

ORIGINAL COMMUNICATION

Are lifestyle factors good predictors of retinol and vitamin C deficiency in apparently healthy adults?

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Objective: To examine interrelationships between (1) dietary habits, (2) socioeconomic and (3) environmental factors, and their impact on plasma retinol and plasma ascorbic acid.

Design: Cross-sectional study on adults from Western India.

Setting: Rural, semi urban, urban higher/middle/lower socioeconomic regions (HSE/MSE/LSE) having diverse dietary habits and environmental conditions.

Subjects: A total of 214 men and 108 women (20–50 y), apparently healthy and non-anemic.

Main outcome measures: Food intake by food frequency questionnaire, weight, height, age, smoking, environmental score, education, income, plasma retinol and plasma ascorbic acid.

Results: Mean plasma retinol in women ($24.84 \pm 5.1 \mu\text{g/dl}$) and men ($24.75 \pm 4.53 \mu\text{g/dl}$) were not significantly different and 21% had plasma retinol below $20 \mu\text{g/dl}$. Mean plasma ascorbic acid in women ($0.35 \pm 0.12 \text{ mg/dl}$) and men ($0.30 \pm 0.12 \text{ mg/dl}$) was similar with 75% having plasma ascorbic acid below 0.4 mg/dl . Vitamin A intake (as retinol equivalent) and plasma retinol showed a significant dose response ($P < 0.05$) but not vitamin C intake and plasma ascorbic acid. Plasma retinol showed significant correlation with income ($\rho = 0.24$), education ($\rho = 0.27$), and environment ($\rho = 0.21$; $\rho = 0.0001$). Similar correlations with plasma ascorbic acid were 0.29, 0.31, -0.23 respectively ($P = 0.0001$). Logistic regression showed education, environment, green leafy vegetables (GLV) and milk intake as predictors of plasma retinol deficiency, while non-sweet fruit intake, education and passive smoking for plasma ascorbic acid deficiency ($P < 0.05$).

Conclusions: Subnormal status of retinol and vitamin C emphasizes the need to increase consumption of fruit, GLV and milk products, and also better education and environment. Avoiding passive smoking demands attention in order to improve levels of these vitamins.

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Keywords: plasma retinol; plasma ascorbic acid; socioeconomic status; fruit/vegetable intake; passive smoking

Introduction

Adequate nutritional status of carotenoids and vitamin C has attained a greater importance during the past decade, as a protective role of vitamins due to their antioxidative action

has been suggested in some health disorders (Saubert & Machlin, 1992; Basu & Dickerson, 1996). Major dietary sources of carotenoids and vitamin C for vegetarians are fruit and vegetables. Increased intake of fruit and vegetables has been associated with a reduced risk of various diseases including cancer and cardiovascular diseases (Ness & Powles, 1997; Law & Morris, 1998; Ness *et al*, 1999; Steinmetz & Potter, 1996; Gillman *et al*, 1995; Regina *et al*, 1991).

Dietary levels of vitamins also depend on several other factors such as age and socioeconomic status (Hjartaker & Lund, 1998). In developing countries like India, socioeconomic factors and customary restrictions govern intake of foods rich in vitamin A and C (eg fruit, vegetables, animal foods, milk products). Assessment of vitamin status and identifying influencing factors in apparently healthy individuals from different socioeconomic and environmental background has significance for drafting preventive health

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Contributors: SAC was the principal investigator of the project, participated in data collection, completed data analysis and statistical treatment of the data, and contributed to writing the manuscript. VVA was co-investigator of the projects, participated in data collection, was in charge of biochemical (food, blood) estimations, and contributed to writing the manuscript. SSM and KVT worked as Project Assistants, assisted in the survey and laboratory analysis of food and blood samples.

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measures against risk of vitamin deficiency. There is a need therefore to explore interrelationships of dietary intakes, socioeconomic status and environment with plasma levels of these vitamins and identify influential factors.

Most of the studies reporting vitamin A status in Indians are done on pregnant women and children. Information on plasma levels of vitamin A and C in an apparently healthy vegetarian population is scarce (Sinha & Sharma, 1998, Vijayalakshmi & Rema, 1994). The present paper aims at (i) assessing the retinol and vitamin C status of an adult population, and (ii) examining risk of plasma retinol deficiency and plasma ascorbic acid deficiency using dietary intake, age, socioeconomic and environmental conditions as explanatory variables.

Methods

Study design

The study was conducted in Maharashtra State, which is one of the major states in India having an area of 307 713 square kilometers and a population of around 100 million with 51.7% males and 48.3% females. Some 61.3% of the population is in rural areas and 38.6% in urban and semi-urban zones. Rural and tribal populations have agriculture as their main occupation, while semi-urban and urban areas included individuals from all socioeconomic classes having occupations like businessmen, industrial workers, employees of banks/offices/institutes, students, housewives, hawkers etc. Accordingly the urban population was divided into higher/middle/lower socioeconomic classes (HSE/MSE/LSE). Rural and tribal populations have to depend on the local food commodities, while semi-urban and urban populations have better access to a variety of foods in the market.

Subjects

Criteria for selection of subjects were: (1) not suffering from major illnesses like diabetes or hypertension; and (2) not taking medicines or vitamin-mineral supplements. A random sample of 500 individuals (20–45 y) was selected through several health camps organized by local voluntary organizations in different areas in Maharashtra state of Western India viz. rural, semi-urban and urban zones during February 1998 to June 2000. Out of these 500, 75 had a history of anaemia, while 32 were found to have joint problems/inflammation, or recurrent infections. Information on 71 could not be obtained because of inadequate blood samples, or incomplete questionnaires, although their general characteristics matched with the study group. Complete information was available for 322 individuals (214 men and 108 women) apparently healthy and non-anaemic, and these data have been reported in the present paper. An informed written consent was obtained from subjects towards voluntary participation. Subjects were asked to come in the morning at 8am on an empty stomach and a fasting blood sample of 10ml was collected by the doctor

while doing a clinical examination. A food frequency questionnaire and proforma of socioeconomic, environmental information was filled in by interview method.

Cooked food frequency questionnaire (FFQ) and nutrient intakes

A semi quantitative FFQ was designed to ask questions about usual dietary intake. The period of the FFQ was taken as 1 y to cover all seasonal fruit and vegetables. The FFQ covered 278 food items, traditionally consumed in India, which were classified under food categories: cereals, legumes, vegetables, fruit, milk, animal food, snacks, sweets and deserts. The questionnaire was administered by trained investigators by interview. The subjects were asked how often on average they had consumed each food item during the year, the frequency of consumption per day, and amount in terms of standard measures like cup, bowl, spoon etc. For example, for some food items, consumption may be seasonal or on festive occasions, while for others, like milk, consumption may be twice a day, one glass at a time. FFQ was standardized to get adequate information about food choices of different sections of population and to obtain suitable consumption categories.

Frequency for each food item was defined as never/once per month/once in two months/once a week/once per week/twice per week/three times per week/once a day/twice a day and so on. The response intervals were adjusted to the food item and in case of reversal items in one category, a weighted frequency was computed. Weights of the portion units were decided on the basis of average weight of each food item for that portion size collected from different households representing the sample. As the questionnaire was interview-administered, there were no blanks in the form. Subjects were asked about any multi-vitamin supplements.

Repeatability of the FFQ was tested by again administering the same FFQ to a random sample of 30 individuals after one month through an interview by the same trained investigator. The response to both the FFQ agreed well with respect to consumption of foods and computed intakes of energy and nutrients (value of test statistic t ranged from 0.2 to 0.98. $P > 0.1$ and Pearson's correlation coefficient r from 0.79 to 0.96 for different nutrient and food intakes.

Daily food consumption in eight food groups was computed for every individual using the information in the FFQ about frequency and quantity. Fruit consumption was further classified into sweet fruit, eg banana or chiku, and non-sweet fruit like guava, orange or lime.

Measurements

Dietary intake of energy, protein, fat, β -carotene and vitamin C was computed using nutrient values generated in our laboratory for individual cooked food items. The estimation of nutrient contents was done as per Raghuramulu *et al*

(1983). Retinol and β -contents were estimated for all the foods but retinol was detectable only in animal foods. Vitamin A intake was expressed as retinol equivalents per day.

Height and weight recorded using measuring scale and weighing balance to the nearest 0.1 cm and 0.5 kg respectively. Subjects underwent a routine medical check-up, which included physical examination, blood pressure, and past and present health complaints by a medical doctor. Subjects having medical fitness were included in the analysis.

Education was asked as an open-ended question as illiterate, primary school, high school, graduate and postgraduate studies. The data was subsequently divided into four classes giving years of education: less than 4, 5–10, 11–15 and above 15, as per the existing educational system.

Information on age, environment and socioeconomic status were collected by the investigator through a separate questionnaire by interview. Income was asked for as total monthly income of the participant and the family, and family size. Per capita daily income was calculated and classified into four categories of income using the National Sample Survey (NSS, 1996) as the basis. Per capita per day income categories were: less than Rs20; Rs20–50; Rs50–100; and Rs100 and above. The average cost of 1 litre of milk during the period of survey was Rs14, wheat Rs10 per kg, rice Rs14 per kg, vegetables Rs12–30 per kg (cabbage, bottle gourd, cauliflower, beans) and fruit Rs15–60 per kg (banana, guava, apple, cherries, strawberries), indicating the relative value of these income levels. Thus a person earning Rs20/day having an average family size of four would find it difficult to spare money for milk/fruit/vegetables.

Environmental grading was considered as an explanatory variable. Under environmental conditions, factors such as type of housing, ventilation, surroundings, overcrowding, water supply, sewage system etc were studied. Each of these factors was given a score on a 10-point scale. Scores of all the variables were added, the maximum score being 100. A pooled score above 70 was termed good, between 50 and 70 fair, 35–50 bad and less than 35 poor (NSS, 1996; Gokhale *et al*, 1993).

Blood analysis

Fasting venous blood (10 ml) was collected in EDTA bulbs for every subject (not more than 12 h fasting). The samples were brought to the laboratory on the same day in ice bags within 2 h and centrifuged at 2000 rpm for 15 min. Plasma was separated and levels of retinol and ascorbic acid were estimated.

Plasma retinol was estimated by photofluorometer (Raghuramulu *et al*, 1983) using excitation at 350 nm and fluorescence at 490 nm. Plasma ascorbic acid was estimated by spectrophotometer. Reduction of ascorbic acid to dehydro

ascorbic acid by 2,6-dichlorophenol indophenol was estimated at 520 nm.

Haemoglobin was estimated by cyanmethaemoglobin method using standard kit (Qualigens Diagnostics, Glaxo India Ltd.).

Statistical methods

Nutrient calculations were done by computer program in C and Microsoft Excel version MS-OFFICE 2000. Statistical analyses were done by SPSS version 7.5.2 for Windows 16 May, 1997). Results were considered significant at $P < 0.05$. Parametric methods were used when analyzing anthropometric and socioeconomic variables, as these distributions were found to be normal by Kolmogorov–Smirnov test. The distributions of intake of food and nutrients were positively skewed; therefore non-parametric methods were used to test the differences between groups. Median values were reported for these variables. Univariate analyses were first carried out for the probable factors. Any factor with a probability of rejection less than 0.25 was considered as a candidate for the multivariate model along with all variables of known biological significance. Multiple logistic regression analyses were used to examine simultaneously the effect of food consumption, age, body mass index (BMI), education and socioeconomic status on the adequacy of plasma levels of retinol and vitamin C. A forward selection procedure was employed to include variables in the multivariate model. Importance of variables included in the model was verified by comparing each estimated coefficient with the coefficient from the univariate model containing that variable. All possible interactions of independent factors which were plausible, such as income and education, income and milk consumption, age and fruit–vegetable intakes etc, were entered in the model initially. However, no interaction terms remained in the final equation. To test adequacy of the model a small data set was set aside from the same survey. This was referred as the external data set. Goodness of fit of the model was tested by Hosmer–Lemshow test.

Results

Characteristics of the study participants are presented in Tables 1 and 2. Mean age ranged from 28.19 ± 8.62 in urban LSE to 32.67 ± 7.31 y in rural area in men. The age of the women ranged from 28.0 ± 9.72 in urban MSE to 31.29 ± 8.36 y in rural area. Mean BMI of the men and women was highest in the urban HSE (23.11 ± 3.9 , 21.55 ± 3.3 kg/m², respectively) and lowest in the rural population (19.42 ± 3.5 , 18.15 ± 3.2 kg/m²).

Table 2 gives socioeconomic and environmental characteristics along with distributions for plasma retinol and ascorbic acid. For estimating marginal to severe retinol and vitamin C deficiency, the classification given by Matilainen *et al*, (1996) and Grusse & Watier (1993) was used. Plasma ascorbic acid level was within normal range for 24% of men

and 33% of women. Only 12.8% of men and 14.8% of women showed normal plasma retinol status. With the cut-off of 20 µg/dl for deficient retinol concentration as per IVACG recommendations (Olson, 1990), 20.6% of men and 30.2% of women showed deficient levels.

Further examination of interrelationships among these characteristics was undertaken. Level of education was negatively associated with age ($r = -0.23$, $P = 0.0001$) but positively linked with income ($r = 0.536$, $P = 0.0001$). Education was also positively correlated with plasma retinol ($r = 0.46$, $P = 0.0001$) and ascorbic acid. Plasma retinol and ascorbic acid were not correlated with age and BMI. Environment was positively correlated with plasma retinol ($r = 0.272$, $P = 0.0001$) and ascorbic acid ($r = 0.255$, $P = 0.0001$).

Table 3 gives observed food intakes, which reveal the general dietary habits of the subjects. Median intake of cereals and legumes was below recommended dietary intakes for Indians (RDI; ICMR 1990) in case of men and women from different regions. Fruit intakes were below RDI in almost 44% of women and 34% of men from different

regions. All the subjects consumed leafy vegetables and other vegetables in meager amounts. Animal food intakes were greatly below RDI in most of the subjects and a quarter of the subjects were not eating animal food at all. Intake of milk and milk products was adequate in only 34.6% of men and 25% of women, mainly from urban HSE and MSE classes.

Table 2 Socioeconomic, environmental and biochemical parameters of the study population

Variable	Percentage of men	Percentage of women
Education (number of years)		
< 4	11.2	23.4
5–10	50.0	29.0
11–15	30.4	30.8
> 15	8.4	16.8
Per capita income (Rs/day)		
< 20	31.3	39.3
20–50	40.2	29.9
50–100	17.3	11.2
≥ 100	8.4	17.8
Environmental conditions		
Good	18.7	20.6
Fair	42.5	45.8
Average	32.2	29.0
Poor	4.2	3.7
Plasma retinol (µg/dl)		
≤ 10	0.4	1.4
11–20	20.2	26.8
21–30	66.6	57.0
> 30	12.8	14.8
Plasma vitamin C (mg/dl)		
< 0.2	19.62	12.96
0.2–0.4	55.14	51.85
≥ 0.4	24.30	33.33

The cost of 1 litre of milk during the period of survey was Rs16, while a cereal like wheat was Rs10 per kg, indicating the relative value of these income levels. Of the total subjects, only 5.6% were smokers while 15.1% of subjects, a family member was a smoker.

Table 1 Characteristics of the study population

Variable	Women (n = 108)		Men (n = 214)	
	Mean ± s.d.	95% CI	Mean ± s.d.	95% CI
Age (y)	29.17 ± 7.9	(27.4, 30.9)	29.8 ± 7.9	(28.5, 31.0)
Height (cm)	151.84 ± 7.3	(150.2, 153.4)	163.3 ± 7.5	(162.1, 164.5)
Weight (kg)	48.03 ± 9.9	(45.8, 50.2)	55.5 ± 9.2	(54.2, 56.8)
BMI (kg/m ²)	20.81 ± 3.9	(19.9, 21.7)	20.8 ± 3.3	(20.3, 21.3)
Plasma retinol (µg/dl)	24.84 ± 5.1	(23.7, 25.9)	24.75 ± 4.53	(24.0, 25.5)
Plasma ascorbic acid (mg/dl)	0.35 ± 0.12	(0.32, 0.37)	0.30 ± 0.116	(0.28, 0.32)
Hb (g/dl)	13.01 ± 1.05	(12.8, 13.2)	15.41 ± 1.03	(15.3, 15.6)

Mean BMI of both men and women in rural semi-urban higher, middle and lower socioeconomic classes was within normal range (20–25), but was subnormal in rural population (19.4, 18.1).

Table 3 Median food consumption (g/day)

Region	Cereal	Legume	Fruit	GLV	Other vegetables	Animal food	Milk product	Non-sweet fruit
Men								
Rural (59)	312.4	23.8	41.4	19.2	34.6	12.7	57.4	10.9
Semiurban (74)	312.3	20.2	44	15.8	25	11.9	51.7	11
Urban HSE (24)	331.1	21.5	39.7	18.1	30.5	5.6	189.5	21.7
Urban MSE (31)	357.8	35.7	51.6	18.8	49.8	31.6	87.9	17.6
Urban LSE (26)	331.4	21.2	26.7	10.6	33.7	13.5	114.2	9.6
Balanced diet ^a	460	40	30	40	75	60	150	—
Women								
Rural (27)	247.2	19.9	18.4	15.1	28.8	4.8	6.2	7.4
Semiurban (26)	238.2	20.8	30	13.8	19.6	5.7	42.3	11.8
Urban HSE (21)	241.1	20.8	62.4	20.5	32	3.9	158.9	23
Urban MSE (15)	243.2	18.2	44.3	15.9	18.8	2.2	121.1	14.1
Urban LSE (19)	259.3	9.4	38.3	14.5	22.9	9.9	32.1	21.8
Balanced diet ^a	410	40	30	100	75	60	100	—

^aICMR Report (1990).

Percentage of energy from fat was around 26–31% while that from protein was 9–11%. Median nutrient intakes for β -carotene as retinol equivalent and ascorbic acid were lower than the RDA (Table 4).

The intake of vitamin A as retinol equivalent showed a dose responsive behavior with plasma retinol (Figure 1a, Table 5, $P < 0.05$). However such a marked dose response was not seen in intake of ascorbic acid and plasma ascorbic acid (Figure 1b, Table 5, $P > 0.2$). Differences between rural population and urban HSE were highly significant in plasma retinol as well as in plasma ascorbic acid levels (Figures 2 and 3, $P = 0.0001$), but the differences between rural and semi urban sections were not significant nor those between different urban classes ($P > 0.1$).

Table 5 shows rank correlations between intakes of fruit, non-sweet fruit, green leafy vegetables, other vegetables and milk products with plasma levels of both the vitamins along with some of the socioeconomic characteristics. Intake of fruit, green leafy vegetables and milk products were significantly associated with plasma levels of retinol and ascorbic acid. Plasma ascorbic acid was more closely associated with non-sweet fruit intake. Age, education and income were strongly associated with fruit, vegetable intake and milk product consumption.

To investigate interrelationships of factors influencing plasma vitamin levels, multiple logistic regression analyses were carried out (Table 6). In the univariate analyses, age, BMI, sex, consumption of other vegetables and Hb, were found not to be statistically significant ($P > 0.25$). Hence in the multivariate model these factors were not included. Education level, environmental conditions, green leafy vegetables and milk product consumption were observed to be good predictors of plasma retinol, while education, non-sweet fruit consumption, and passive smoking were the influential factors for plasma ascorbic acid level. No interaction terms entered the model. By excluding smokers, the associations were the same, while exclusion of passive smokers reduced the coefficient of determination.

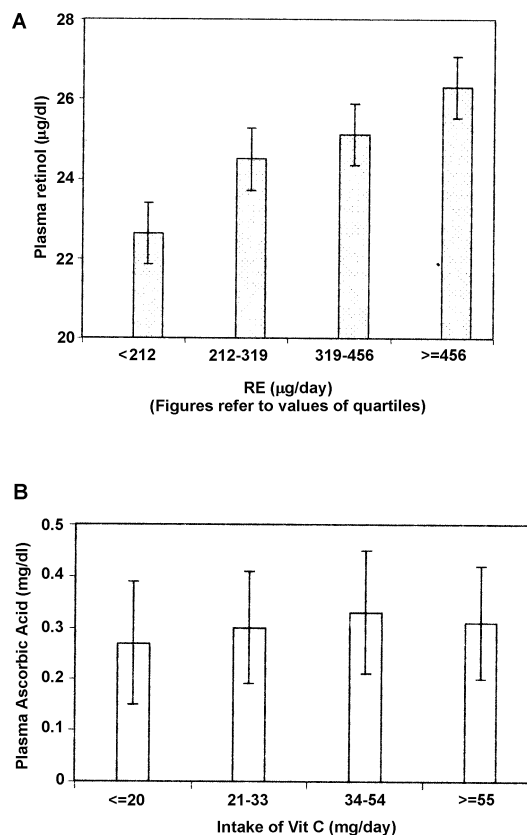


Figure 1 (A) Increase in plasma retinol with dietary intake. (B) Change in plasma ascorbic acid with dietary intake of vitamin C.

Discussion

The main aim of the present investigation was to model linkages between 19 dietary and non-dietary factors with plasma status of vitamin C and retinol. Although only a

Table 4 Median daily nutrient intake in men and women from different regions

Nutrient	Region				
	Rural	Semiurban	Urban HSE	Urban MSE	Urban LSE
Men					
Calories kJ (kcal)	8470 (2025)	8791 (2102)	10049 (2403)	11743 (2808)	9008 (2154)
Protein (g)	52.9	55.5	66.1	64.2	55.2
Fat (g)	58.7	60.4	82.5	92.1	63.9
β -carotene as retinol equivalent (μ g)	337	327	440	341	326
Vitamin C (mg)	36.1	36.4	42.2	58.6	33.6
Women					
Calories (kcal)	5840 (1396)	6716 (1606)	8226 (1967)	6926 (1656)	6637 (1611)
Protein (g)	36.1	42.3	50.0	41.7	40.3
Fat (g)	37.1	45.9	59.3	48.5	46.4
β -carotene as retinol equivalent (μ g)	179	254	386	391	265
Vitamin C (mg)	23.7	27.9	44.7	19.7	29.3

Table 5 Rank correlation among food intake (g/day) and some selected parameters

Variable	Plasma retinol ($\mu\text{g}/\text{dl}$)	Plasma ascorbic acid (mg/dl)	Age (y)	Education (level)	Per capita income (Rs)
Fruit	0.12*	0.17**	-0.20*	0.19**	0.14*
Non-sweet fruit	0.02 ^{NS}	0.30**	-0.10 ^{NS}	0.23***	0.13*
GLV	0.20***	0.10 ^{NS}	-0.12*	0.12*	0.11*
Other vegetables	0.11 ^{NS}	0.15*	-0.07 ^{NS}	0.24**	0.13*
Milk and milk products	0.19**	0.093 ^{NS}	-0.17**	0.44***	0.32***
RE intake	0.35**	0.07 ^{NS}	-0.04 ^{NS}	0.03 ^{NS}	0.26*
Vitamin C intake	0.06 ^{NS}	0.12 ^{NS}	0.02 ^{NS}	0.14 ^{NS}	0.29*

* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$; NS statistically not significant.

quarters of the study group was strictly vegetarian, the remaining population had very low and infrequent intakes of animal foods. The study population were consuming vegetarian foods for the majority of the days in the year.

Our data refers to 322 men and women covering major sections of the society, viz. rural, tribal, semi-urban, and urban with economically three broad classes, higher, middle and lower. The entire data were collected under identical experimental conditions by the same team of investigators. Apart from nutritional, anthropometrical and socioeconomic status, many new factors like passive smoking, and non-sweet fruit intakes were considered as variables in the data analyses. Most of the studies reporting associations of fruit and vegetable intakes refer to cancer patients, cardiovascular patients, smokers or alcoholics (Willett, 1998). Our study reports such associations in apparently healthy individuals and therefore adds valuable information not known in the Indian subcontinent. This is an essential addition to the world database and further help in defining dietary modifications required for such at-risk populations. Moreover the data represents individuals not taking any vitamin-mineral supplements, thus avoiding confounding effect on linkages observed between vitamin status and other factors. Considering the diverse environmental exposures, these data with varied dietary patterns are important for understanding interrelationships.

Consumption of cereals, legumes, fruit, vegetables and milk in the present study (Table 3) matches national sample data reported for various regions (NNMB, 1991) indicating better representation of the population in the study group. A high correlation of education, income and intake of milk products, fruit and vegetables was observed in our data (Table 5). Association of socioeconomic factors and consumption of fruit and vegetables has also been reported by studies in Western population (Hulshof *et al*, 1991; Smith & Baghurst, 1992; Johanson *et al*, 1997).

The design of the FFQ enabled us to extract more detailed information on any specific parameters of interest. For example, association of fruit consumption with plasma ascorbic acid level has been reported in Western populations either as individual fruit or total fruit intake (Matilainen *et al*, 1996). Our data on sour and non-sweet fruit consumption showed a strong association with the plasma ascorbic acid level but a weak association with total fruit consumption. Association of vegetable intakes and plasma vitamin C concentrations was found to be weak in the present study. Our results agree well with those reported by Drewnowski *et al* (1997). Secondly, our data permitted us to assess the impact

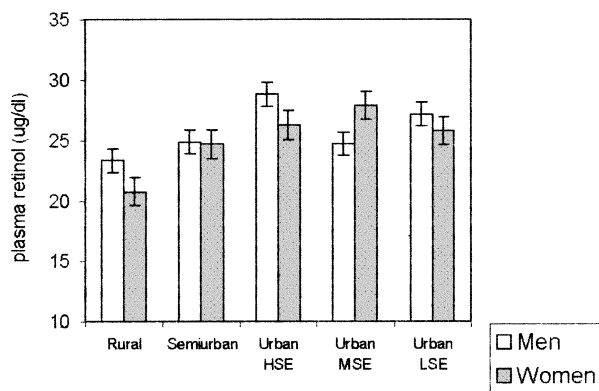


Figure 2 Plasma levels of retinol by different regions in men and women.

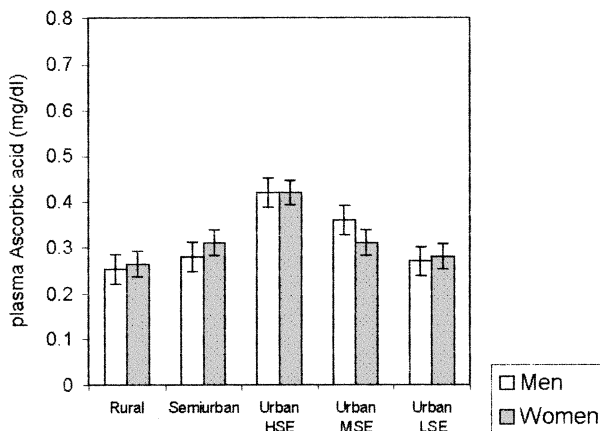


Figure 3 Plasma levels of ascorbic acid by different regions in men and women.

Table 6 Odds ratios for having plasma retinol concentration above median of the population ($\geq 25.53 \mu\text{g/dl}$) and plasma ascorbic acid concentration above median ($\geq 0.30 \text{ mg/dl}$)

Variable	Plasma retinol ($\mu\text{g/dl}$)		Plasma ascorbic acid (mg/dl)	
	Odds ratio	95% CI	Odds ratio	95% CI
Education (y)				
≤ 4	1.00		1.00	
5–10	0.87	(0.58, 1.30)	0.78	(0.50, 1.21)
11–15	0.98	(0.64, 1.52)	0.94	(0.59, 1.49)
≥ 15	2.38	(1.14, 4.98)	3.90	(1.70, 8.92)
P-value	0.004		0.003	
Environment				
Poor	1.00		—	—
Bad	0.67	(0.37, 1.20)		
Average	0.76	(0.93, 2.35)		
Fair	1.80	(0.45, 7.18)	NS	
P-value	0.10		NS	
Green leafy vegetables	1.01	(1.00, 1.03)	0.99	—
P-value	0.024		NS	
Non-sweet fruit	—	—	1.03	(1.01, 1.05)
P-value	NS		0.01	
Milk products	1.03	(1.00, 1.08)	—	—
P-value	0.021		NS	
Passive smoking				
No	—	—	1.00	
Yes	NS		0.68	(0.50, 0.94)
P-value			0.017	

of green leafy vegetables and other vegetables separately. Results indicated that green leafy vegetables have a larger impact than other vegetables on plasma retinol. The influence of vegetables, as a single food group, on plasma vitamin A has been reported by other studies (Drewnowski *et al*, 1997; Matilainen *et al*, 1996). Additionally milk consumption was also found to have a beneficial effect on plasma retinol. Considering the vegetarian eating habits in the Indian sub-continent and subnormal status of vitamin A, these results emphasize the greater need to increase consumption of green leafy vegetables and milk products.

The range of plasma vitamin C in our data was 0.2–0.6 mg/dl. Similar levels of plasma ascorbic acid have also been reported in the case of Russian men (0.21–0.57 mg/dl) and women (0.32–0.5 mg/dl; Spirichev *et al*, 1995). Mean plasma vitamin C levels in our data are 0.30 for men and 0.35 mg/dl for women, which is equivalent to 17–20 $\mu\text{mol/l}$ (Table 1). These are lower than the values in French adults (50 \pm 29 $\mu\text{mol/l}$ for 407 men and women; Drewnowski, 1997) or values of plasma vitamin C in south Asians residing in England (37.4 $\mu\text{mol/l}$ Ness *et al*, 1999). This may also be due to environmental pollution and absence of vitamin supplements. Further plasma ascorbic acid in women was reported to be significantly higher than in men (Matilainen *et al*, 1996). This is also true in our study, although not statistically significant. In one Indian study on 20 urban men (mean age 52 y), serum vitamin C was reported as 1.14 \pm 0.38 mg/dl

(Sinha & Sharma, 1998). However their intakes of vitamin C are very high (110 \pm 69 mg/day) as against our intakes of 33.6–58.6 mg/day (Table 4) and reported average vitamin C intakes (NNMB, 37 mg/day).

Our values of plasma retinol ranged from 12.8 to 38.3 $\mu\text{g/dl}$ in women and 12.3 to 40.8 $\mu\text{g/dl}$ in men, which are lower than the reported Western data (de Pee *et al*, 1998; Spirichev *et al*, 1995) and Indian data on university students (Vijayalakshmi & Rema, 1994). However our intakes of beta-carotene as retinol equivalent (179–440 $\mu\text{g/day}$ in men and women) are in agreement with the NNMB reports (294 $\mu\text{g/day}$ retinol). Mean retinol intakes reported by Vijayalakshmi *et al*, were higher than this range (785 $\mu\text{g/day}$). Our data on retinol intake and plasma retinol levels of urban and rural groups match well with those reported by Sankhla *et al*, (1991) on 30 urban and 30 rural men (18–50 y) from North India (Agra district).

A dose–response relationship between computed retinol equivalent intakes and plasma retinol (Figure 1) is in agreement with de Pee *et al* (1998). Since our calculation of vitamin A content was done on an individual food basis with our own laboratory estimates of nutrient contents, our values are more accurate than estimates based on a group level. Similar dose–response behaviour was not seen in the case of ascorbic acid. This may be because of the fact that vitamin A is fat-soluble and can be stored in the body. Therefore plasma vitamin A represents long-term status so

also do the FFQ estimates, while ascorbic acid, being water-soluble, has limited stores. Cooking losses in ascorbic acid can vary depending upon the variations in cooking procedures.

The multiple logistic regression model for the chance of having a plasma vitamin level above or below the median of the population was based on the actual consumption of different foods rather than computed nutrient intakes. This was felt to be necessary because of the fact that dietary intakes were computed from FFQ rather than the weighing method. Secondly, although nutrient contents of all recipes in FFQ were estimated in the laboratory, assumption of common recipes has to be made when converting FFQ data to nutrient intakes.

An interesting observation emerged from the study, that passive smoking had an adverse effect on plasma ascorbic acid level. The number of smokers was very small in the study population. However inquiry about any family member smoking revealed that passive smoking was a problem amongst the study group. There are a number of studies reporting the effect of smoking on blood parameters. However effects of passive smoking have been documented clearly in the present study. This emphasizes the need for public health measures to strictly maintain a non-smoking atmosphere in places of work and at home. The model envisaged while taking up the study was as shown in Figure 4.

Referring to the above model, our data has given leads to possible linkage between low plasma status of vitamin A and C and dietary and socioenvironmental factors in normal individuals. Thus the importance to dietary inadequacy needs to be reconsidered in the case of aetiology of vitamin A or C deficiency. This has important implications in arresting the process of developing disease.

Considering the difficulties in estimating the biochemical parameters in field studies, our results on logistic regression can be utilized as guidelines for identifying groups at risk of deficiency. For example, information on green leafy vegetable intake, milk product intake, education and environmental conditions of a group of individuals can predict risks of vitamin deficiency.

In summary, our study population is leaner and has considerably lower intakes of vitamin A and C and lower levels of vitamin C than Western populations. Subnormal status of both vitamin A and C in apparently healthy individuals emphasizes the need for increasing consumption of fruit, green leafy vegetables and milk products, and literacy and pollution-free environments as well.

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