

Vitamin A status of pre-school-age children aged 6 to 59 months in the National Capital District, Papua New Guinea

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SUMMARY

Assessing the vitamin A status among pre-school-age children is essential for evaluating the magnitude and public health status of vitamin A deficiency in a population. This cross-sectional study assessed the vitamin A status of children aged 6 to 59 months resident in the National Capital District (NCD), Papua New Guinea. Children attending the Children's Outpatient Clinic at Port Moresby General Hospital participated in this study. Informed consent was obtained from parents before using blood samples from their children. Assay of plasma retinol was carried out using the 'Clin-Rep' complete kit for assay of vitamins A and E in plasma by high performance liquid chromatography (HPLC). A commercial enzyme immunoassay kit was used to assay C-reactive protein (CRP) in plasma. Of the 132 children in the study 108 (82%) had received vitamin A capsules. The median plasma retinol concentration of the 132 children was 0.98 $\mu\text{mol/l}$ and the interquartile range 0.65-1.38 $\mu\text{mol/l}$. Of the 132 children, 35 (27%) had a plasma retinol concentration below 0.70 $\mu\text{mol/l}$. 75 children (57%) had normal plasma CRP levels and in 57 (43%) the CRP levels were elevated. The median plasma retinol concentration of the children with normal plasma CRP was 1.19 $\mu\text{mol/l}$ and the interquartile range 0.93-1.50 $\mu\text{mol/l}$. The prevalence of vitamin A deficiency (VAD) in the children with normal plasma CRP was 11%, indicating a moderate public health problem. 74 (56%) males and 58 (44%) females were included in the study. The prevalence of VAD in the male and female children with normal plasma CRP was 14% and 8%, respectively, indicating a moderate public health problem among the male children and a mild public health problem among the female children. The prevalence of subclinical (mild to moderate) and marginal VAD among the children with and without elevated CRP strongly suggests the need for continuous monitoring of the vitamin A status of the vulnerable groups in NCD.

Introduction

Vitamin A (retinol) is required for regulation of the visual cycle and haematopoiesis, for differentiation and maintenance of the biological integrity of epithelial tissues, for mucin production, and for regulation of cell-mediated immunity and humoral antibody responses (1-4). The consequences of vitamin A deficiency (VAD) are manifold. VAD

is among the 'top 10' risk factors contributing to the global burden of disease among pre-school-age children in resource-limited countries (1-6). According to recent World Health Organization (WHO) estimates, about 0.2 million deaths among pre-school-age children were due to VAD, with most occurring in children with subclinical (mild to moderate) VAD (1,2). Thus the need for continuous monitoring of this vulnerable group in the

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community cannot be overemphasized.

The vitamin A status of individuals in target populations can be assessed by quantification of their plasma retinol concentrations, using high performance liquid chromatography (HPLC) (7-10). However, the carrier protein for plasma retinol, ie, retinol-binding protein, is one of the acute phase reactants that decrease during inflammation, infection and trauma (3-6). As a consequence, plasma retinol concentrations may not accurately reflect the vitamin A status of populations with high prevalence of clinical or subclinical inflammation (3-6). One of the recommended procedures for improving the accuracy of interpreting plasma retinol data is to measure the plasma concentration of C-reactive protein (CRP), which is an indicator of both inflammation and infection (2-6,11). The plasma CRP results can then be used to stratify the plasma retinol data according to the health status of individuals in the target population (3-5).

According to the recommended cut-off points, plasma retinol concentrations below 0.70 $\mu\text{mol/l}$ indicate VAD and concentrations between 0.70 and 1.05 $\mu\text{mol/l}$ indicate marginal VAD (1,2,4,9,10). Furthermore, VAD can be characterized as a severe, moderate or mild public health problem, when the plasma retinol concentration is below 0.70 $\mu\text{mol/l}$ in over 20%, 10% to 20% or 2% to 10% of the target population, respectively (1,2,4,9,10).

Published data on vitamin A status of pre-school-age children in Papua New Guinea (PNG) is scanty (12-14). Some studies conducted in hospitals in several provinces indicated a prevalence of clinical xerophthalmia of 0.6% to 1.0% in pre-school-age children (12-14). A mini-survey of vitamin A status of pre-school-age children in coastal and highland provinces in PNG detected no clinical signs of VAD (13,14). The authors reported a prevalence of subclinical VAD of 9.1%, 10% and 15% in children in Western Highlands, Madang and Sepik Provinces, respectively (13,14). In the National Nutrition Survey conducted between 1996 and 1998, night-blindness was reported in 0.9% of pre-school-age children (13,14). There are, however, no data to indicate the vitamin A status of pre-school-age children in the National Capital District (NCD), PNG.

In an effort to prevent VAD among pre-

school-age children in PNG, the distribution of vitamin A capsules was included as part of the expanded program on immunization (EPI) in 2002 (13,14). According to the PNG National Department of Health (NDoH) protocol, the first dose of vitamin A is given together with the first measles vaccination at six months, and the second dose is given about six months later (13,14). Thus vitamin A supplementation (VAS) of pre-school-age children in PNG has been going on for several years. The NDoH protocol for supplementation includes monitoring of vitamin A status in areas already covered by supplementation programs. Recent reports from the PNG National Nutrition Survey carried out in 2005 (PNG NNS 2005) indicate a prevalence of moderate to severe VAD among pre-school-age children in the four regions of PNG (15). The PNG NNS 2005 did not provide data on the vitamin A status of pre-school-age children in the various provinces and districts in PNG (15).

The major objective of this study was to assess the vitamin A status of pre-school-age children resident in NCD, which is one of the districts in PNG that has been covered by the vitamin A supplementation program.

Subjects and Methods

Study site

The study was conducted between April and September 2008 in the NCD, which is one of the districts in the Southern Region in PNG. The NCD is the incorporated area around Port Moresby, the capital of PNG. Because of the difficulty in obtaining blood samples from healthy pre-school-age children in the households in NCD, the Children's Outpatient Clinic at Port Moresby General Hospital (PMGH) was selected as the specific study site. The PMGH is the major public general, specialist and reference hospital in NCD and PNG; it also serves as the teaching hospital for the School of Medicine and Health Sciences (SMHS), University of Papua New Guinea (UPNG).

Sample size

The sample size was calculated using a design effect of one, relative precision of 10%, and assumed prevalence rate of 20%, with a confidence level of 95% (16). The sample size of about 180 pre-school-age children

was considered adequate for a study with a predicted non-response rate of 25% (16).

Study design and sampling

This was a hospital outpatient-based cross-sectional study. All children aged between 6 and 59 months from whom blood was being collected for various reasons were enrolled in the study. Children with significant illness, those admitted to the ward and those who were not resident in NCD were excluded from the study. Further selection was by parental consent.

Collection of blood samples and data by questionnaire

About 0.3 ml of venous blood was transferred into an EDTA-coated microtainer, which was immediately put into a microtainer-box wrapped with aluminum foil for protection from light. The box was then put into a cool-box, kept at 4-8°C in the field and during transport to the Micronutrient Laboratory (MNL) in the SMHS, UPNG. The blood samples were centrifuged, after which aliquots of plasma were kept frozen at -70°C. A self-designed pretested questionnaire was used to collect demographic and other information, including the vitamin A supplementation history of each child.

Sample analysis and quality control

All reagents, including the internal standard, were of analytical grade and were components of the 'Clin-Rep' complete kit for assay of vitamins A and E in plasma by HPLC (17). A special reverse phase column, which was a component in the 'Clin-Rep' complete kit, was used to measure the concentration of vitamin A as retinol (17). The flow rate of the mobile phase was set at 1.5 ml/minute. The operating HPLC system used was the Waters Empower 2.0 software, configured for analysis of retinol in plasma. The wavelength of the HPLC detector was set at 325 nm (17).

The 'Levy-Jennings' charts and 'Westgard' rules were used for monitoring the internal bench quality control (QC) of the HPLC output data. Four levels of 'Clin-Chek' plasma retinol control samples were used for QC monitoring (17). The intra-assay coefficient of variation (CV) for each of the four QC samples ranged from 4.0% to 7.5%. The percent recovery of

plasma retinol was 95-98.5%.

The C-reactive protein in plasma was assayed using a commercial enzyme immunoassay kit (QuikRead – 101 Orion Diagnostica, Finland) (11). Inter- and intra-assay coefficients of variation for CRP were measured, using the controls provided by the manufacturer. The intra-assay CV was 3.5% and the inter-assay CV 4.0%.

Data analysis and interpretation

Data analysis was carried out using the Statistical Package for Social Sciences (SPSS) Version 11 for Windows. The Shapiro-Wilks test was used to assess normality of data. The Wilcoxon rank sum test, Mann-Whitney U test, chi-squared (Fisher's exact test) and Student's t-test were used as appropriate. Correlations were determined by Spearman's rank correlation coefficient ($\rho = \text{rho}$).

For interpretation of the data in the present study, plasma retinol concentrations below 0.70 $\mu\text{mol/l}$ and between 0.70 and 1.05 $\mu\text{mol/l}$ were used to indicate VAD and marginal VAD, respectively (1,2,4,9,10). VAD levels of 2-10%, 10-20% and above 20% were used to indicate mild, moderate and severe public health problems, respectively (4,9,10). As recommended by the manufacturer (11), elevated CRP indicating inflammation was defined as CRP >8.0 mg/l.

Ethical clearance

Ethical clearance and approval for this study were obtained from the Ethical and Research Grant Committee in the SMHS, UPNG, the Medical Research Advisory Committee (MRAC) in the National Department of Health (MRAC No 08/28) and the PMGH. Participation was voluntary; oral and signed informed consents were obtained from the parents of each of the children. The blood samples used were only from children whose parents gave consent.

Results

Permission to include their child in the study was sought from 181 parents or guardians. Informed consent was obtained from 140, but permission to use the blood samples was obtained from only 132 (response rate 73%). This gave a non-response rate of 27%, which was higher than the predicted non-response

rate of 25% used for calculating the sample size.

Of the 132 mothers, 92 (70%) did not have their baby book with them at the time of data collection for this study. In these cases, the information obtained was based on the mother's recollection.

The median age of the study children was 16.0 months and the interquartile range (IQR) 10.3 to 24.0 months.

The distribution curve of the plasma retinol concentration for the study children was not normal, according to the Shapiro-Wilks test ($p < 0.001$, $df = 132$). Figure 1 shows the box plot of the plasma retinol concentrations. The median plasma retinol concentration was $0.98 \mu\text{mol/l}$ and the IQR was $0.65\text{--}1.38 \mu\text{mol/l}$ (Table 1).

Of the 132 children, 35 (27%) had a plasma retinol concentration below $0.70 \mu\text{mol/l}$ and 37 (28%) had a plasma retinol concentration between 0.70 and $1.05 \mu\text{mol/l}$ (Table 1). The Spearman correlation ($\rho = -0.519$) indicated

a significant ($p = 0.01$) inverse relationship between the plasma retinol concentration and the plasma CRP level among the study children.

The 132 children were separated according to their plasma CRP levels. 75 (57%) of them had normal plasma CRP levels and in 57 (43%) the levels were elevated (CRP $> 8.0 \text{ mg/l}$).

The median age of the children with normal plasma CRP levels was 14.0 months (IQR = $9.0\text{--}19.0$ months), while for those with elevated plasma CRP levels it was 19.0 months (IQR = $15.0\text{--}26.0$ months). The difference in the mean age after log transforming the data was significant ($p = 0.018$).

The box plots in Figure 1 also represent the distributions of plasma retinol concentrations for the children with normal and elevated plasma CRP levels. The data were not normally distributed. Table 1 shows the medians and IQRs of the plasma retinol concentrations for children with normal and elevated plasma CRP levels. Statistical analysis, using the Mann-

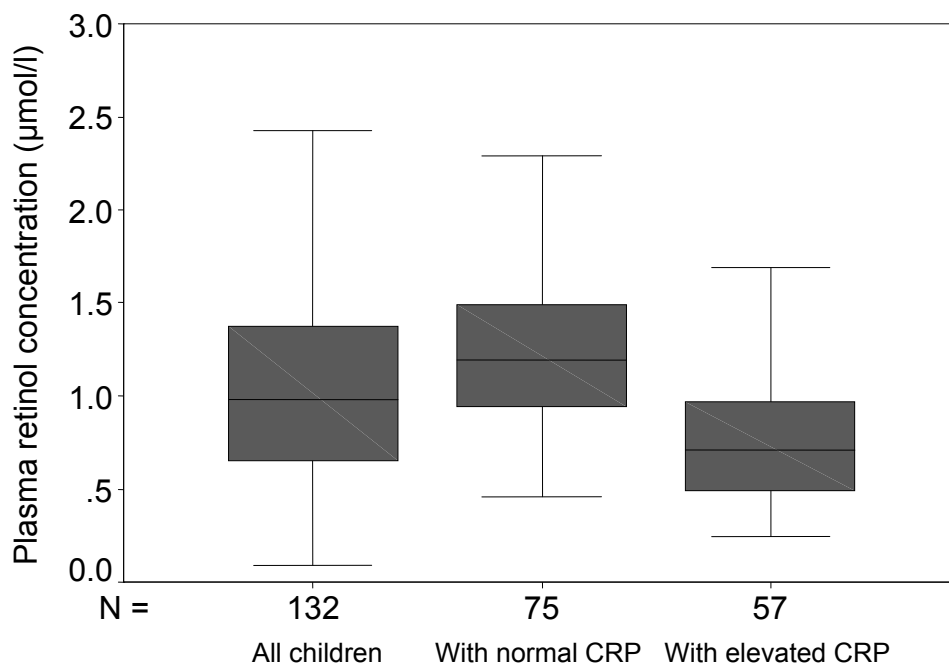


Figure 1. Box plots of plasma retinol concentration ($\mu\text{mol/l}$) for all the study children and for those with normal and elevated plasma C-reactive protein (CRP) levels.

TABLE 1

PLASMA RETINOL CONCENTRATION AND PERCENT BELOW CUT-OFF POINT THAT INDICATES VITAMIN A DEFICIENCY (VAD) FOR ALL THE STUDY CHILDREN AND FOR THOSE WITH NORMAL AND ELEVATED PLASMA C-REACTIVE PROTEIN (CRP) LEVELS

	All children (n = 132)	Children with normal CRP (n = 75)	Children with elevated CRP (n = 57)
Median retinol ($\mu\text{mol/l}$)	0.98	1.19	0.71
Interquartile range ($\mu\text{mol/l}$)	0.65-1.38	0.93-1.50	0.49-1.00
Mean retinol ($\mu\text{mol/l}$)	1.09	1.29	0.82
Standard deviation	0.61	0.64	0.43
95% CI ($\mu\text{mol/l}$)	0.98-1.19	1.15-1.44	0.70-0.93
Percent (number) with retinol between 0.70 and 1.05 $\mu\text{mol/l}$ (marginal VAD)	28.0% (37)	26.7% (20)	29.8% (17)
Percent (number) with retinol <0.70 $\mu\text{mol/l}$ (VAD)	26.5% (35)	10.7% (8)	47.4% (27)

CI = confidence interval

Whitney U test, indicated that the plasma retinol concentrations for the children with normal plasma CRP levels were significantly ($p < 0.001$) higher than for those with elevated plasma CRP levels.

The plasma retinol concentration was below 0.70 $\mu\text{mol/l}$ in 11% of the 75 children with normal plasma CRP levels and 27% of them had a plasma retinol concentration between 0.70 and 1.05 $\mu\text{mol/l}$.

A significant inverse relationship ($p = -0.455$, $p = 0.01$) was obtained between plasma retinol concentration and plasma CRP level for children with elevated plasma CRP levels.

In order to assess the vitamin A supplementation coverage among the population in this study, each mother was asked whether their child had ever received vitamin A capsules. According to their recall, 108 (82%) of the 132 mothers responded in the affirmative, and 24 (18%) "did not remember" whether their children had ever received vitamin A capsules (Table 2). There was no significant ($p > 0.05$) difference in the plasma retinol concentrations between these two groups of children. However, the

plasma retinol concentration was significantly ($p = 0.038$) higher in the children with normal CRP in the 'affirmative' group than in their counterparts in the 'did not remember' group. In addition, plasma retinol concentration was below 0.70 $\mu\text{mol/l}$ in 7% of those with normal plasma CRP in the 'affirmative' group compared to 22% of their counterparts in the 'did not remember' group (Table 2).

Further analysis of the data indicated that 31 (29%) of the 108 children in the 'affirmative' group received a vitamin A capsule within the previous six months. This information was further confirmed in the baby book of each of these children. Table 2 shows the plasma retinol concentration of this group of children. The plasma retinol concentration in the children with normal plasma CRP was significantly higher ($p = 0.001$) than in those with elevated plasma CRP. In addition, 4% of the children with normal plasma CRP had a plasma retinol concentration below 0.70 $\mu\text{mol/l}$ compared to 57% of their counterparts with elevated plasma CRP.

Of the 108 mothers in the 'affirmative' group 37 (34%) indicated that their children had received vitamin A capsules on two different occasions since birth. This information was

TABLE 2

PLASMA RETINOL CONCENTRATION OF STUDY CHILDREN GROUPED ACCORDING TO THEIR REPORTED HISTORY OF VITAMIN A SUPPLEMENTATION

	Affirmative, received vitamin A capsule		Did not remember ever receiving vitamin A capsule		Received vitamin A capsule within the previous 6 months		Received vitamin A capsule twice since birth				
	All	Elevated CRP	All	Elevated CRP	All	Elevated CRP	All	Elevated CRP			
Percent (number)	81.8% (108)	52.8% (57)	47.2% (51)	75% (18)	25% (6)	28.7% (31)	77.4% (24)	22.6% (7)	34.3% (37)	73.0% (27)	27.0% (10)
Median retinol (µmol/l)	0.99	1.28	0.66	0.93	1.11	1.08	1.17	0.61	1.07	1.33	0.68
Interquartile range (µmol/l)	0.62-1.37	1.00-1.28	0.49-0.94	0.72-1.44	0.70-1.81	0.97-1.42	1.00-1.47	0.49-0.97	0.59-1.35	1.07-1.57	0.49-0.99
Mean retinol (µmol/l)	1.07	1.36	0.76	1.12	1.21	1.30	1.47	0.73	1.04	1.34	0.73
Standard deviation	0.62	0.66	0.37	0.56	0.66	0.82	0.86	0.28	0.48	0.45	0.30
95% CI (µmol/l)	0.96-1.19	1.18-1.53	0.65-0.86	0.84-1.40	0.52-1.90	1.00-1.60	1.10-1.83	0.47-0.99	0.81-1.26	1.02-1.66	0.53-0.95
Percent (number) with retinol <0.70 µmol/l (VAD)	27.8% (30)	7.0% (4)	51.0% (26)	22.2% (4)	16.7% (1)	16.1% (5)	4.2% (1)	57.1% (4)	16.2% (6)	3.7% (1)	50.0% (5)
Percent (number) marginal VAD	26.9% (29)	22.8% (13)	31.4% (16)	38.9% (7)	33.3% (2)	25.8% (8)	25.0% (6)	28.6% (2)	8.1% (3)	-	30.0% (3)

CRP = C-reactive protein; CI = confidence interval; VAD = vitamin A deficiency; marginal VAD = retinol level between 0.70 and 1.05 µmol/l

confirmed in the baby book of 9 of the 37 children. The plasma retinol concentration of this group of children is presented in Table 2. The plasma retinol concentration for those with normal plasma CRP was significantly ($p = 0.002$) higher than for those with elevated CRP.

In all instances, significant ($p < 0.05$) inverse relationships were obtained between plasma retinol concentrations and plasma CRP levels for children with elevated plasma CRP levels.

Of the 108 mothers in the 'affirmative' group, 40 (37%) could not state the exact number of times their children had received vitamin A capsules.

For further analysis of the supplementation data (Table 3), the 132 children were separated into three age groups. There were 37 (28%) in the 6 to 11 months age group, 65 (49%) in the 12 to 24 months age group and 30 (23%) in the over 24 months age group.

The results show that 29 (78%) of the 37 children in the 6 to 11 months age group had received a vitamin A capsule within the previous six months. The age range of the remaining 8 children that had not received any vitamin A capsule was 6 to 8 months (plasma CRP was elevated in these children).

Our results also show that 54 (83%) of the 65 children in the 12 to 24 months age group and 25 (83%) of the 30 children in the over 24 months age group had received a vitamin A capsule at least once. Two of the children in the 12 to 24 months age group received a vitamin A capsule within the previous six months, as was recorded in their baby books.

The plasma CRP levels were normal in 78%, 52% and 40% of the children in the 6 to 11 months, 12 to 24 months and over 24 months age groups, respectively. The plasma retinol concentration and percent below the cut-off points that indicate marginal VAD and VAD for all the children and for those with normal plasma CRP in the three age groups are presented in Table 3. The plasma retinol concentration of the children with normal plasma CRP in the over 24 months age group was significantly lower than that for the children with normal plasma CRP in the 6 to 11 months ($p = 0.035$) and 12 to 24 months ($p = 0.019$) age groups.

The plasma retinol concentrations were significantly higher ($p < 0.05$) in the children with normal plasma CRP than in their counterparts with elevated plasma CRP in the corresponding age groups.

The plasma retinol concentration was below $0.70 \mu\text{mol/l}$ in 7%, 9% and 25% of the children with normal plasma CRP in the 6 to 11 months, 12 to 24 months and over 24 months age groups, respectively.

74 (56%) of the 132 children were males with a median age of 17.0 months (IQR = 12.0-25.0 months) and 58 (44%) were females with a median age of 14.0 months (IQR = 8.8-19.0 months). The age of the male children was significantly higher ($p = 0.018$) than that of the female children.

The box plots of the plasma retinol concentrations for all the male and female children are presented in Figure 2.

The median and IQR plasma retinol concentrations for the male children were $0.93 \mu\text{mol/l}$ and $0.65\text{-}1.32 \mu\text{mol/l}$, respectively (Table 4). The corresponding values for the female children were $1.08 \mu\text{mol/l}$ and $0.71\text{-}1.43 \mu\text{mol/l}$. There was no significant ($p = 0.091$) difference between the plasma retinol concentrations of the male and female children. The plasma retinol concentration was below $0.70 \mu\text{mol/l}$ in 28% of the male and in 24% of the female children. The Spearman correlation indicated a significant inverse relationship between the plasma retinol concentration and plasma CRP level for the male ($p = -0.414$; $p = 0.01$) and the female ($p = -0.617$; $p = 0.01$) children.

Results for the male and female children were separated according to their plasma CRP levels. Of the 74 male children, 50% had normal and 50% had elevated plasma CRP levels. Of the 58 female children, 66% had normal and 34% had elevated plasma CRP levels. Figure 2 shows the box plots for the plasma retinol concentrations for these male and female groups. The medians and IQRs of the plasma retinol concentrations for the male and female children with normal and elevated plasma CRP levels are presented in Table 4. The table also shows the percent of children in each group with plasma retinol concentration below $0.70 \mu\text{mol/l}$ and those between 0.70 and $1.05 \mu\text{mol/l}$.

TABLE 3

PLASMA RETINOL CONCENTRATION AND PERCENT BELOW CUT-OFF POINT THAT INDICATES VITAMIN A DEFICIENCY (VAD) FOR STUDY CHILDREN IN THE VARIOUS AGE GROUPS AND FOR THOSE WITH NORMAL PLASMA C-REACTIVE PROTEIN (CRP) LEVELS

	Children aged 6-11 months		Children aged 12-24 months		Children aged >24 months	
	All (n =37)	Normal CRP (n = 29)	All (n = 65)	Normal CRP (n = 34)	All (n = 30)	Normal CRP (n = 12)
Median retinol (µmol/l)	1.07	1.09	0.95	1.29	0.83	1.05
Interquartile range (µmol/l)	0.87-1.07	0.98-1.46	0.62-1.45	0.93-1.63	0.53-1.17	0.62-1.33
Mean retinol (µmol/l)	1.26	1.38	1.07	1.34	0.91	0.95
Standard deviation	0.77	0.81	0.54	0.50	0.45	0.43
95% CI (µmol/l)	1.00-1.51	1.07-1.69	0.94-1.21	1.16-1.51	0.74-1.07	0.68-1.23
Percent (number) with retinol between 0.70 and 1.05 µmol/l (marginal VAD)	32.4% (12)	34.5% (10)	24.6% (16)	23.5% (8)	33.3% (10)	25.0% (3)
Percent (number) with retinol <0.70 µmol/l (VAD)	16.2% (6)	6.9% (2)	30.8% (20)	8.8% (3)	30.0% (9)	25.0% (3)

CI = confidence interval

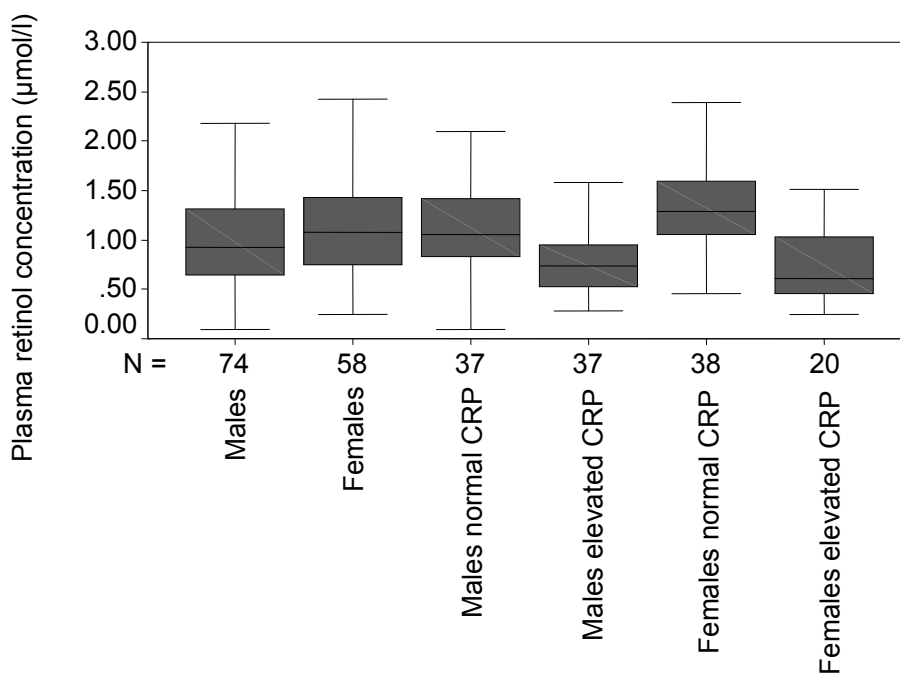


Figure 2. Box-plots of plasma retinol concentration (µmol/l) for all the male and female study children and for those with normal and elevated plasma C-reactive protein (CRP) levels.

The plasma retinol concentrations for the male children with normal plasma CRP was significantly ($p = 0.001$) higher than for those with elevated plasma CRP. A similar result ($p = 0.001$) was obtained for the female children.

No significant difference ($p = 0.054$) was obtained when the plasma retinol concentration for the male children with normal plasma CRP was compared with that of the females with normal plasma CRP.

Of the 37 male children with normal plasma CRP there were 14% with plasma retinol concentration below 0.70 µmol/l and 38% with plasma retinol concentration between 0.70 and 1.05 µmol/l. Of the 38 female children with normal plasma CRP only 8% had a plasma retinol concentration below 0.70 µmol/l and 16% had a plasma retinol concentration between 0.70 and 1.05 µmol/l.

The median and mean plasma retinol concentrations were higher, and the proportions with plasma retinol below 0.70

µmol/l were lower, in both the male and female children with normal plasma CRP levels than in their counterparts with elevated plasma CRP levels.

Discussion

The high non-response rate (27%) indicates some of the problems encountered by research projects that require collection of biological samples from 'apparently healthy' children. Higher non-response rates have been reported by others (18,19). Parents' awareness of the voluntary nature of their consent to the collection of biological samples from their children for the purpose of research may be one possible explanation for the negative response from some parents.

In the present study, VAD was prevalent in 27% of the study children. This was higher than the 9.1%, 10% and 15% prevalence reported respectively in children in Western Highlands, Madang and Sepik Provinces in PNG (13,14), but similar to the 25.6% prevalence recently

TABLE 4

PLASMA RETINOL CONCENTRATION AND PERCENT BELOW CUT-OFF POINT THAT INDICATES VITAMIN A DEFICIENCY (VAD) FOR ALL THE MALE AND FEMALE STUDY CHILDREN AND FOR THOSE WITH NORMAL AND ELEVATED C-REACTIVE PROTEIN (CRP) LEVELS

	Male children			Female children		
	All (n = 74)	Normal CRP (n = 37)	Elevated CRP (n = 37)	All (n = 58)	Normal CRP (n = 38)	Elevated CRP (n = 20)
Median retinol (µmol/l)	0.93	1.06	0.73	1.08	1.29	0.61
Interquartile range (µmol/l)	0.65-1.32	0.82-1.44	0.52-0.96	0.71-1.43	1.05-1.61	0.43-1.08
Mean retinol (µmol/l)	0.99	1.13	0.85	1.21	1.47	0.76
Standard deviation	0.46	0.42	0.45	0.74	0.78	0.40
95% CI (µmol/l)	0.88-1.09	0.99-1.27	0.70-1.00	1.02-1.41	1.21-1.73	0.58-0.94
Percent (number) with retinol between 0.70 and 1.05 µmol/l (marginal VAD)	35.1% (26)	37.8% (14)	32.4% (12)	19.0% (11)	15.8% (6)	25.0% (5)
Percent (number) with retinol <0.70 µmol/l (VAD)	28.4% (21)	13.5% (5)	43.2% (16)	24.1% (14)	7.9% (3)	55.0% (11)

CI = confidence interval

reported for pre-school-age children in PNG (15) and within the 20.5% to 35.3% range of prevalence in the four regions of PNG (15).

The VAD prevalence in the present study was also higher than the 11.0% and 15.6% reported for pre-school-age children in Manisa and Izmir regions in Turkey (20,21), the 24.1% in Brazil (3) and the 7.8% to 15.75% in Beijing and Guizhou in China (22). However, it was lower than the 30.0%, 29.5%, 58.7%, 61.2% and 32.1% prevalence reported for pre-school-age children in Beijing, Nigeria, West Bengal in India, Republic of the Marshall Islands and Brazil, respectively (22-26).

Marginal VAD was prevalent in 28% of the study children. This was higher than the 16.8% and 23.2% reported for pre-school-age children in urban areas in Beijing, China (22), but lower than the 41.8% and 52.4% reported for the rural areas around Beijing (22).

Our results indicated a prevalence of VAD of 28% in the male and 24% in the female study children. These values were lower than the 60.3% and 62.0% prevalence of VAD reported for male and female pre-school-age children in West Bengal in India (24).

Marginal VAD was higher in the male (35%) than the female (19%) study children in NCD. Our findings support the observation by Benn et al. (27) that infant boys may be more vitamin A deficient than infant girls. These results did not reflect the plasma CRP levels in the children.

According to the recommended criteria (4,9,10), VAD should be characterized as a severe public health problem among pre-school-age children in NCD at the time of this study. However, this characterization was of concern because of the inverse correlation obtained between the plasma retinol concentration and plasma CRP level (Spearman $\rho = -0.519$, $p = 0.01$). The high percentage (43%) of study children with an elevated plasma CRP level indicates a high prevalence of subclinical infection. This figure, however, was lower than the 49.6% of pre-school-age children with an elevated plasma CRP level reported in the Republic of the Marshall Islands (25).

The prevalence of VAD in the study children with normal plasma CRP was 11%, which indicated a moderate public health problem.

Children with an elevated plasma CRP had a much higher (47%) prevalence of VAD, which indicated a severe public health problem.

Our findings are consistent with recent scientific opinion, which increasingly suggests that the negative impact of subclinical infection on plasma retinol concentration may lead to overestimation of VAD (4-6).

The VAS coverage (82%) obtained in the present study was lower than the 83.1% and 96.0% reported for pre-school-age children in Ethiopia and Nepal, respectively, but higher than the 42.8% and 74.0% reported from Cambodia and the Welayta Zone in Ethiopia, respectively (28,29).

Our data indicate that the VAS coverage in NCD at the time of this study was 'on track' to achieve the WHO recommended coverage of approximately 80% of children aged 6-59 months within target populations (30). Despite this achievement, the VAS coverage (78%) among the younger (6 to 11 months) children was slightly lower than the 83% coverage obtained for the older children in the present study. This indicates the need for program planners to implement additional strategies to improve and sustain the VAS program among pre-school-age children in the NCD.

The prevalence of VAD (7%) and marginal VAD (23%) in the study children with normal plasma CRP in the VAS 'affirmative' group was significantly lower than the prevalence of VAD (22%) and marginal VAD (39%) in the study children with normal plasma CRP in the 'did not remember' VAS group. This tends to confirm the general observation that VAS is an effective intervention strategy for reducing the incidence of illness among children aged 6-59 months (3,5,28).

Our data also indicate that among the children in the VAS 'affirmative' group both VAD and marginal VAD were significantly associated with elevated plasma CRP levels.

Mild VAD was prevalent in the children with normal plasma CRP in the 6 to 11 months and 12 to 24 months age groups, comparable to the prevalence of severe VAD in the over 24 months age group. Marginal VAD was, however, more prevalent in the 6 to 11 months old children with normal plasma CRP than in the other two age groups. This indicates that the younger children are at greater risk

of developing moderate to severe VAD in the event of mild infection without a corresponding increase in the intake of vitamin A. These data underscore the need to further strengthen and expand the on-going VAS program in the NCD.

The prevalence of VAD in the male and female children with normal plasma CRP was 14% and 8%, which indicates moderate and mild public health problems, respectively.

The high prevalence of marginal VAD in the children with normal plasma CRP obtained in this study should be of concern to program planners, because, from the public health point of view, it indicates that a significant number of these children are at risk of developing VAD. This further indicates the urgent need to strongly advocate for the implementation of appropriate strategies and policies aimed at improving the vitamin A status of pre-school-age children in NCD. There is a need for intensive nutrition education, information and parental awareness campaigns, emphasizing the significance of vitamin A for children.

Our findings should be of concern to program planners involved in the VAS program in NCD. There is a need to improve and strengthen the existing monitoring of the VAS program in NCD, to ensure its efficiency, sustainability and functionality.

Although this was a hospital-based study, the prevalence of subclinical and marginal VAD in the study children with and without elevated plasma CRP strongly suggests the need for continuous monitoring of the vitamin A status of vulnerable groups (children aged 6-59 months and pregnant and lactating mothers) in NCD.

Conclusions

The prevalence of VAD in the pre-school-age children aged 6-59 months with normal plasma CRP was 11%, indicating a moderate public health problem. The prevalence of VAD in the male and female children with normal plasma CRP was 14% and 8%, which indicates moderate and mild public health problems, respectively.

The high prevalence of marginal VAD in the children with normal plasma CRP should be of concern to program planners, because, from the public health point of view, it indicates that

a significant number of these children are at risk of developing VAD.

The VAS coverage in NCD is 'on track' to achieve the WHO recommended coverage of approximately 80% of children aged 6-59 months within target populations.

There is, however, a need for intensive nutrition education, information and awareness campaigns emphasizing the significance of vitamin A for pre-school-age children, and for an efficient, sustainable and functional monitoring system to strengthen and improve the VAS program in NCD.

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