and vegetarian diets. *Med J Aust* **199**, s11–s16.
- Asakura K, Sasaki S, Murakami K et al. (2009) Iron intake does not significantly correlate with iron deficiency among young Japanese women: a cross-sectional study. Public Health Nutr 12, 1373–1383.
- Hallberg L, Brune M & Rossander L (1989) Iron absorption in man: ascorbic acid and dose-dependent inhibition by phytate. Am J Clin Nutr 49, 140–144.

