Original Article

Anemia in Cambodia: a cross-sectional study of anemia, socioeconomic status and other associated risk factors in rural women

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Iron deficiency and associated anemia are severe public health problems, which are prevalent in the developing world. We conducted a cross-sectional survey, comprised of written interview questions and laboratory analysis of blood biomarkers, in Kandal Province, Cambodia. The objective of this study is to examine possible factors that are associated with anemia in rural Cambodia. Data on socioeconomic status, water source/treatment practices, and meat consumption was also collected. Of the 297 women surveyed, 51.2% were anemic. Of those women found to be anemic, iron deficiency was implicated in 9.7% of cases (SF <15 ng/L), with an additional 18.5% reported to be borderline iron deficient (serum ferritin=15-30 ng/L). Meat consumption was very low, with nearly one-half of the women consuming meat one time per month or less. This study highlights the multi-faceted etiology of anemia in Cambodia and emphasizes the need for comprehensive nutrition surveying in order to better inform prevention and treatment programming and policy development.

Key Words: anemia, socioeconomic status, meat consumption, Cambodia, risk factor

INTRODUCTION

Despite the efforts of governments, multilateral agencies and nongovernmental organizations worldwide, anemia remains a major public health problem. An estimated 3.6 billion people are iron deficient and of these more than two billion are anemic, especially women of reproductive age, infants and young children.¹

Iron status flows from iron deficiency anemia, to iron deficiency in the absence of anemia, to normal iron status, and finally to iron toxicity.² Globally, the most significant contributor to the onset of anemia is iron deficiency, which is estimated to be responsible for approximately 50% of all cases.²

The most recent Cambodian Demographic and Health Survey (CDHS) reported a country-wide prevalence of anemia of 44.4% in women of reproductive age and 55.1% in children aged 6-59 months.³ To date, the prevalence of iron deficiency is unknown and there is a dearth of information describing the nutrition situation and associated factors in Cambodia. Baseline data of this sort is needed in order to determine the extent of the public health problem and is essential for the development of effective and targeted treatment and prevention programmes.

The current paper describes a cross-sectional study that was conducted to investigate the prevalence of iron deficiency and associated anemia in rural Cambodian women aged 16 years and older prior to initiation of an iron intervention. Particular emphasis has been placed on understanding the role that socioeconomic status has on the prevalence of anemia and the degree of meat consumption in the study population.

MATERIALS AND METHODS Study site and subjects

This cross-sectional study was conducted in Tuol Trea and Preak Khmeng villages in Preak Khmeng Commune, Lvea Aem District, Kandal Province, Cambodia. Approximately 2,313 people live within the study area of Preak Khmeng Commune, with 1,563 in Preak Khmeng Village and 750 in Tuol Trea Village. For the purposes of the current study, Preak Khmeng village was considered as two distinct study sites, Preak Khmeng 1 (PK1) and Preak Khmeng 2 (PK2). These two areas within Preak Khmeng are separated geographically by a river system, have distinct village councils, experience different degrees of flooding and drought, and for all intents and purposes can be examined as two distinct locales.

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Households were selected using a systematic random sample by approaching every fourth household until the pre-determined sample size was met during a 2-week period in June 2010, the beginning of the monsoon season. The project was explained briefly and those women who consented to participate were then surveyed and a blood sample was collected for biomarker measurement. Women were included if they lived in the study area and did not intend to move in the following 12-months and were 16 years of age and older; exclusion criteria included unwillingness to give a blood sample, participants who had received a blood transfusion in the last 5 years, regular use of iron supplements, and/or pregnant women (for detailed analysis). Sample size was calculated to produce a power of 95% (α =0.05) with an allowable error of 5%; the calculation was performed to detect a difference in hemoglobin concentration following a dietary intervention of 10 g/L (not described herein). A total of 310 women first completed a structured, coded survey, after which a blood sample was taken to establish anemia status and the size of iron stores.

Survey procedures

Trained village health personnel from a local nongovernmental organization, Resource Development International Cambodia (RDI), were employed to conduct the cross-sectional survey and to collect the biomarker data. Data were collected on the size and demographics of the household, as well as the participant's pregnancy status and parity history, history of anemia and infectious disease, education level attained, literacy, common water, and household possessions. Frequency of the consumption of meat and meat products (including poultry, pork, beef, goat, buffalo, and rodent) was assessed through a series of open-ended questions. A detailed socioeconomic analysis was also performed and was based on information collected on household possessions, housing quality and livestock possession. The socioeconomic asset score was derived using the approach of Morris and colleagues.4

Laboratory methods

A 3 mL sample of blood was collected following venipuncture (Greiner Bio-One, Shanghai, China) for each participant. A portable hemoglobin analyzer (HemoCue AB, Angelholm, Sweden) was used on-site to assess hemoglobin level. The remaining blood sample was transported on ice within a 6 hour time period to Phnom Penh for analysis at Paramed Laboratory (Paramed Laboratoire D'Analyses Medicales, Phnom Penh, Cambodia). Serum ferritin (SF) was measured using an Abbott AxSYM system (Abbott Laboratories, North Chicago, IL, USA) and serum C-reactive protein (CRP) was assessed by an ELI-SA (AssyMax Human CRP ELISA Kit, St. Charles, USA); appropriate controls and reference standards were used to ensure accuracy of the laboratory analysis.

Serum ferritin was the biomarker chosen to assess iron stores because it most closely correlates with relative total body iron stores.² Serum ferritin is an acute-phase reactant protein, which is raised in response to infection or inflammation. A CRP concentration >10 mg/L indicated

infection/inflammation.⁵ Anemia was indicated with hemoglobin concentration <120 g/L, and iron deficiency with a SF level <15 ng/mL, according to World Health Organization (WHO) recommendations for non-pregnant women greater than 15 years of age.² Anemia confounded by co-infection was identified in women who had abnormal hemoglobin levels (Hb <120 g/L) together with abnormal CRP concentration (CRP >10 mg/L), and these women (n=13) were excluded from detailed analysis for both Hb and SF.⁶ The overall socioeconomic status scores were also broken down into quartiles for detailed analysis.

Statistical analysis

Data were initially recorded by hand onto coded spreadsheets and later transcribed into SPSS Version 18.0 (SPPS Inc., Chicago, IL, USA). Hemoglobin, SF and socioeconomic score were evaluated to determine whether they were normally distributed using normality plots and the Shapiro-Wilk test for normality of data. Serum ferritin and asset scores values were right-skewed, and were therefore log-transformed.

Descriptive statistics were used to examine baseline demographic information. Baseline demographics, Hb and log-transformed serum ferritin were compared among villages using linear regression for continuous variables and chi-square tests for binary variables. Additional descriptive analysis was used to examine biomarker data and meat consumption. The geometric mean was calculated for SF and asset score data, and the median meat consumption was determined. The baseline demographic data was compared among the three study sites to determine if village should be included as a fixed effect.

Serum ferritin values were further classified into borderline (15-30 ng/mL), low (<30 ng/mL), and very low (<15 ng/mL) in order to capture those women who are sub-clinically iron deficient. Meat consumption was ranked according to high (>9 servings per month), medium (2-9 servings per month), and low (<2 servings per month) intake. These values, together with socioeconomic status and a self-reported history of infectious disease (malaria and hookworm) were compared between women found to be anemic versus those who were non-anemic at baseline using univariate logistic regression incorporating the clustering effect of village.

Multiple regression analysis was used to evaluate the associations between either hemoglobin or log-transformed SF and meat consumption, the demographic variables of age, education level attained, number of pregnancies, socioeconomic status and menopausal status after controlling for village as a fixed effect. Examining the standardized residuals and ensuring homogeneity of variances tested the assumptions of the final models.

RESULTS

The demographic data for the 310 women included in the study are presented in Table 1. Of the 310 women entered into the study, 13 were omitted from detailed analysis: six were pregnant, while seven had both abnormal CRP (>10 mg/L) and abnormal Hb concentrations, signalling anemia of chronic disease.⁶

The baseline survey was used to collect information on

Category	Variable	PK1	PK2	Tuol Trea	Combined
N		91	119	100	310
Participant	Age^{\dagger}	40.0 (14.2)	41.1 (15.8)	44.8 (13.8)	42.0 (14.8)
-	Height [†] (cm)	154 (5.1)	153 (7.1)	151 (7.7)	153 (6.9)
	Weight [†] (kg)	56 (51.4)	49 (7.7)	49 (8.9)	51 (28.8)
Anemia risk factors	Mean parity [†]	5.2 (3.9)	4.4 (3.3)	4.8 (3.5)	4.8 (3.6)
	Pre-menopausal (%)	69 (75.8)	80 (67.2)	52 (52.0)	201 (64.8)
	Diagnosed anemia (%)	7 (7.7)	3 (2.5)	2 (2)	12 (3.9)
	Diagnosed hookworm (%)	13 (14.3)	38 (31.9)	31 (31)	82 (26.5)
	Latrine use (%)	0 (0.0)	1 (0.003)	2 (0.007)	3 (1.0)
	Diagnosed malaria (%)	2 (2.2)	7 (6.3)	7 (7)	16 (5.2)
Education	None (%)	38 (41.8)	28 (23.5)	37 (37)	103 (33.2)
	Grade 6-9 (%)	9 (9.9)	14 (11.8)	21 (21)	44 (14.2)
	$> 9^{\text{th}}$ grade (%)	5 (5.5)	7 (5.9)	3 (3)	15 (4.8)
Literacy	None (%)	44 (45.0)	39 (32.8)	42 (42.0)	125 (40.3)
	Low (%)	25 (27.5)	58 (48.7)	34 (34.0)	117 (37.7)
	High (%)	22 (24.2)	22 (18.5)	24 (24.0)	68 (21.9)

Table 1. Demographic data for women in two villages of Lvea Aem District, Kandal Province, Cambodia

[†]Mean (Standard Deviation).

PK1: Preak Khmeng Village - section 1; PK2: Preak Khmeng Village - section 2.

Table 2. Hemoglobin, iron indices, and meat consumption for 297 women surveyed in Lvea Aem District, Kandal Province, Cambodia

		PK1	PK2	Tuol Trea	Combined
Ν		82	115	100	297
Hemoglobin (g/L)	Mean (SD)	124 (14.0)	115.2 (14.8)	117.9 (13.7)	118.4 (14.6)
Proportion anemic (%)	<12 g/L (anemic)	39.0 (49.0)	59.1 (49.4)	52.0 (50.2)	51.2 (50.1)
• · · ·	<10 g/L (moderate)	1.2 (11.0)	12.2 (32.8)	5.0 (21.9)	6.7 (25.1)
	<7 g/L (severe)	0	0.9 (9.3)	0	0.3 (5.8)
Ferritin (ng/mL)	Mean of raw data (SD)	76.7 (107.6)	57.0 (39.1)	62.1 (54.2)	64.2 (69.3)
	Geometric mean for log data (SD)	51.3 (2.3)	41.7 (2.4)	44.7 (2.4)	45.7 (2.4)
	<15 ng/mL (%)	9.7 (29.9)	14.8 (35.6)	11.0 (31.4)	12.1 (32.7)
	<30 ng/mL (%)	20.7 (40.8)	33.0 (47.2)	31.0 (46.4)	29.0 (45.4)
	15-30 ng/mL (%)	11.0 (31.5)	18.3 (38.8)	20.0 (40.2)	16.8 (37.5)
Iron deficiency anemia	Anemic women with low ferritin (<15 ng/mL) (%)	6.1 (24.1)	12.2 (32.8)	10.0 (30.2)	9.8 (29.7)
	Anemic with borderline-low ferritin (<30 ng/mL) (%)	12.2 (32.9)	22.6 (42.0)	19.0 (39.4)	18.5 (38.9)
Infection	Mean CRP concentration (SD)	5.4 (5.6)	2.8 (3.0)	2.7 (1.5)	3.5 (3.7)
Meat consumption	Mean meat serves per month (SD)	4.9 (4.2)	5.1 (4.9)	5.3 (6.0)	5.1 (5.1)
I	Median (range)	2 (0-20)	2 (0-20)	2 (0-20)	2 (0-20)

PK1: Preak Khmeng Village - section 1; PK2: Preak Khmeng Village - section 2.

food, water and hygiene practices. During the dry season (December to June), the river system was the primary source of water for residents in PK1 (95.6%) and PK2 (87.4%), while ground water collected from a tube well was most often used in Tuol Trea (81.0%). During the monsoon season (July to November), however, the majority of villagers in all three study sites relied heavily upon stored rain water (72.9%), which is typically collected in large, concrete tanks. Water treatment was achieved through boiling drinking water (85.8%), with a small subset reliant upon chemical treatment or the use of water filters. Few women regularly used latrines 0.97% (3/310). Approximately one-third of the participants consumed meat at least once a week, while almost half (45.8%) reported to consume meat only on special occasions (i.e., festivals, weddings, funerals).

Detailed analysis was performed on a truncated data set (n=297), not including pregnant women or women with anemia of chronic disease (Table 2). The prevalence of anemia, as defined by hemoglobin concentration <120

g/L, was 51.2% (152/297). The prevalence of iron deficiency, defined by SF concentration <15 ng/mL was 12.1% (36/297); an additional 16.8% (50/297) were found to be borderline iron deficient (15-30 ng/mL). The prevalence of iron deficiency anemia, that is both abnormal hemoglobin and SF levels, was observed in 9.8% (29/297) of the participants. Borderline iron deficiency anemia, taking into account those individuals with both abnormal hemoglobin and decreased iron stores (<30 ng/mL) included an additional 18.5% (55/297) of the women surveyed.

A comparison of iron status, animal-source protein consumption, socioeconomic status and infectious disease between anemic and non-anemic populations is shown in Table 3. The mean indices of iron status in the anemic group were different from those in the non-anemic group. Participants who were anemic (n=152) were 4.5 times more likely to have very low (ferritin <15 ng/mL) iron stores (OR=4.5, 95% CI: 1.9-10.8, p=0.001) when compared to the non-anemic study population. Women with

Variable	Proportion	Proportion	<i>p</i> -value	Odds Ratio	95% CI of Odds
	Non-Anemic	Anemic			Ratio
Ν	145	152			
Ferritin					
Percent ferritin very low (<15 ng/mL)	4.8% (7)	19.1% (29)	0.001	4.5*	1.9-10.8
Percent ferritin low (<30 ng/mL)	21.4% (31)	36.2% (55)	0.01	2.0^{*}	1.2-3.3
Percent ferritin borderline (15 to <30 ng/mL)	16.6% (24)	17.1% (26)	0.9	1.0	0.5-1.8
Socioeconomic status					
Very low (Q1)	30.3% (44)	19.7% (30)	0.3	0.7	0.4-1.3
Low (Q2)	24.1% (35)	25.7% (39)	0.9	1.0	0.6-1.8
Medium (Q3)	22.8% (33)	27.6% (42)	0.7	1.1	0.7-1.9
High (Q4)	22.8% (33)	27.0% (41)	0.7	1.1	0.6-1.9
Meat consumption					
<2 servings per month	5.5% (8)	19.5% (16)	0.2	1.9	0.7-4.5
2 to 9 servings per month	69.7% (101)	55.3% (84)	0.02	0.6^{*}	0.3-0.9
>9 servings per month	24.8% (36)	34.2% (52)	0.09	1.6	0.9-2.6
Infectious disease					
Diagnosed with malaria	5.5% (8)	5.3% (8)	0.8	0.9	0.3-2.4
Diagnosed with hookworm	31.0% (45)	21.7% (33)	0.02	0.5^{*}	0.3-0.9

Table 3. Proportion of iron deficiency, socioeconomic status, low meat consumption, prior history of malaria and hookworm by anemia classification (Hb <120 g/L) and the association with being anemic in women from 3 villages in rural Cambodia

For this analysis, socioeconomic status was treated as categorical, determined by the quartiles. Referent group is non-anemic. *Significant p < 0.05

Table 4. Multiple regression analysis of risk factors (demographic, socioeconomic status, and meat consumption) associated with hemoglobin and serum ferritin concentrations in women from 3 villages residing in rural Cambodia

	Hemoglobin		Serum ferritin [†]		
	Coefficient (SE)	р	Coefficient (SE)	р	
Age (years)	-0.42 (0.095)*	< 0.001	$0.98(1.0)^{*}$	0.004	
Education (years)	1.38 (1.03)	0.18	1.0 (1.07)	0.92	
Number of pregnancies (N)	0.34 (0.28)	0.22	$1.04(1.02)^*$	0.033	
Socioeconomic status	-0.003 (0.016)	0.874	1.0 (1.0)*	0.025	
Pre-menopausal	-6.09 (2.49)*	0.015	$0.49(1.17)^*$	< 0.001	
Less than 2 meat servings per month [‡]	1.95 (3.17)	0.54	1.15 (1.22)	0.49	
2-9 meat servings per month [‡]	4.33 (1.78)*	0.016	1.09 (1.12)	0.45	

[†]Log-transformed serum ferritin used for analysis (back-transformed values shown).

[‡]Referent group: high meat consumption (9+ servings per month).

*Significant *p*<0.05

moderately low serum ferritin levels (<30 ng/mL) may be at a higher risk of developing iron deficiency and/or iron deficiency anemia than the general study population given that iron status can change rapidly, therefore this group was analyzed separately. Approximately 36% of anemic participants, compared with 21.4% of non-anemic participants were found to have low SF levels (<30 ng/mL) (OR=2.0, 95% CI: 1.2-3.3, p=0.01).

The proportion of participants consuming meat between two and nine times per month was greater among the anemic population (OR=0.6, 95% CI: 0.3-0.9, p=0.02) than the non-anemic population. Low meat consumption (less than the median of two times per month) and high meat consumption (more than nine servings per month) did not differ by anemia status (p>0.05).

The results of multiple regression analyses of various risk factors on hemoglobin and SF concentrations are given in Table 4. There was a negative association between the demographic variables age and menstrual status (pre-menopausal) with hemoglobin and iron stores (p-values<0.05). As women aged by five years, their Hb

level was predicted to decrease by 2.1 g/L, while serum ferritin increased by 4.9 ng/mL. Post-menopausal women had an Hb level that was predicted to be 6.1 g/L lower, and serum ferritin level that was 0.48 ng/mL higher than pre-menopausal women. Additionally, increasing socio-economic status (p=0.03) and total number of pregnancies (p=0.03) were associated with higher SF concentrations. Although low meat consumption was not associated with high meat consumption (>9 servings per month), women consuming meat 2-9 times per month had higher Hb than those consuming meat >9 times per month (p=0.016).

DISCUSSION

To our knowledge, the current study is the first to describe the prevalence of anemia, iron deficiency and associated risk factors in Preak Khmeng Commune, Lvea Aem District in Kandal Province, Cambodia. We found the prevalence of anemia, iron deficiency, and iron deficiency anemia to be 51.2%, 12.1%, and 9.7%, respectively. Anemia in this study site, in keeping with the overall national prevalence, is therefore considered to be a severe public health problem according to WHO criteria.⁷ Because nutritional anemia is the result of severe iron deficiency, these findings highlight the multi-faceted nature of anemia and suggest that some other etiology(ies) may be responsible for the high prevalence of anemia without iron deficiency in the study area. Helminth infection, inherited hemoglobin disorders, malaria, and other micronutrient deficiencies are well known to exist in rural Cambodia and may be responsible for some of the anemia observed in the study area.⁸ While simple iron supplementation programs might work elsewhere, the multifaceted nature of anemia in Cambodia highlights the importance of an interdisciplinary approach to the problem.

Abnormally low SF concentrations were associated with women of low socioeconomic status and those who had a low number of pregnancies. It is not clear why increasing number of pregnancies is associated with increasing SF concentration, but it may be that pregnant women had access to iron supplements and were therefore able to improve overall iron status. The observation that decreasing socioeconomic status was associated with lower SF concentration suggests that poorer women might be unable to afford a diet rich in iron. In addition, a poor understanding of nutrition has been shown to result in the failure to consume nutritious, iron-rich foods.^{9,10} In fact, we found that women belonging to the higher socioeconomic group were just as likely to be anemic as those in lower status groups. This finding would suggest that women who have more money do not choose to spend it on improving the household diet by purchasing iron-rich foods. This observation again implicates poor nutritional knowledge as a reason for the severe problem of anemia throughout the country.

Consumption of iron-rich foods was also investigated. Meat, including chicken, pork, beef, water buffalo, goat, several rodent species, and associated organ meat was not found to be a main constituent of the diet. The reliability of this data is likely quite high given that the majority of participants never purchase meat for household use, instead consuming meat only when it is offered on special occasions (i.e., weddings, funeral ceremonies, festivals, etc.) In fact, nearly one half of the participants reported meat consumption on special occasions only, and the median meat consumption was two servings per month. We found that those participants who consumed meat between two and nine times per month were less likely to be anemic than those who consumed meat less frequently. It is unclear why those women consuming meat >9 times per month were not protected against anemia and/or iron deficiency but this finding in an observational study may indicate a cause of anemia in these women other than iron deficiency. In contrast, the 2005 CDHS revealed that 98.4% of women aged 15-49 consumed meat, fish, and/or eggs on the day before the survey.¹¹ These findings would suggest that fish and eggs, which have lower iron content,¹² are much more commonly consumed than meat and poultry, though we did not investigate this directly. Previous research in the area has shown that per capita consumption of fish, including both fresh and processed, is 71.1 kg per annum, placing Cambodia among the top fish-consumption countries in the world.¹³

Poor water and sanitation practices were observed. Latrine use was reported for <1% of the participants, who instead used fields for urination and defecation. Regular use of heavily polluted river water during the wet and dry seasons was also observed. Preak Khmeng village, in particular, is situated along a river that is used for urination, defecation, clothes cleaning, washing of cattle and other livestock, along with fish-rearing, thus the use of this untreated water for drinking and cooking highlights the possibility for infection with various bacteria and parasites, including those that are known to cause anemia. These findings underscore the need for an integrated approach to anemia control in the area, with targeted interventions to improve water and sanitation practices.

The current study confirmed the severe public health problem of anemia in women in Cambodia. Poor diet, including limited meat consumption, together with infection are contributing factors to anemia and must be considered in programming efforts to ameliorate iron deficiency and iron deficiency anemia in this region. Current programs in Cambodia to improve nutrition rely on the use of plant-based iron stores, however this study highlights the need to improve intake of meat and other heme iron sources, particularly as animal-source iron is more readily bioavailable and therefore more likely to have an impact on nutrition status. The National Nutrition Program has focused thus far on the use of iron supplements and micronutrient powders as a way to improve iron status of populations, however the current study indicates that more effort should be placed on eliminating other risk factors including hookworm infection, proper sanitation, and hygiene. Additional research on the etiology of anemia in the area would benefit the development of health intervention strategies.

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AUTHOR DISCLOSURES

No competing interests are reported.

ETHICAL APPROVAL

Informed verbal consent was obtained from all participants and study procedures were approved by the Research Ethics Board at the University of Guelph (which conforms to the ethical standards of the Helsinki Declaration of the World Medical Association) and the Ministry of Rural Development, Kingdom of Cambodia.

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Original Article

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柬埔寨农村妇女中贫血与社会经济状况和其他相关危险 因素的横断面研究

缺铁性贫血是发展中国家一个严重的公共卫生问题。我们在柬埔寨干丹省做了 一个横断面调查,有书面问卷和血样分析。本研究的目标是检测柬埔寨农村与 贫血相关的潜在因素。同时也收集了社会经济状况、水来源/处理措施和肉类消 费数据。接受调查的 297 名妇女中,51.2%患有贫血。这些贫血的妇女中,9.7% 缺铁(血清铁蛋白<15 ng/L),还有 18.5%是临界缺铁(血清铁蛋白=15-30 ng/L)。肉类消费很少,将近一半的妇女每月仅摄入一次或少于一次的肉类。 本研究重点在于对柬埔寨贫血的多元病因分析,强调了营养普查的必要性,以 便更好地发展防治规划和政策。

关键词:贫血、社会经济状、肉类消、柬埔寨、危险因素