

## Low folic acid status and its association with anaemia in urban adolescent girls and women of childbearing age in Sri Lanka

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Folic acid deficiency is implicated in the aetiology of nutritional anaemia and adverse pregnancy outcomes for the fetus. Data on folic acid status among adolescent girls and non-pregnant, non-lactating young women are limited. We assessed folic acid status in a random sample of 552 subjects (277 adolescent girls aged 15–18.9 years and 275 women aged 19–30 years) living in Colombo, Sri Lanka. The association of low folic acid status with anaemia was evaluated. Socio-economic, food intake and anthropometric data were obtained. Hb, serum folic acid, vitamin B<sub>12</sub> and ferritin and plasma homocysteine concentrations were measured. Forty-three per cent of subjects studied had low serum folic acid concentrations (<3 ng/ml) and 47% had low Fe stores (serum ferritin <20 µg/l). Overall prevalence of anaemia was 12.9%, and 43.9% of anaemic subjects had both low folic acid status and depleted Fe stores (serum ferritin <12 µg/l). Both low folate status and depleted Fe stores were significantly associated with anaemia (odds ratio = 2.32; 95% CI 1.34, 4.01 and odds ratio = 5.98; 95% CI 3.36, 10.63, respectively). Serum folic acid concentration was associated ( $r = 0.108$ ,  $P = 0.015$ ) with folate intake as indicated by a computed folate index. Folate index was associated inversely with household size and positively with economic status and education level. In this study population low folic acid status, besides depleted Fe stores, was associated with anaemia. The high prevalence of low folic acid status observed highlights the need for nutrition education to improve intakes of folate, Fe and other micronutrients among adolescent girls and young women.

### Folic acid: Iron deficiency: Anaemia

Micronutrient deficiencies and nutritional anaemia are major problems, especially among children and women in South East Asian countries including Sri Lanka (WHO, 2000). Of the micronutrients known to contribute to nutritional anaemia, Fe and folic acid have been suggested to play major roles. As in most other developing countries with a high prevalence of anaemia, the commonest cause of nutritional anaemia in Sri Lanka is thought to be Fe deficiency (Tudawe & Wikramanayake, 2000). However, the contribution of Fe, folic acid or other micronutrients towards nutritional anaemia has not been assessed in national surveys in Sri Lanka. Folic acid is a micronutrient that has been reported to be deficient in diets particularly of low-income groups (Ballew & Sugeran, 1995; Giskes *et al.* 2002). Increased demand for folate during pregnancy resulting in low folic acid status is well established (Caudill *et al.* 1998) and is a contributing factor to maternal anaemia (Sood *et al.* 1975; Seshadri, 2001).

Despite awareness of the significance of folic acid during the periconceptional period, data on folic acid status are limited, especially among adolescent girls and women of childbearing age. A recent study among adolescent schoolgirls in a rural area in Sri Lanka indicated that 22.8% have low folic acid status (Atukorala & Lanerolle, 1998). Low folic acid status is associated with adverse pregnancy outcomes, such as neural tube defects (MRC Vitamin Study Group, 1991; Berry *et al.* 1999), other birth defects (McDonald

*et al.* 2003), low birth weight (Christian *et al.* 2003) and preterm delivery (Scholl *et al.* 1996). Low folic acid status can also lead to hyperhomocysteinaemia, an independent risk factor for CVD (Clarke *et al.* 1991). Although national periconceptional folic acid supplementation programmes have already been initiated, supplementation often commences after confirmation of pregnancy in most cases. Antenatal Fe–folate supplementation has been shown to improve micronutrient status during pregnancy but is not effective in correcting the pre-existing deficits (de Silva & Atukorala, 1996; Casanueva *et al.* 2003). Furthermore, as folic acid status before conception determines folic acid status during pregnancy (Caudill *et al.* 1997), it is essential to maintain adequate levels in adolescent girls and women of childbearing age before they become pregnant.

The present study was carried out to assess folic acid status in urban adolescent girls and women of childbearing age and the association of its deficiency with anaemia. Factors leading to folic acid deficiency were also determined.

### Methods

#### Sample size estimation

Sample size was calculated based on the prevalence of folic acid deficiency reported for a selected group of adolescent

Abbreviation: FI, folate index.

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schoolgirls (14–18 years of age) from a rural area in Sri Lanka (Atukorala & Lanerolle, 1998) assuming that the prevalence would be similar in adolescent girls and women of childbearing age. The degree of precision was set at 4% and the expected prevalence at 25%. Therefore, a minimum sample size of 600 was required (Lwanga & Lemeshow, 1991) for a final number of 451, assuming a drop-out rate of 25%.

### *Study population*

Six hundred adolescent girls and women of childbearing age were enrolled for the study. Ten out of forty-seven divisions in the Municipality of Colombo, Sri Lanka were randomly selected. The participants were selected randomly from lists of persons available with the Medical Officer of Health at each of the ten divisions. Five hundred and seventy-eight subjects participated with a response rate of 96%. Adolescent girls who had attained menarche at least 2 years prior to recruitment and women who were not pregnant in the past 2 years or not breast-feeding during the past year were included. Exclusion criteria were evidence or history of any systemic illness such as CVD, hepatic disorders, renal disease, thyroid disease, cancers, arthritis, blood disorders or diabetes mellitus.

Of a total of 578 participants, twenty-six were excluded (systemic illness,  $n = 2$ ; lack of Hb data,  $n = 24$ ) from the analysis. The final sample ( $n = 552$ ) consisted of 277 adolescent girls in the age group of 15–18.9 years and 275 women aged between 19 and 30 years. Serum folic acid and ferritin concentrations were measured in 530 and 525 subjects, respectively. Five hundred and twelve subjects had complete laboratory data including Hb, serum folic acid and ferritin concentrations. Dietary data from a food frequency questionnaire were available for 526 subjects.

### *Ethical considerations*

The Ethical Review Committee of the Faculty of Medicine, University of Colombo, Sri Lanka, approved the study. Permission was obtained from the Colombo Municipal Council and written informed consent was obtained from all participants and parents/guardians of participants under 18 years of age. Results of biochemical analyses were given to the subjects, and subjects with low Hb levels were referred to the Medical Officer of Health for further management.

### *General information and anthropometric measurements*

Information regarding socio-economic status, past medical and obstetric history (if any), intake of drugs (e.g. anti-epileptics, oral contraceptives), vitamins, minerals and other supplements was obtained using a pre-tested interviewer-administered questionnaire.

Weight and height were measured by trained personnel, using standardized scales (weight to the nearest 0.1 kg using an electronic balance and height to the nearest 1 mm using a stadiometer) and BMI was calculated. Waist circumference was measured using a measuring tape. In adolescent girls, the cut-off values for underweight and overweight were taken as BMI < 5th percentile for age and BMI  $\geq$  85th percentile for age, respectively (WHO, 1996). The WHO cut-off values for Asian adults (WHO Expert Consultation, 2004) were considered as the

cut-off values for women (BMI < 18.5 kg/m<sup>2</sup> for underweight and BMI  $\geq$  23 kg/m<sup>2</sup> for overweight). For comparison, the global cut-off (WHO, 1996) for overweight (BMI  $\geq$  25 kg/m<sup>2</sup>) was also considered for women.

### *Dietary assessment*

Intake of foods rich in folate was assessed using a pre-tested food frequency questionnaire designed for this population. Participants were asked to recall the frequency of intake of each food item during the past year, choosing between seven frequency categories ranging from 0 (never/almost never) to 6 (two or three times per day). Frequencies of intakes were recorded by trained interviewers.

### *Blood sample collection*

A venous blood sample (10 ml) was obtained between 07.30 and 11.00 hours from the cubital fossa under aseptic conditions. Aliquots of blood were transferred to sample tubes coated with EDTA for measurement of Hb and plasma homocysteine, and stored in ice. The remainder was collected in sample tubes without anticoagulant for estimation of serum folic acid/vitamin B<sub>12</sub> and ferritin and left at room temperature to clot, and then stored in ice. All samples were transported to the laboratory in ice. Serum and plasma were separated by centrifugation at 3000 rpm for 7 min within 3 h of blood collection, then aliquoted into small sample vials and stored at  $-20^{\circ}\text{C}$  until analysis.

### *Laboratory measurements*

Hb was estimated by the cyanmethaemoglobin method (Randox, Co. Antrim, UK), within 5 h of collection of blood. Folic acid and Fe status were assessed by determination of serum folic acid and serum ferritin concentrations, respectively. Serum folic acid and vitamin B<sub>12</sub> concentrations were measured by competitive protein binding assay (SimulTRAC SNB radioassay for folate and B<sub>12</sub>; MP Biomedicals, Orangeburg, New York, USA). Serum ferritin concentration was measured by an immunoradiometric assay (Coat-A-Count Ferritin IRMA; EURO/DPC Ltd, Gwynedd, UK). Plasma homocysteine concentration was measured using an enzyme immunoassay method (Abbot IMx; Abbot Diagnostics, Dundee, UK) in a randomly selected sub-sample of 253 subjects. All samples were assayed in duplicate and pooled blood/sera served as internal controls.

### *Statistical analysis*

Serum folic acid, vitamin B<sub>12</sub>, ferritin and plasma homocysteine concentrations were log-transformed and analysed. A variable termed the folate index (FI) was computed by summing up frequencies of intake of dark green leafy vegetables, other vegetables and fruits to assess their combined effect. The FI ranged from 1 to 17.

Associations were tested using the  $\chi^2$  test, Pearson correlation coefficients, Student's  $t$  test and ANOVA. As there was no difference between Hb, folic acid and ferritin concentrations among adolescent girls and women, the two groups were pooled for statistical analysis. Logistic regression analysis was used to model anaemia status. Odds ratios and their 95% CI were calculated. All tests of significance were two-sided and all analyses were performed using SPSS software version 10.01 (SPSS Inc., Chicago, IL, USA).

**Table 1.** General characteristics of the study population  
(Values are mean and standard deviation or % (*n*))

	All subjects ( <i>n</i> 551)		Adolescent girls ( <i>n</i> 276)		Women ( <i>n</i> 275)	
	Mean	SD	Mean	SD	Mean	SD
Age (years)	19.6	3.28	17.0	1.32	22.3	2.31
Total monthly income/family (Rs)†						
< 5000	34.6 (189)		41.1 (113)		28.0* (76)	
5000–10 000	47.9 (261)		46.2 (127)		49.6 (134)	
> 10 000	17.5 (96)		12.7 (35)		22.4* (61)	
Level of education						
No formal education	1.3 (7)		0.70 (2)		1.8 (5)	
Primary or secondary	70.1 (384)		70.7 (195)		68.7 (189)	
Tertiary	28.6 (160)		28.6 (79)		29.5 (81)	
BMI (kg/m <sup>2</sup> )	19.7	3.86	19.3	3.59	20.2*	4.07
Waist circumference (cm)	61.3	8.23	60.5	7.75	62.2*	8.23

Rs, Sri Lankan Rupees (£1 = Rs 189.00).

Value significantly higher than in adolescent girls: \**P*<0.05.

† Data on economic status were unavailable for one adolescent girl and four women.

## Results

### Subject profile

General characteristics of the study population are shown in Table 1. Most of the subjects were from the low socio-economic class, with 82.5% having a total family income of less than Sri Lankan Rupees 10 000 (£53) per month. Nearly 83% of subjects were financially dependent on their parents/guardians/spouses. Approximately 29% had tertiary education and only seven subjects (1.3%) had no formal school education. Among the adolescent girls aged 15–18.9 years, 41.5% (*n* 115) were school dropouts. Of the women 19–30 years of age, 26.9% (*n* 74) were married, whereas only ten adolescent girls were married. Of the married women and girls, 61.9% (fifty-one women and one girl) had given birth to at least one child.

Mean BMI and waist circumference values were higher (*P*<0.05) in women of childbearing age than in adolescent girls (Table 1).

The cut-off values for underweight (BMI <5th percentile for age) were: 15 years, 16.0 kg/m<sup>2</sup>; 16 years, 16.4 kg/m<sup>2</sup>; 17 years, 16.6 kg/m<sup>2</sup>; 18 years, 16.7 kg/m<sup>2</sup>. The cut-off values for overweight (BMI ≥85th percentile for age) were: 15 years, 24.3 kg/m<sup>2</sup>; 16 years, 24.7 kg/m<sup>2</sup>; 17 years, 25.2 kg/m<sup>2</sup>; 18 years, 25.6 kg/m<sup>2</sup>. Eighteen per cent of adolescent girls (15–18.9 years) were underweight (BMI <5th percentile for age) and 8.7% were overweight (BMI ≥85th percentile for age), while 43.3% of women were underweight (BMI <18.5 kg/m<sup>2</sup>) and 22.5% were overweight (BMI ≥23 kg/m<sup>2</sup>). When the global cut-off value for overweight for adults (BMI ≥25 kg/m<sup>2</sup>) was considered, only 11.7% of women were overweight.

### Folate intake

Frequency of intake of common folate-rich foods, as indicated by the FI, was similar (Table 2) among adolescent girls and women

**Table 2.** Folate status and selected biochemical indices of the study population\*

	All subjects ( <i>n</i> 551)		Adolescent girls ( <i>n</i> 276)		Women ( <i>n</i> 275)	
	Mean†	95% CI	Mean†	95% CI	Mean†	95% CI
Folate index						
Mean	11.6		11.7		11.5	
SD	1.96		1.94		1.98	
Serum folic acid (ng/ml)	3.66	3.46, 3.87	3.52	3.27, 3.79	3.80	3.49, 4.13
< 3 ng/ml (%)	43.6		45.1		42.0	
Plasma homocysteine (μmol/l)	11.7	11.2, 12.2	11.4	10.7, 12.2	11.9	11.3, 12.6
≥ 15 μmol/l (%)	19.0		19.3		18.7	
Hb (g/l)						
Mean	132.3		131.9		131.3	
SD	12.2		11.9		12.5	
< 120 g/l (%)	12.9		13.1		12.7	
Serum ferritin (μg/l)	19.7	18.3, 21.3	18.7	16.8, 20.9	20.8	18.7, 23.1
< 12 μg/l (%)	25.3		25.0		25.6	
12–19.99 μg/l (%)	21.3		24.7		18.0	
Vitamin B <sub>12</sub> (pg/ml)‡	706	676, 737	705	662, 750	707	667, 749

\* Folate intake is indicated by the folate index (range 1–17). Data were available for 525 subjects for ferritin and 530 subjects for folic acid. Homocysteine was measured in a sub-sample of 253 subjects. There were no significant differences between adolescent girls and women for any of the parameters considered.

† Geometric mean.

‡ Percentage with low vitamin B<sub>12</sub> (<150 pg/ml) concentration was 0.44% (*n* 2).

**Table 3.** Comparison of folic acid and iron status of anaemic (Hb < 120 g/l) and non-anaemic (Hb ≥ 120 g/l) subjects\* (Values are % and *n* or geometric mean (95% confidence interval))

	Anaemic ( <i>n</i> 66)		Non-anaemic ( <i>n</i> 446)		Significance test	<i>P</i>
	%	<i>n</i>	%	<i>n</i>		
Serum folic acid (ng/ml)	2.98 (2.50, 3.54)		3.77 (3.56, 4.00)		<i>t</i> = 2.97	0.003
Serum ferritin (μg/l)	9.48 (7.20, 12.4)		21.9 (20.3, 23.6)		<i>t</i> = 7.26	< 0.001
Serum folic acid < 3 ng/ml	62.1	41	41.3	184	χ <sup>2</sup> = 10.2	0.001
Serum ferritin < 12 μg/l	65.2	43	18.8	84	χ <sup>2</sup> = 66.1	< 0.001
Serum ferritin < 20 μg/l	74.2	49	42.2	188	χ <sup>2</sup> = 23.8	< 0.001
Serum folic acid < 3 ng/ml and serum ferritin < 12 μg/l	43.9	29	8.30	37	χ <sup>2</sup> = 65.1	< 0.001
Serum folic acid < 3 ng/ml and serum ferritin < 20 μg/l	51.5	34	17.7	79	χ <sup>2</sup> = 38.2	< 0.001
Serum folic acid < 3 ng/ml and serum ferritin ≥ 20 μg/l	10.6	7	23.5	105	χ <sup>2</sup> = 5.63	0.017
Serum folic acid ≥ 3 ng/ml and serum ferritin < 20 μg/l	22.7	15	24.4	109	χ <sup>2</sup> = 0.09	0.762

\* Subjects with complete laboratory data including Hb, serum ferritin and folic acid (*n* 512).

of childbearing age (mean 11.7 (SD 1.94) in adolescent girls *v.* mean 11.5 (SD 1.98) in women). Mean FI in the whole study group was 11.6 (SD 1.96).

*Folate, iron and anaemia status*

Mean Hb, folic acid and ferritin concentrations were similar among adolescent girls and women (Table 2). Significantly, 43.6% of the study population (45.1% of adolescent girls and 42.0% of women) had serum folic acid concentrations < 3 ng/ml. Nineteen per cent of subjects (19.3% of adolescent girls and 18.7% of women) had plasma homocysteine concentrations ≥ 15 μmol/l, the concentrations being similar in adolescent girls and women. The prevalence of anaemia (Hb < 120 g/l) in our study population was 12.9%. Depleted Fe stores (serum ferritin < 12 μg/l) were observed in 25.3% (25.0% of adolescent girls and 25.6% of women). An additional 21.3% had low Fe stores, indicated by serum ferritin concentrations between 12 and 19.99 μg/l (24.7% of adolescent girls and 18.0% of women). Serum vitamin B<sub>12</sub> concentrations were in the normal range, except for two subjects with low values (< 150 pg/ml).

Comparison of folic acid and Fe status in anaemic and non-anaemic subjects is given in Table 3. Anaemic subjects (Hb < 120 g/l) had significantly lower concentrations of serum folic acid (*P* = 0.003) and ferritin (*P* < 0.001) than non-anaemic subjects. There was no significant difference in serum vitamin B<sub>12</sub> concentration between anaemic and non-anaemic subjects (*t* = 0.566; *P* = 0.571). Of the sixty-six anaemic (Hb < 120 g/l) subjects, 62.1% (*n* 41) had low serum folic acid concentrations (< 3 ng/ml), while 41.3% of non-anaemic subjects had low serum folic acid concentrations (*P* = 0.001). Serum ferritin concentrations < 12 μg/l were observed in 65.2% of anaemic subjects (*n* 43) and 18.8% of non-anaemic subjects (*P* < 0.001). Forty-four per cent of anaemic subjects (*n* 29) had both depleted Fe stores (serum ferritin < 12 μg/l) and low serum folic acid (< 3 ng/ml) status, compared with 8.3% of non-anaemic subjects (*P* < 0.001).

Results of logistic regression analyses using anaemia status as the dependent variable are summarized in Table 4. Low folate status (serum folic acid concentration < 3 ng/ml) and depleted Fe stores (serum ferritin concentration < 12 μg/l) were significant predictors of anaemia after adjusting for each other. Subjects with depleted Fe stores were almost six times

**Table 4.** Logistic regression analysis using anaemia status\* as the dependent variable

Independent variable	Regression coefficient	<i>P</i>	Odds ratio (95% CI)
Intercept	-2.29		
Low folate status†	0.842	0.003	2.32 (1.34, 4.01)
Depleted Fe‡	1.79	0.000	5.98 (3.36, 10.63)
Low Fe§	-0.365	0.444	0.694 (0.27, 1.77)

\* Anaemic, Hb < 120 g/l; non-anaemic, Hb ≥ 120 g/l.

† Serum folic acid concentration < 3 ng/ml.

‡ Serum ferritin concentration < 12 μg/l.

§ Serum ferritin concentration < 20 μg/l.

more likely to be anaemic (95% CI 3.36, 10.63) than Fe-replete subjects (serum ferritin concentration ≥ 20 μg/l). Subjects with low folic acid (< 3 ng/ml) status were approximately 2.3 times more likely to be anaemic than subjects with serum folic acid concentrations ≥ 3 ng/ml (95% CI 1.34, 4.01). Subjects with both low Fe and low folate status were not at a significantly greater risk of being anaemic compared with subjects with low status on either parameter, *i.e.* non-significant interaction term.

Serum folic acid concentration was positively correlated with FI (*r* = 0.108; *P* = 0.015). FI was significantly lower (*t* = 3.70; *P* < 0.001) in subjects with low serum folic acid concentrations (< 3 ng/ml) than in subjects with serum folic acid concentrations ≥ 3 ng/ml. FI was significantly associated with level of education (*F* = 4.998; *P* = 0.007) and total monthly income (*F* = 6.151; *P* = 0.002). Mean FI was higher (*t* = 2.057; *P* = 0.041) among subjects having fewer household members (four or less) as compared with those having more (fifteen or more) household members. There were significant differences in mean serum folic acid concentration among different income groups (*F* = 3.77; *P* = 0.011). A significantly lower (*t* = 5.012; *P* < 0.001) mean serum folic acid concentration was noted among parous women than nulliparous women (2.77 *v.* 5.76 ng/ml). Plasma homocysteine concentrations were negatively correlated (*r* = -0.157; *P* = 0.014) with serum folic acid concentrations. Further, 47.9% of subjects with moderately elevated plasma homocysteine concentration (≥ 15 μmol/l) had a low serum folic acid (< 3 ng/ml) concentration.

## Discussion

The present study was an attempt to assess the magnitude of the problem of folic acid deficiency and its importance in the aetiology of anaemia among urban adolescent girls and non-pregnant, non-lactating women of childbearing age. A serum folic acid concentration  $< 3$  ng/ml reflects a low folate status (Herbert, 1987). The percentage of subjects with low folate status (43.6%) was higher in our study sample than that reported in a previous study (22.8%) for rural adolescent schoolgirls (Atukorala & Lanerolle, 1998). This difference could be attributed to the higher accessibility of foods rich in folate in rural areas compared with urban areas. In a recent study on a rural community in India, serum folic acid concentrations  $< 3$  ng/ml were reported in 27.7% of non-pregnant young women (Pathak *et al.* 2004a).

Inadequate dietary intake of folate-rich foods is considered a major cause of folic acid deficiency (Krumdieck, 1991; Tucker *et al.* 1996; Herbert, 1999). In the present study, intake of dark green leafy vegetables, other vegetables and fruits, expressed as FI, was associated with a higher folate status. The significant positive association observed with folate intake and monthly income is in agreement with the report by Pathak *et al.* (2004b). The intake of folate-rich foods was lower when family size was large and higher among subjects with a higher level of education. Rahmann (1994) has suggested that women's level of education is likely to play a major role in food selection.

The prevalence of anaemia observed in the present study was lower than in previous studies (Lanerolle & Atukorala, 1998; Medical Research Institute, Sri Lanka, 2003). This could be due to differences in the age groups studied. Serum ferritin concentration  $< 12$   $\mu$ g/l indicates depleted Fe stores (Beard, 1994), while concentrations between 12 and 19.9  $\mu$ g/l reflect low Fe stores (Bothwell *et al.* 1979). By these criteria, nearly 50% of our population had either low or depleted Fe stores, indicating that subclinical Fe deficiency is a significant problem in this community. A high prevalence of subclinical Fe deficiency has also been reported by Lanerolle & Atukorala (1998).

Previous studies have not assessed the role of folic acid in the aetiology of nutritional anaemia in Sri Lanka. The national survey used the prevalence of anaemia as a proxy for Fe deficiency (Medical Research Institute, Sri Lanka, 2003). It is important to note that this is true only in settings where Fe deficiency is known to be the major cause of anaemia (WHO/UNICEF, 2004). Our findings indicate that both folic acid and Fe deficiency are important in the aetiology of nutritional anaemia in the present population, with Fe appearing to play the leading role. Data on the aetiology of anaemia in other South Asian countries is limited. Ahmed (2000) suggested that Fe deficiency could be the major factor in the aetiology of anaemia in the Bangladeshi population. Dietary intakes among women indicate a similar situation in India (Pathak *et al.* 2004a). In contrast, Ronnenberg *et al.* (2000) observed a higher contribution of B-vitamin deficiency (folic acid, vitamin B<sub>12</sub> and B<sub>6</sub>) than of Fe deficiency in the aetiology of anaemia among non-pregnant female textile workers of childbearing age in China. In the present study population, low serum vitamin B<sub>12</sub> concentrations were noted in  $< 1$ % of subjects, and is unlikely to be important in the aetiology of anaemia.

The high prevalence of low folic acid status observed in the present population highlights the importance of establishing folate status on a national scale. There are no national-level studies in the South Asian region. Moreover, the negative effect

of childbearing on folate status noted in the present study and by Pathak *et al.* (2004b) should be considered when developing strategies to improve women's nutritional status. Improved folic acid status will not only prevent anaemia, but is likely to prevent neural tube defects, other birth defects and pregnancy complications. However, the amount of folic acid needed to avert adverse pregnancy outcomes is beyond the threshold for deficiency (Scott, 2001) and there can be a graded risk reduction between improved status and risk of neural tube defects even when the initial folate status is normal (Wald *et al.* 2001). The observation of an inverse association between low folic acid status and moderately elevated plasma homocysteine concentrations suggests that low folate status could be a predisposing factor for hyperhomocysteinaemia in this population.

In conclusion, low folic acid status and low Fe stores were common among adolescent girls and women of childbearing age in an urban area in Sri Lanka. Depleted Fe stores are likely to be a major factor in the aetiology of nutritional anaemia in this population. However, low folic acid status also plays a significant role in the aetiology of anaemia. Our analysis showed that frequent intake of common folate-rich foods was associated with a better folic acid status. The folate-rich foods frequently consumed by this population are also common sources of Fe and other micronutrients. Nutrition education strategies are required to increase the awareness of the benefits of preventing folic acid deficiency among adolescent girls and women of childbearing age and also to help in making correct food choices.

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