Association of micronutrient status with subclinical health complaints in lactovegetarian adults

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Abstract

Background: Micronutrient malnutrition has many adverse effects on human health, not all of which may be clinically evident.
Objective: To explore linkages between subclinical health complaints and micronutrient status in lactovegetarian Indian adults.
Design: Health was assessed in 502 lactovegetarian adults (275 men, 227 women, aged 30.6 ± 9 years) using a structured questionnaire for existing complaints with severity of symptoms on a four-point scale and morbidity over the preceding month. Subjects were categorized as having no complaints (NC), or complaints of mild (MI) or moderate (MD) degree, using cluster analysis. Diet was recorded by a semi-quantitative food frequency questionnaire and nutrient intakes were computed using standard databases. Levels of haemoglobin, vitamin C, retinol, ceruloplasmin, riboflavin (EGRAC), folic acid, vitamin B_{12} and erythrocyte membrane zinc (RBCMZn) in blood were estimated.
Results: Health complaints of a mild and moderate degree were observed in 30.5% and 24.7% of the subjects, respectively. Average dietary intakes of β-carotene, riboflavin, iron and zinc were observed to be only about half of the recommended dietary intakes. There was a decreasing trend in micronutrient intakes from NC to MD. Intakes of iron, zinc, niacin and thiamin of men from the MI group tended to be lower than in the NC group ($p < 0.1$). Men from the MD group had significantly lower intakes of calcium, zinc and riboflavin than those from the NC group ($p < 0.05$). The intakes of these nutrients in women from NC, MI and MD were not significantly different. Multinomial logistic regression of health status revealed that plasma vitamin C and RBCMZn were negatively associated with MD and RBCMZn with MI.
Conclusions: The study indicates a need to increase micronutrient intakes of vegetarian populations, especially regarding vitamin C and zinc for maintenance of health.

Keywords: *health complaints*; *micronutrients*; *vitamin C*; *zinc*

Introduction

More than 2 billion people in the world today are estimated to be deficient in key vitamins and minerals, particularly vitamin A, iodine, iron and zinc (1). These deficiencies are likely to be prevalent in traditionally vegetarian populations such as Indians, since the micronutrient content of cereals (especially after milling), roots and tubers is low, so these foods typically provide only a small proportion of the daily requirements of most vitamins and minerals. Unless augmented with a variety of fruit and vegetables and taking measures to enhance bioavailability, these populations are at increased risk of several micronutrient deficiencies simultaneously. Dietary micronutrient intakes in

Indians have been observed to be much lower than the recommendations (2, 3).

Micronutrient malnutrition has many adverse effects on human health, not all of which are clinically evident (4). Current research has shown that apart from deficiency disorders, micronutrients play a crucial role as cofactors in the activity of almost all enzymes. Many antioxidants such as vitamin C and carotenoids have been reported as having health-protective effects (5, 6). Inadequate micronutrient intakes may reduce resistance to infections and adversely affect the immune system (7). Therefore, there is a need to estimate the magnitude of micronutrient deficiencies and assess their consequences on health (8). The present paper

attempts to explore linkages of common mild health complaints with micronutrient status in lactovegetarian Indian adults.

Materials and methods

Subjects

This cross-sectional study was conducted in a traditionally vegetarian population who consumed milk and milk products. Health surveys were undertaken through private hospitals, clinics and health camps in rural and urban communities in and around Pune city, Maharashtra state, India, in the same season. A total of 598 adults participated in the study voluntarily and gave informed written consent. Of these, 96 participants who had a major illness such as hypertension or diabetes mellitus were excluded from the study. Finally, 502 individuals (227 women and 275 men) were examined for their health and nutritional status.

Dietary nutrient intakes

A semi-quantitative food frequency questionnaire (FFQ) was used for evaluating habitual dietary intake based on 278 food items consumed throughout the year. The questionnaire was administered by trained investigators using standard measures and portion sizes. Details of the FFQ are given elsewhere (3). Average daily nutrient intakes, i.e. energy, protein, fat, riboflavin, folic acid, β-carotene, vitamin C, copper, zinc and iron, were computed from food consumption data of the FFQ using the nutritive value database of the authors' laboratory for a variety of cooked foods (9) and the nutritive value tables by Gopalan et al. (10) for raw foods.

Clinical assessment

With the help of a medical doctor a questionnaire was designed based on different health questionnaires for population survey. These involved self-reported complaints related to general health as well as complaints specifically related to, for example, bone, skin and the respiratory tract (11–13). This questionnaire was pretested on a pilot sample. Another questionnaire was pretested and used to record clinical signs of nutritional deficiencies (14). Clinical examination was carried out by a medical doctor for blood pressure, reflexes of limbs, eyes, chest and abdomen and existing complaints, if any, as reported when filling in the above-mentioned two questionnaires. Morbidity during the preceding 1

month was assessed as occurrence and duration of complaints related to upper respiratory tract infections (URT), eye, gastrointestinal system (GI), skin, bones and joints, central nervous system (CNS) and reproductive system. Complaints such as cough and cold, acidity, anorexia, eye strain, red eyes, photophobia, Bitot's spots, dryness of skin, skin pallor, skin pigmentation, delayed wound healing, muscle pain, bone and joint pain, irritability and nervousness were recorded with grading of severity on a four-point scale: absence of symptom $=0$, mild $=1$, moderate $=2$ and severe $=3$.

Biochemical measurements

Fasting venous blood (10 ml) was collected in the morning for each subject (not more than 12 h fasting). Laboratory estimations of plasma retinol, ascorbic acid, folic acid, erythrocyte glutathione reductase activity coefficient (EGRAC) and serum ceruloplasmin were carried out using the methods described in the National Institute of Nutrition manual (15), and erythrocyte membrane zinc (RBCMZn) was estimated as described by Ruz et al. (16).

Criteria used for identifying low micronutrient status were as follows: EGRAC >1.2, folic acid $<3.0\,\mu\mathrm{g\,l}^{-1}$, retinol $\leq 200\,\mu\mathrm{g\,l}^{-1}$, ascorbic acid $\leq 0.2\,\mathrm{mg\,l}^{-1}$, vitamin B_{12} $<160\,\mathrm{ng\,l}^{-1}$ (17); serum ceruloplasmin $<20\,\mathrm{mg\,dl}^{-1}$, serum iron $<600\,\mu\mathrm{g\,l}^{-1}$ (18); haemoglobin $<14\,\mathrm{g\,dl}^{-1}$ for men and $<12\,\mathrm{g\,dl}^{-1}$ for women (19).

Statistical methods

All statistical analyses were performed using SPSS for Windows, version 11.0. A K-means cluster analysis was used to identify relatively homogeneous groups of subjects defined from their degree of health complaints (20). The technique aims to group subjects in such a way that the distance between subjects within a group is minimized and the distance between the group centres is maximized. It does this using Euclidean distance measures in K-dimensional space, where K is the number of different variables considered. In other words, it aims to put similar subjects in the same group. The three-cluster solution had the best fit to the data. These three clusters were termed no complaints (NC), mild health complaints (MI) and moderate health complaints (MD), representing three levels of the variable health status. Characteristics of the subjects were analysed for

main effects of health risk and gender with interaction terms using a two-way anova (health status at three levels by gender at two levels). Gender differences were tested using anova. Differences between nutrient intakes in normal (no complaints) versus health complaint groups were tested using the generalized linear models (GLM) univariate procedure with gender and health status as factors. Health status was defined as a three-category variable (NC, MI and MD) with age- and gender-matched individuals in all three groups. To investigate associations of health complaints with biochemical indices of micronutrient status and to judge the risk of suboptimal health, multinomial logistic regression was performed after adjusting for age and weight. Health status of subjects was taken as the dependent variable, taking a value of 0 for MD, 1 for MI and 2 for NC, and the blood levels of micronutrients and lipids were taken as the set of predictor variables.

Results

General characteristics of the subjects in the three groups are given in Table 1. Differences between body mass index (BMI) or waist to hip circumference ratio (WHR) in different health groups were not statistically significant. Blood pressure was within the normal range in all subjects. There were no significant differences in blood pressure between

Table 1. Characteristics of the subjects

Parameter	No complaints (NC)	Mild complaints (MI)	Moderate complaints (MD)
Men ($n = 275$)			
n	127	93	55
Age	29.7 ± 1.1	29.7 ± 1.0	30.6 ± 1.3
BMI	20.5 ± 0.5	19.8 ± 0.5	21.6 ± 0.6
WHR	0.88 ± 0.02	0.88 ± 0.04	0.87 ± 0.02
SBP	121 ± 2	115 ± 2	123 ± 2
DBP	80 ± 2	77 ± 1	80 ± 2
Women ($n = 227$)			
n	97	61	69
Age	31.6 ± 1.6	32.8 ± 1.2	31.1 ± 1.2
BMI	20.7 ± 0.8	21.1 ± 0.8	22.1 ± 1.0
WHR	0.84 ± 0.02	0.85 ± 0.02	0.86 ± 0.05
SBP	114 ± 2	112 ± 2	113 ± 5
DBP	74 ± 2	74 ± 1	73 ± 2

Data are shown as mean $\pm$ SE.

BMI: body mass index; WHR: waist to hip ratio; SBP: systolic blood pressure;

different groups and also no significant differences between men and women.

Bone and joint pain were reported by 27.5%, URT trouble by 39.6%, eye complaints by 45.2%, GI complaints by 44.5%, skin complaints by 34.1% and CNS problems by 38.9% of the individuals. Gynaecological complaints were reported by 21.2% of the women.

In men, average daily intake of cereals, legumes, vegetables, fruit, milk and milk products was 478 ± 192 g (mean $\pm$ SD), 148 ± 109 g, 62 ± 32 g, 50 ± 26 g and 101 ± 42 g, respectively. In women, these intakes were 320 ± 107 g, 122 ± 69 g, 71 ± 43 g, 63 ± 38 g and 65 ± 33 g, respectively. Average daily energy intake by men was 2192 ± 100 kcal and that by women 1678 ± 73 kcal (Table 2). These were close to the respective recommended dietary intakes (RDIs) for Indians (21). There were no significant differences in macronutrient intakes or polyunsaturated fatty acids (PUFA) intakes among the NC and MI, MD groups, although men's intakes were higher than those of women ($p < 0.05$).

Mean daily intakes of folic acid, copper, calcium and thiamin met the RDI in all subjects. However, average iron intakes in men (18.7 ± 1.5 mg day^{-1}) and women (13.5 ± 1.0 mg day^{-1}) were much lower than the RDI of 28 mg day^{-1} for Indians (21). Similarly, average zinc intakes of men and women (7.4 ± 0.16 and 5.2 ± 0.14 mg day^{-1}, respectively) were also lower than the RDI of 15 mg day^{-1}. Mean intakes of β-carotene (1425 ± 52 and 1356 ± 68 µg day^{-1} in men and women, respectively) were lower than the RDI. Although there was a decreasing trend in micronutrient intakes from NC to MD (Table 2), differences in NC, MI and MD among women were not statistically significant. Intakes of iron, zinc, niacin and thiamin in men were also similar in the different groups. Intakes of calcium, zinc, riboflavin and vitamin C in men from the MD group were significantly lower than in the NC group ($p < 0.05$).

The erythrocyte sedimentation rate (ESR) was within the normal range for all subjects, indicating the absence of chronic infection or inflammation (Table 3). Glucose levels and total cholesterol were also within the normal range for all subjects. Triglycerides were slightly high, between 150 and 175 mg dl^{-1} for three women and two men from the MI group, and two women and two men from the MD group. A significant difference was observed in

Table 2. Average daily nutrient intakes of the subjects

Nutrient	RDI[a]	NC	MI	MD	5th percentile	95th percentile
Men						
Energy (kcal)	2425	2322±96	2134±98	2118±105	1121	3630
Protein (g)	60	53.8±2.4	49.2±2.2	48.3±3.2	23.5	88.9
Total fat (g)	20	69.8±3.4	66.9±3.7	67.5±4.2	32.1	128.4
PUFA[b] (g)	3% of energy	39.1±2.0	39.5±2.1	41.5±5.5	17.9	63.7
Carbohydrates (g)	–	368±15	371±12	354±16	170	528
Calcium (mg)	400	561±37	583±42	476*±74	191	1035
Iron (mg)	28	20.4±1.2	17.6†±0.9	18.1±2.5	6.3	32.8
Copper (mg)	2	2.4±0.1	2.2±0.1	2.2±0.2	1.04	3.97
Zinc (mg)	15	8.1±0.4	7.2†±0.3	6.2*±0.8	3.2	12.4
β-Carotene (μg)	2400	1418±130	1322±121	1263±201	340	3298
Thiamin (μg)	1200	1476±76	1298†±63	1346±180	516	2373
Riboflavin (μg)	1400	797±37	744±38	709*±87	316	1275
Niacin (mg)	16	15.8±0.8	13.6†±0.7	13.8±1.7	5.5	23.9
Folic acid (μg)	100	159±9	148±7	147±17	72.3	256.6
Vitamin C (mg)	40	52±8	41±5	34*±6	8.1	107.1
Women						
Energy (kcal)	1875	1712±70	1676±62	1644±86	870	2598
Protein (g)	50	37.3±2.1	36.7±1.8	36.9±1.5	19.3	58.2
Total fat (g)	20	60.1±3.6	57.8±3.3	56.2±3.7	22.1	96.6
PUFA (g)	3% of energy	29.4±1.6	29.2±2.5	28.1±2.2	12.9	49.0
Carbohydrates (g)	–	242±14	236±13	228±10	123	378
Calcium (mg)	400	486±32	453±46	462±44	192	792
Iron	30	14.2±0.7	13.4±1.2	12.8±1.1	6.1	23.8
Copper (mg)	2	1.6±0.07	1.6±0.12	1.5±0.11	0.8	2.6
Zinc	15	5.4±0.3	5.2±0.4	4.2±0.4	2.6	8.3
β-Carotene (μg)	2400	1561±143	1508±224	1316±209	306	3369
Thiamin (μg)	900	991±44	960±86	928±71	422	1656
Riboflavin (μg)	1100	606±30	598±50	556±43	283	967
Niacin (mg)	12	10.6±0.5	10.0±0.8	9.6±0.7	4.8	16.3
Folic acid (μg)	100	121±7	118±11	107±9	51.3	196
Vitamin C (mg)	40	72±17	48±18	46±10	6.3	150.6

Data are shown as mean±SE.

[a]Recommended dietary intakes (RDI) for Indians (21).

PUFA: polyunsaturated fatty acids.

*$p < 0.05$, †$p < 0.1$.

triglycerides between the MD group and the NC group ($p = 0.015$).

Haemoglobin was marginally below the normal range in 25% of the men and 36.4% of the women. Mean plasma vitamin C and RBCMZn were lower in the MD group than in the NC group ($p < 0.05$) (Table 3). No other micronutrient levels were found to be significantly different between the groups ($p > 0.1$). However, the prevalence of low micronutrient status was higher in both men and women in the MD group than in the corresponding NC group (Fig. 1). Low levels of plasma vitamin C (< 0.2 mg dl^{-1}) were seen in 16% of the NC group, 26% of the MI group and 35% of the MD group. Low plasma retinol levels (< 20 μg dl^{-1}) were found more often in women (14–21%) than in men (6–12%; $p < 0.05$). The prevalence of low serum ceruloplasmin (< 20 mg dl^{-1}) was the same in men and women (16–30%). Around 29% of men and 48% of women had low RBCMZn (< 0.45 mg dl^{-1}). Low riboflavin status (EGRAC > 1.2) was observed in 40.5% of men and 52.1% of women, with no significant differences between groups. The prevalence of low plasma levels of vitamin B$_{12}$ (< 160 ng l^{-1}) and folic acid (< 3 μg l^{-1}) was in the order of 2–4.5% in all groups, and in both men and women.

To study associations between low micronutrient status and health complaints, correlations of individual complaints with micronutrient levels

Table 3. Blood levels of haemoglobin, cholesterol, high-density lipoprotein (HDL), glucose, triglycerides and blood micronutrients, and erythrocyte sedimentation rate (ESR), in men and women

Parameter	Normal range (17–19)	NC	MI	MD	5th percentile	95th percentile
Men						
Haemoglobin (g dl^{-1})	14–16	14.8±0.2	14.7±0.2	14.8±0.2	12.8	17
ESR (mm h^{-1})	1–13	11±1	13±1	10±1	3.3	25.0
Total cholesterol (mg dl^{-1})	<200	164±4	160±5	170±5	118	214
HDL (mg dl^{-1})	40–60	45±1	46±3	48±2	26.5	61.7
Glucose (mg dl^{-1})	70–110	93±4	95±4	94±5	53	108
Triglycerides (mg dl^{-1})	<150	91±5	99±4	97*±4	43	143
Plasma vitamin C (mg l^{-1})	>0.2	0.31±0.02	0.26±0.02	0.25*±0.02	0.12	0.44
Plasma retinol (μg dl^{-1})	>20	25.6±0.9	25.0±0.6	25.1±0.5	19.0	31.3
Serum ceruloplasmin (mg l^{-1})	>20	20.2±0.7	21.3±0.8	19.7±1.0	12.2	30.1
RBCMZn (μmol g^{-1} protein)	<0.45	0.54±0.01	0.46±0.01	0.44*±0.02	0.35	0.61
EGRAC	<1.2	1.12±0.03	1.13±0.02	1.1±0.02	0.83	1.6
Plasma folic acid (μg l^{-1})	>3.0	4.6±0.1	4.3±0.1	4.5±0.1	3.1	5.9
Serum B$_{12}$ (μg l^{-1})	>160	434±12	438±10	426±13	285	614
Women						
Haemoglobin (g dl^{-1})	12–14	12.0±0.3	12.1±0.2	12.1±0.2	8.7	14.5
ESR (mm h^{-1})	1–20	20±2	24±2	25±3	7	46
Total cholesterol (mg dl^{-1})	<200	163±4	159±4	171±6	113	205
HDL (mg dl^{-1})	40–60	50±2	52±2	47±2	30	75
Glucose (mg dl^{-1})	70–110	78±2	81±3	87±4	54	106
Triglycerides (mg dl^{-1})	<150	71±3	81±5	89*±6	45	131
Plasma vitamin C (mg l^{-1})	>0.2	0.32±0.02	0.30±0.02	0.22*±0.01	0.13	0.49
Plasma retinol (μg dl^{-1})	>20	24.5±0.7	24.2±0.6	24.0±1.0	16.7	30.7
Serum ceruloplasmin (mg l^{-1})	>20	23.1±1.2	22.5±1.1	22.9±0.7	13.4	43.3
RBCMZn (μmol g^{-1} protein)	<0.45	0.52±0.01	0.49±0.02	0.43*±0.01	0.30	0.60
EGRAC	<1.2	1.2±0.02	1.22±0.01	1.24±0.02	1.0	1.6
Plasma folic acid (μg l^{-1})	>3.0	4.4±0.2	4.1±0.1	3.8±0.1	3.2	5.5
Serum B$_{12}$ (μg l^{-1})	>160	424±11	413±11	388±13	276	543

Data are shown as mean±SE.

RBCMZn: erythrocyte membrane zinc; EGRAC: erythrocyte glutathione reductase activity coefficient.

*$p < 0.05$.

were first examined. Plasma vitamin C exhibited a significant negative correlation with almost all individual complaints ($p < 0.05$). RBCMZn, retinol, haemoglobin and riboflavin also showed an association with health complaints (Table 4). For other micronutrient levels, i.e. vitamin B$_{12}$, folic acid and ceruloplasmin, correlations were very low and not statistically significant.

To estimate the relative influences of these micronutrients on health status, a multinomial logistic regression was performed after accounting for differences in age and weight. The dependent variable health status had three categories: NC, MI and MD. Blood levels of eight micronutrients, triglycerides and cholesterol were the 10 independent variables. Comparison of the NC group with the MI group showed that RBCMZn had a significant negative association with health status ($p = 0.001$) (Table 5). When the MD group was compared with the NC group, RBCMZn and serum vitamin C were negatively related factors ($p < 0.01$).

Discussion

In this study population of apparently healthy individuals, subclinical health complaints of mild degree were reported by 30.5% of the subjects and of moderate degree by 24.7%. Although the assessment of subclinical health complaints may involve subjective bias, care was taken to reduce this bias in self-reporting by using standard techniques and examination by the doctor. Moreover, such health status measures have been reported to provide reliable data (11).

Regarding macronutrients, mean fat intakes were higher than the RDI in both men and women in all three health status groups. Similar high fat intakes, especially higher saturated fat intakes, have been reported previously in Indian adults (3, 22, 23).
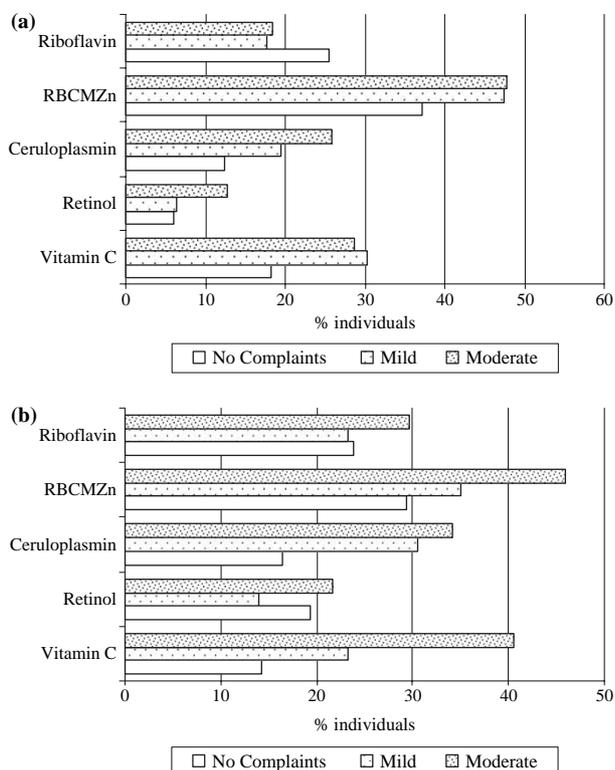
Fig. 1. Percentage prevalence of low blood levels of micronutrients in (a) men and (b) women. RBCMZn: erythrocyte membrane zinc.

The invisible fats present in many Indian foods have a high linoleic acid content (rice 50%, wheat 55%, Bengalgram 65%, redgram 58%) and linolenic acid content of about 3%. As a result, the average Indian diet, which is predominantly based on cereals and some legumes, contains sufficient linoleic and linolenic acid (21). Together with cooking oils, this may be the reason for the high PUFA intakes of the study groups. Studies on tissue levels of long-chain *n*-3 fatty acids show that these are depressed in vegetarians, particularly in vegans (24). However, a study of normal healthy Indians indicated blood levels of essential fatty acids to be

satisfactory (25). Fatty acid levels in blood were not measured in the present study, but no significant differences were seen in PUFA intakes between the three health status groups.

The results showed that mean daily iron intakes were only about half of the RDI. Furthermore, 74.4% of adults had zinc intakes less than half the RDI, 52% had low β-carotene intakes, 44% had low riboflavin intakes and 34% had low vitamin C intakes. Low micronutrient intakes have also been reported by other Indian studies (2) and in Western populations (26, 27). The present study group was lactovegetarian, with low consumption of vegetables (66 ± 45 g day^{-1}) and fruit (66 ± 45 g day^{-1}), and also of milk and milk products (86 ± 67 g day^{-1}). This may be the reason for their low intakes of vitamins and minerals. Similar low consumption of fruit and vegetables has been found to result in low vitamin C intakes (27).

Intake of animal foods in the study group was restricted only to milk products. This could be one of the factors behind low micronutrient status, especially regarding iron (28). Iron and zinc deficiencies have been reported in the developing as well as the developed world (29, 30). For instance, lifelong vegetarians in Belgium were also found to have deficiencies of calcium, zinc, iron, and vitamins B$_6$ and B$_{12}$ (30). There is a real risk of vegan populations becoming deficient unless they change their eating habits or take a supplement (31). However, micronutrient status can be enhanced by increasing the intake of vegetables and fruit, and by improving the bioavailability of micronutrients by appropriate methods such as sprouting or fermentation (32, 33).

There was a decreasing trend in micronutrient intakes from NC to MD. This was reflected in the corresponding blood levels. Even in the NC group,

Table 4. Correlations between individual health complaints and blood levels of micronutrients

Complaints	Plasma vitamin C	RBCMZn	Plasma retinol	Haemoglobin	Riboflavin (EGRAC)
Bones and joints	−0.23 ($p=0.001$)	−0.10 ($p=0.09$)	0.045 ($p=0.49$)	0.09 ($p=0.16$)	0.07 ($p=0.33$)
URT	−0.12 ($p=0.053$)	−0.08 ($p=0.24$)	0.131 ($p=0.03$)	(0.01) ($p=0.9$)	0.12 ($p=0.06$)
Eye	−0.145 ($p=0.012$)	−0.11 ($p=0.07$)	0.08 ($p=0.17$)	−0.07 ($p=0.24$)	0.14 ($p=0.018$)
GI	−0.16 ($p=0.006$)	−0.12 ($p=0.049$)	0.013 ($p=0.82$)	−0.03 ($p=0.61$)	0.08 ($p=0.15$)
Skin	−0.12 ($p=0.048$)	−0.07 ($p=0.32$)	0.10 ($p=0.13$)	−0.11 ($p=0.08$)	0.01 ($p=0.89$)
CNS	−0.091 ($p=0.118$)	−0.08 ($p=0.21$)	0.082 ($p=0.17$)	–	0.08 ($p=0.17$)
Total health complaints	−0.17 ($p=0.017$)	−0.155 ($p=0.034$)	0.054 ($p=0.45$)	−0.04 ($p=0.53$)	0.15 ($p=0.03$)

RBCMZn: erythrocyte membrane zinc; EGRAC: erythrocyte glutathione reductase activity coefficient; URT: upper respiratory tract; GI: gastrointestinal; CNS: central nervous system.

Table 5. Maximum likelihood fit of the multinomial logistic model

Variable	Estimate of β-values	
	Moderate (MD) vs normal (NC)	Mild (MI) vs normal (NC)
Intercept	0.523	1.98
Haemoglobin	0.012	−0.032
Plasma vitamin C	−4.292**	−0.154
Plasma retinol	0.057	0.01
Plasma folic acid	0.049	0.037
Serum ceruloplasmin	−0.022	0.002
Serum total cholesterol	0.01	−0.001
Triglycerides	0.005	0.007
RBCMZn	−5.849**	−4.072**
EGRAC	0.471	−0.611
Vitamin B$_{12}$	−0.002	0.001

RBCMZn: erythrocyte membrane zinc; EGRAC: erythrocyte glutathione reductase activity coefficient.

**$p < 0.01$.

14% of subjects had low serum levels of various micronutrients, and the prevalence increased to 35% in the MI group and 47% in the MD group. Further examination of the data by multinomial logistic regression analysis revealed that low levels of vitamin C and zinc were risk indicators for subclinical health complaints. The association of low zinc status, as indicated by low RBCMZn, with health complaints suggests a need for improving bioavailable zinc intakes.

These results on young adults agree well with many studies on older age groups reporting an association between micronutrient deficiencies and health disorders. Low serum micronutrient concentrations in disabled older women were found to be an independent risk factor for frailty, and the risk of frailty increased with the number of micronutrient deficiencies (34). Zinc has been found to be one of the significant predictors of immune function in older women (35).

Evidence from epidemiological studies supports the importance of an optimal micronutrient status to help to prevent chronic diseases such as coronary heart disease, cancer, mental degeneration, diabetes and age-related macular degeneration. Antioxidant micronutrients such as vitamin C or β-carotene may be particularly important (5, 36, 37). Increased fruit and vegetable consumption has been associated with a lower prevalence of coronary heart disease and some cancers, although reductions are sometimes modest and not statistically significant (38).

The results demonstrated associations between low zinc and vitamin C status and common health complaints in young adults, indicating the need for alleviating micronutrient malnutrition. One of the limitations of this study, however, is the relatively small sample size. Further studies are necessary to confirm the results. Another limitation is that the clusters identified in this paper apply specifically to the clinical signs of nutritional deficiencies and general health complaints and should not be extrapolated to the entire spectrum of health disorders. In conclusion, however, this study highlights the importance of increasing the dietary intake of micronutrients, especially vitamin C and zinc, for the maintenance of good health.

Acknowledgements

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