Original Article

Dietary vitamin A may be a cardiovascular risk factor in a Saudi population

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Traditional risk factors do not appear to explain fully the variation in the incidence of the cardiovascular diseases (CVD). Epidemiological studies have not been entirely consistent with regard to the relationship between antioxidant vitamin intake and CVD and there appears to be little data on this relationship in non-Caucasian populations. This study aimed to investigate the dietary intake of vitamin A, C, and vitamin E, and carotenoids, serum concentrations of vitamin E and A and indices of lipid peroxidation were measured in male Saudi patients with established CVD and age-matched controls. We assessed the dietary intakes of vitamins A, C, and E and carotenoids, by a food frequency questionnaire. Serum vitamins A and E concentrations were measured by HPLC, in 130 Saudi male subjects with established CVD, and 130 age-matched controls. We also determined serum lipid profiles (total cholesterol, triglycerides, HDL-C, LDL-C), lipoprotein (a), oxidized LDL, and serum lipid peroxide concentrations. Diabetes mellitus (P<0.0001), a positive smoking habit (P<0.0001) and hypertension (P<0.05) were more prevalent among CVD patients. Levels of dietary vitamin E and A were also significantly higher among cases. In conditional logistic regression analysis, the most significant characteristics differentiating CVD patients from controls were diabetes mellitus (Odds ratio 2.49, CI 1.42-4.37, P<0.001), total fat intake (Odds ratio 1.02, CI 1.01-1.03, P<0.01), serum vitamin A (Odds ratio 0.72, CI 0.53-0.99, P<0.05), and the vitamin A/total fat intake ratio (Odds ratio 1.04, CI 1.01-1.06, P<0.01). In a Saudi population, smoking habit and hypertension were significantly more common among patients with CVD. Multivariate analysis showed that dietary total fat and vitamin A and the presence of diabetes mellitus were independent coronary risk factors. This is the first report of a potentially deleterious effect of dietary vitamin A in a non-Caucasian population. However it is possible that unidentified residual confounding factors may account for this finding.

Key Words: vitamins A, C, and E, coronary disease, risk factors, lipid peroxides, dietary antioxidant vitamin intake, oxidative stress, coronary atherosclerosis, Saudi Arabia, Jeddah.

Introduction

Traditional coronary risk factors do not appear to explain fully the variation in the incidence of the cardiovascular disease in all populations and the search for other risk factors continues.[1] There are several strands of evidence supporting the hypothesis that oxidation of LDL-C has a role in the etiology of atherosclerosis and that antioxidant status may be an important determinant of cardiovascular disease (reviewed by[2,3]). The major dietary sources of antioxidants are fruits and vegetables. Vitamin A has an antioxidant activity against the thiyl radical whilst its precursor, β-carotene is a multifunctional lipid soluble antioxidant capable of physiologically quenching singlet oxygen and inhibiting free radical chain reactions.[4]

Vitamin E may contribute to the prevention of atherosclerosis by inhibiting LDL oxidation via its reaction with lipid peroxy-radicals, thereby, suppressing lipid peroxidation before conjugated dienes are formed.[5] Vitamin C may exert an effect on redox cycling of vitamin E within lipoproteins and membranes.[6] Moreover, this interaction may involve the putative vitamin C sparing effect of vitamin E.[7] Epidemiological studies have suggested that antioxidant supplements, specifically vitamin E, can prevent coronary events.[8-10] The ability of antioxidant vitamins to inhibit the formation of modified LDL, potentially slowing the rate of atherogenesis, has been tested in animal and human intervention studies using antioxidant vitamins[11-14] and other

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antioxidant compounds such as probucol and BHT.[15-17]

Vitamins E and C, and β-carotene, either singly or combination have been investigated extensively in coronary prevention trials. The results of these trials have been inconsistent.[18-20] The discrepancies may be explained by differences in the inclusion criteria, the antioxidant content of the basal diet of the sample population under investigation, and the dose composition of antioxidant supplements used. Although the prevalence of coronary heart disease is increasing in Saudi Arabia, there is little data on antioxidant vitamin status in the Saudi population[21,22] and in particular in patients with CVD.

The aim of this study was to investigate the relationship between: self-reported dietary intake of the antioxidant vitamins A, C, E, and the carotenoids; serum vitamin E and A concentrations; and indices of lipid peroxidation in Saudi males with established CVD and a population of age-matched subjects without CVD.

Materials and methods

Subjects

Individuals with established CVD were identified by positive findings on angiography (*N*=66), or by a past medical history of myocardial infarction or angina (*N*=64). They were recruited from The King Fahad Armed Forces Hospital and The King Abdulaziz Univer-sity Hospital, Jeddah, Kingdom of Saudi Arabia, respectively. Patients were classified according to the number of stenosed (defined as >50% occlusion of luminal diameter), or occluded vessels (1 to 3 vessel disease). The severity of angiographically-defined disease was scored from 0 (no CHD) to 3 (stenosis >50% in 3 vessels) as previously described.[23]

A diagnosis of a myocardial infarction was made in accordance with Joint European Society of Cardiology/ American College of Cardiology Committee criteria[24]: chest pain with elevated cardiac enzymes, and characteristic ECG changes. None of the patients had suffered a heart attack within 6 months of recruitment into the study. Age-matched individuals, who were at risk for developing coronary artery disease, but were without the disease, served as controls and were recruited from both hospitals. Identification of controls was made by a cardiologist in each hospital. They were usually referred for risk factor modification, principally hyperlipidemia, hypertension, and diabetes mellitus. The drawing of controls from the same patient pool is a proper setting that enhances the compatibility of coronary risk estimation in cases and controls. Subjects with established renal or hepatic disease were excluded, as were those on treatment with antioxidants. Fasting blood and urine samples were obtained from all subjects. The ethical committee of each hospital approved the study.

Demographic characteristics

Cardiovascular risk factors including: age, height, weight, body mass index (BMI), systolic and diastolic blood pressure, physical activity, smoking status, history of hypercholesterolemia, diabetes, family history of heart diseases (first-degree relative with myocardial infarction or cardiac death before age 55) were assessed for each subject. Hypertension was defined as a systolic blood pressure above 140mmHg, and or a diastolic blood pressure above 90mmHg, or current use of antihypertensive therapy.[25] BMI was calculated as weight (kg)/ height (in metres).[2] Normal weight was defined as a BMI <25, overweight as a BMI 25-29.9, and obese as >30 kg/m^2.[26] Dyslipidaemia was defined as total cholesterol level ≥ 5.2 mmol/L, LDL-C ≥ 3.36 mmol/L, and/or HDL-C< 1.04 mmol/L.[27]

Diabetes was defined as a history of diabetes, with a documented, or measured fasting glucose >7 mmol/L, or treatment with either insulin or oral hypoglycaemic agents.[28] With regard to smoking habits the following categories were used: nonsmokers, former smokers, and current smokers. Current smokers were further categorized into those who smoked <20 cigarettes/day and those who smoked ≥20 cigarettes/day. Physical activity was graded by the participant according to the number of episodes of exercise undertaken per week and were categorized as active (≥3 times/week) or inactive (<3 times/ week) according to the recommendations of the American Heart Association consensus statement on primary prevention of coronary diseases and from the USA Surgeon General's report .[29]

Laboratory methods

Triglycerides and total cholesterol were measured enzymatically by colorimetric methods. HDL-cholesterol was measured similarly after precipitation of the non HDL-C fraction with phosphotungstate magnesium. LDL-cholesterol was calculated using the Friedwald formula.[30]

Serum lipid peroxides were measured by the ferrous oxidation xylenol orange (FOX) assay[31] or thiobarbituric acid reactive substances (TBARS).[32] Serum lipoprotein(a) levels were measured using an ELISA assay kit (Biopool, CA, USA). Serum oxidized-LDL levels were measured using a competitive ELISA kit (DRG Diagnostics, Germany). The intra-assay and inter-assay coefficients of variation for each variable was found to be <8% and <15% respectively.

Serum antioxidant vitamins analysis

Serum vitamin A (measured as retinol) and E (measured as α-tocopherol) were determined simultaneously using isocratic reverse phased high performance liquid chromatography (HPLC) with multi-wave length detection[33] as a modification of the method Bieri *et al.*[34] Briefly, 200µl of internal standard (10µg/ml δ-tocopherol in isopropanol) was added to 200µl of serum and vortex mixed. Aqueous ammonium sulphate (3.9mol/L, 200µl) was added and again vortex mixed. After centrifugation at 1000g for 5 min, 25µl of the supernatant was injected into a stainless steel (150 x 4.6mm) Prodigy 5-µm ODS2 column (Phenomenex Ltd, Macclesfield, Cheshire, UK), using methanol as the mobile phase and a flow rate of 1.40 ml/min. Peaks were detected at 294nm and 325nm for vitamin E and A respectively. Retention time for internal standard and α-tocopherol was 5.2, and 6.6min respectively and 2.6 min for vitamin A. Standards and quality control material were obtained from BioRad Laboratories Ltd, Hemel Hempstead, UK. Intra-assay and inter-assay variations (CV%) for vitamin A were 4.6% and 6.2% respectively and for vitamin E were 3.2% and 4.2% respectively.

Assessment of dietary intake

Dietary intake over the previous year was estimated using a food frequency questionnaire (FFQ). This approach for estimating nutrient intake has been used in epidemiological studies in the West.[35] The FFQ contained questions about frequency of intake of ninety-four food items and beverages in the past year.[36] Foods and beverages were expressed in serving sizes (grams), household measures (cup, spoon) or natural units (slice of bread, apple). Food items were classified into eleven categories: grains (5 items), bread (5 items), fruits (11 items), vegetables (19 items), legumes (4 items), nuts (4 items), meats, poultry and fish (12 items), dairy products (10 items), fat and oil (3 items), soft drinks (6 items), miscellaneous (12 items) and junk food (3 items). Subjects were asked to select either per day, per week, per month or almost never as a denominator and then to enter frequency of use. Serum levels of vitamin A and E were used to validate micronutrient intake using this questionnaire. The nutrient database used was based on UK food composition tables[37] together with food composition tables for use in East Asia and the United States handbook of food composition.[38] The composition of traditional local foods, not included in the above tables, was derived from another local study.[36] The estimated dietary intake of all nutrients was calculated in terms of percentage recommended nutritional intake (%RNI) for UK adults for each individual, as there are no published data on recommendations for a Saudi population. The most recent version of the United Kingdom Dietary Recommended Values (DRVs)[39] was used to standardize the pattern of nutrient intake except for vitamin E intake which was taken from the most recent US recommended dietary intake.[40]

Statistical analysis

All data are presented as mean and standard deviation for normally distributed parameters or as median and intra quartile range for non-normal distributed parameters. Variables that showed a skewed distribution were log transformed before analyses and then back transformed to their natural units for presentation. Comparison of numeric data was performed using unpaired t-tests for normally distributed variables and by using the Mann Whitney-U test for non-normally distributed parameters. A χ^2 test was used for comparison of categoric data. Serum levels of the antioxidant vitamins were corrected for cholesterol levels as suggested by Jordan *et al.*[41] Associations between dietary intake of antioxidant vitamins and their serum levels were tested using Pearson and/or Spearman's correlation coefficients when appropriate. Partial Pearson correlation was also used to test the association between serum and dietary vitamins levels after taking into account potential confounding factors. Conditional logistic regression analysis was used to determine the magnitude of associations between antioxidant vitamins status and atherosclerosis. The dependent variable was a binary variable with a 0 value representing those with no CVD and a 1 value representing CVD patients. The Wald statistic was calculated to assess the significance of individual logistic regression co-efficients for each independent variable. A P value <0.05 was considered of statistical significant. All statistical analysis was performed using SPSS (version 11.5) soft-ware.

Results

Demographic data

Table 1 shows the distribution of demographic and predisposing coronary risk factors in the study population. Overall, there was an increased prevalence of diabetes mellitus (P<0.0001) and hypertension (P<0.05) in the CVD patients. As might have been expected, positive smoking habit was more common among the CVD patients (P<0.0001).

Table 1. Demographic characteristics of patients with CHD and controls.

	Controls (N = 130)	Cases (N =130)	***P***
Age (years) Mean ± SD	55.6 ± 12.1	55 ± 11.6	NS
Systolic BP (mm Hg) Mean ± SD	128 ± 19.3	127.5 ± 22.5	NS
Diastolic BP (mm Hg) Mean ± SD	79.9 ± 10.4	77.6 ± 11.8	NS
Weight (kg) Mean ± SD	80.8 ± 15.4	81.1 ± 15.8	NS
Height (cm) Mean ± SD	168.6 ± 8.3	166 ± 6.3	<0.05
BMI (Kg/m2) Mean ± SD	28.4 ± 4.9	29.4 ± 5.1	NS
Family history n (%)			
Diabetes	61 (47)	70 (54)	NS
Heart diseases	32 (25)	34 (26)	
Smoking status n (%)			
Never	66 (51)	43 (33)	<0.001
Former	33 (25)	61 (47)	
Current (<20 cigarette)	11 (9)	14 (11)	
Current (≥20 cigarette)	20 (15)	12 (9)	
Body mass index n (%)			
Normal	33 (25)	20 (15)	NS
Overweight	51 (39)	64 (49)	
Obese	46 (35)	46 (35)	
Physical activity status n (%)	38 (29)	34 (26)	NS
Non-active	92 (71)	96 (74)	
Active			
Dyslipidemia n (%)	122 (94)	124 (95)	NS
Diabetes mellitus n (%)	47 (36)	78 (60)	<0.0001
Hypertension n (%)	68 (52)	85 (65)	<0.05
On a diet n (%)	38 (29)	51 (39)	NS

Biochemical data

Table 2 summarizes the biochemical measurements among controls and cases. Lipid profiles were similar for the groups, although serum triglycerides were significantly

higher among the patients (P<0.001), a finding consistent with the higher proportion of diabetics within this group (60% vs. 36%). LDL-C, ox-LDL, and lipid peroxides (measured by TBARS and FOX methods) was similar for both groups (P>0.05).

Antioxidant vitamins status

Table 3 compares the average intake of energy, total fat, and antioxidant vitamins. CVD patients had a higher total energy intake (P<0.05). Although both serum vitamin A and E were similar between the groups (Table 2), the absolute dietary intake of vitamin A and E were higher among cases (P<0.001 and P<0.05 respectively). The difference remained significant for vitamin A intake (P<0.05) after correction for total fat intake. The mean daily intake of vitamin C and carotenoids did not differ significantly between the groups.

Univariate analysis

The univariate association between serum and dietary levels of antioxidant vitamins A and E, lipoproteins and measures of lipid peroxidation are shown in Table 4.

Table 2. Biochemical parameters in patients with CHD and controls.

	Controls (N=130)		Cases (N=130)	
	Mean	SD	Mean	SD
TC [mmol/L]	5.41	1.46	5.32	1.36
TG [mmol/L]	1.65	0.88	1.92 #	0.83
HDL-C [mmol/L]	1.31	0.52	1.36	0.61
LDL-C [mmol/L]	3.78	1.44	3.58	1.39
Atherogenic index (TC/HDL)	4.62	1.87	4.55	2.0
ox-LDL [U/L]	66.13	34.01	60.94	36.39
oxLDL/ LDL-C ratio	20.16	14.22	20.35	16.44
Lp(a) [mg/dl]	77.99	64.63	73.86	57.79
TBARS [uM]	2.29	0.87	2.42	0.88
FOX [uM]	2.01	0.89	2.14	0.85
Vitamin A (umol/L)	2.95	1.01	2.76	.919
Vitamin E (umol/L)	29.21	8.91	28.7	7.65
Vitamin A/TC ratio	0.58	0.25	0.55	0.23
Vitamin E/TC ratio	5.61	1.75	5.56	1.38

# P <0.001

Serum vitamin A concentrations correlated with dietary vitamin A in the group of subjects as a whole (r = 0.13, P<0.05) and in the patients with CHD (r = 0.222, P<0.05) after adjustment for age and serum cholesterol levels, suggesting that the FFQ provided a good estimate of intake. The dietary intake of vitamin E and A were correlated in the group as a whole (r = 0.146, P = 0.019) and among controls only (r=0.174, P=0.048). Serum concentrations of vitamin A and E, both corrected for serum cholesterol level, were strongly positively associated in both groups of subjects (controls r=0.478, P<0.0001; cases r=0.507, P<0.0001), and this finding may be related to their shared transport in serum.

Table 3. Total daily calorie, fat and antioxidant vitamin intake in patients with CHD and controls

	Controls (N= 130)	Cases (N=130)
Energy (kcal)	1848.36 ± 425.72	1974.4 ± 435.02 *
Total fat (gm)	79.59 ± 23.58	88.79 ± 26.62 *
Adjusted fat (gm)	79.59 ± 1.02	79.81 ± 0.96
% Energy supplied by fat	38.6 ±6.21	40.07 ± 5.85
Cholesterol (mg)	223.81 ± 96.68	264.05 ± 121.08 *
Vitamin C (mg)	41.32 ± 16.1	41.24 ± 18.1
Vitamin A (ug)	267.1 (836.3-146.5)	725 (1805.1-257.8) #
Carotenoids (ug)	12026.9 ± 2528.7	12407.0 ± 1978.2
Vitamin E (mg)	13.17 ± 3.69	14.4 ± 4.60 *
Vitamin A/total fat (ug/gm)	9.11 ± 10.59	13.17 ± 16.72 *
Vitamin E/total fat (mg/gm)	0.18 ± 0.07	0.18 ± 0.08

* P <0.05 # P <0.001

Table 4. Correlation coefficient between biochemical and demographic parameters in patients with CHD and controls. Pearson's or Spearman's correlation coefficients were calculated for each group separately

	All (N=260)	Controls (N =130)	Cases (N=130)
Serum vit A vs. LDL	-0.019	-0.032	-0.028
Serum vit A vs. ox-LDL	0.136 *	0.16	0.091
Serum vit A vs. ox-LDL/LDL ratio	0.129 *	0.196 *	0.07
Serum vit A vs. HDL	-0.031	0.111	-0.17
Serum vit A vs. TC/HDL ratio	0.012	-0.08	0.09
Serum vit A vs. TG	0.037	0.078	0.024
Serum vit A vs. dietary vit A (partial= age & total cholesterol)	0.13 *	0.054	0.222 *
Dietary vit A vs. ox-LDL	0.092	0.074	0.18 *
Dietary vit A vs. TBARS	-0.044	-0.099	-0.053
Serum vit E vs. LDL	0.388 #	0.331 #	0.453 #
Serum vit E vs. ox-LDL	0.003	0.084	-0.085
Serum vit E vs. ox-LDL/LDL ratio	-0.204 #	-0.115	-0.291 #
Serum vit E vs. HDL	0.004	0.101	-0.107
Serum vit E vs. TC/HDL ratio	0.257 #	0.188 *	0.322 #
Serum vit E vs. TG	0.296 #	0.306 #	0.29 #
Serum vit E vs. dietary vit E	-0.064	-0.041	-0.084
Dietary vit E vs. ox-LDL	-0.009	-0.129	0.02
Dietary vit E vs. TBARS	0.077	0.037	0.104
Serum vit A vs. vit E	0.279 #	0.315 #	0.229 #
Serum ratio vit A/TC vs. vit E/TC	0.489 #	0.478 #	0.507 #

* P <0.05 # P <0.001

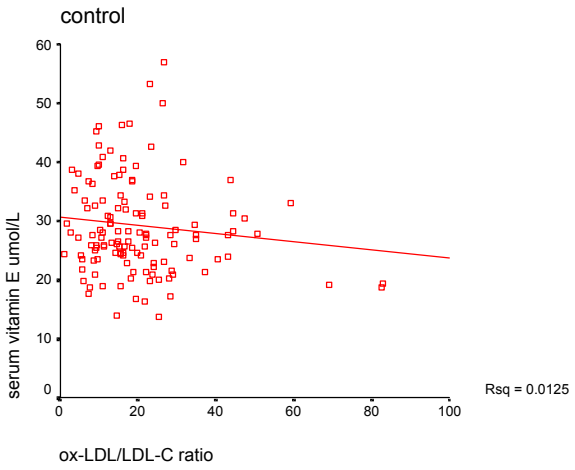

Figure 1 (a). Scatter plot showing correlation between serum vitamin E level and ox-LDL/LDL ratio in controls

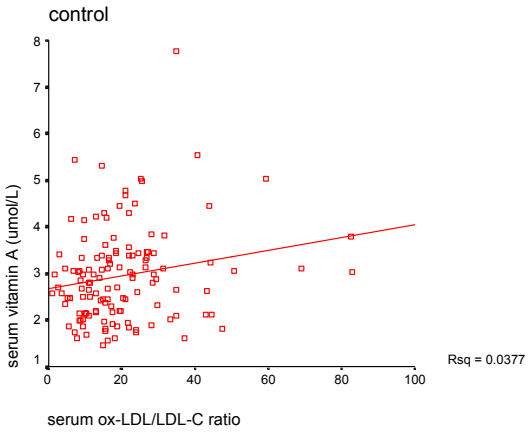

Figure 2 (a). Scatter plot showing correlation between serum vitamin A level and ox-LDL/LDL ratio in controls

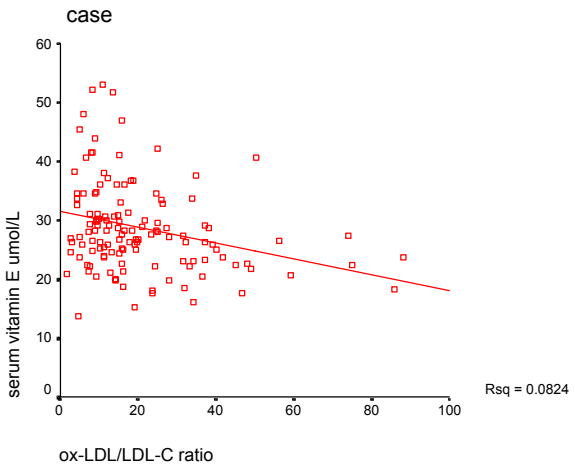

Figure 1(b). Scatter plot showing correlation between serum vitamin E level and ox-LDL/LDL ratio cases

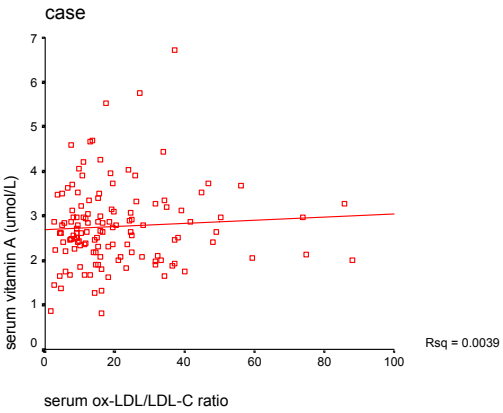

Figure 2(b). Scatter plot showing correlation between serum vitamin A level and ox-LDL/LDL ratio in cases

There was a negative correlation between serum vitamin E level and the ox-LDL/LDL ratio among the combined group (r =-0.204,*P*<0.001) and cases (r =-0.291, *P*<0.001) but not in the controls (*P*>0.05) (Fig. 1). There was also a positive association between serum vitamin A level and the ox-LDL/LDL ratio among the combined group (r =0.129, *P*<0.05) and controls (r = 0.196, *P*<0.05) but not cases (*P*>0.05) (Fig. 2).

Multivariate analysis

Conditional logistic regression analysis was performed to determine the association of antioxidant vitamins status with atherosclerosis. Potentially confounding and effect-modifying factors were included in the model. Diabetes mellitus, total fat intake, serum vitamin A and vitamin A intake corrected for total fat intake were shown to be significant predictors based on this model (Table 5).

Table 5. Adjusted odds ratio of CHD calculated from conditional logistic regression model.

	OR	95% CI		Wald	*P*
		Lower bound	Upper bound		
Serum vitamin A	0.724	0.528	0.992	4.048	0.044
Diabetes mellitus	2.492	1.421	4.370	10.144	0.001
Total fat intake	1.020	1.007	1.032	9.620	0.002
Vitamin A/total fat ratio	1.035	1.011	1.061	7.890	0.005

Discussion

Whilst previous results of observational cohort studies are compatible with a benefit of vitamin E supplements in CVD prevention, this was not the case for vitamin A or C.[8,9] Randomized placebo-controlled trials have generally not confirmed the benefit of antioxidant intervention[11-14] apart from patients post-angioplasty.[20] Findings from observational cohort studies may not be extrapolatable to intervention studies in high-risk patients. It is possible that antioxidant status exerts its effects early in the disease process, rendering late intervention ineffective. Most studies of the association between antioxidants and CVD have been undertaken in Western Caucasian populations. These findings may not be applicable to other populations. Despite the high prevalence of cardiovascular mortality and morbidity in the Middle East,[42-44]

there is only one published study on normative serum antioxidant vitamins concentrations in the Arab Gulf countries[45] and two studies of Saudis.[21,22] However, there are no data on the relationship between antioxidant status and CHD risk in this population.

Serum antioxidant vitamins and indices of lipid peroxidation

Gey[46] has proposed reference values for serum vitamin A (2.2-2.8μmol/L) and vitamin E (28-30) μmol/L). These values were considered optimal for the prevention of ischaemic heart disease and cancer. Approximately half of our study population had serum concentrations of serum vitamin E and A below the lower reference limit; 64% and 66% of controls and patients had serum vitamin E levels below 30 μmol/L, and 49% and 56% of controls and patients had serum vitamin A levels below 2.8 μmol/L.

A significant proportion of tocopherol is transported in cholesterol-rich lipoproteins, LDL, HDL and VLDL. This may explain the observed association between serum vitamin E and LDL (r=0.331, $P<0.0001$), and serum vitamin E and total cholesterol/HDL ratio (r=0.188, $P<0.05$). Serum vitamin E concentrations were also positively correlated with serum triglycerides (r=0.306, $P<0.0001$). Similar findings have been reported in previous studies.[47]

A high vitamin E/total cholesterol ratio should be indicative of LDL particles that are more resistant to peroxidation. In our study populations there was no significant difference in this ratio between the groups. However, within the patient group, there was a strong inverse relationship between serum vitamin E and ox-LDL/LDL ratio ($P<0.001$) (Table 4, Fig. 1). In contrast, there was a significant positive association between serum vitamin A and ox-LDL/LDL ratio suggesting that vitamin A content may not protect LDL from oxidation (Table 4, Fig. 2). The strong positive correlation between serum concentrations of vitamin A and E may be related to the common mode of transportation in serum.

Dietary antioxidant vitamins levels

In our population sample there was a relatively low intake of antioxidant vitamins, compared with the recommended dietary intake among both patients and control subjects (Table 3). Although the dietary intake of vitamin A and E was significantly higher in the CVD patients, this has not reflected in the serum α-tocopherol and vitamin A concentrations, which were similar for the two groups. This may, in part, be related to the higher prevalence of a positive smoking history and diabetes ($P<0.0001$) in the patient group, both conditions being associated with increased oxidative stress, and hence antioxidant consumption. Although the estimated dietary intake of these vitamins was significantly higher in the CVD group (Table 3). This may be related to the higher total fat intake among these patients as we have previously reported.[48]

The higher total calorie intake reported by the CVD patients despite higher proportion of them being on a diet (39%) suggests that these patients have not been fully compliant with dietary advice. It is possible that their original caloric intake was even higher prior to their coronary event.

The recommended intakes for vitamins A and C have recently been reported as 700μg and 40mg respectively.[39] The recommended intake for vitamin E is 15mg.[40] The estimated average requirement (EAR) is used in the assessment of the prevalence of inadequate nutrient intakes. The percentage of subjects in our study who reported consuming less than the EAR of vitamin A were 55% and 31% of controls and CVD patients respectively. The percentage of subjects reporting consuming less than the EAR of vitamin E were 14% and 12% of controls and CVD patients respectively. None of our subjects reported dietary vitamin C consumption less than the EAR.

The estimates of energy intake in our study are in agreement with those reported by other local[21,49] and international studies.[50-54] The modest positive correlation between serum vitamin A levels and dietary vitamin A (r=0.222, $P<0.05$) is similar to previous reports,[55] and provides an objective validation of our FFQ. However higher correlation coefficients between these two parameters have been reported in studies using vitamin supplements.[56] The lack of association between serum vitamin E levels and dietary vitamin E might be due to under-reporting of vitamin E intake from food since major food sources of this nutrient tend to be high in oil and these foods may be systematically under-reported.

Multivariate analysis

For optimal comparison, we attempted to match cases and controls for age. The confounding effects of non-independent factors were eliminated by a multivariate analysis. We found that serum vitamin A concentration was lower among patients with established CVD. Conversely, intakes of total fat and vitamin A levels were higher among patients. The traditional risk factors, diabetes mellitus also emerged as independent risk factors from our analysis. Individuals with diabetes mellitus are known to be exposed to higher levels of oxidative stress.[57] Serum vitamin A levels have shown to be of a protective value with an odds ratio of 0.724 ($P<0.05$, 95% CI: 0.528-0.992). Perhaps unexpectedly dietary vitamin A also emerged as a significant determinant of CVD with an odds ratio of 1.04 ($P<0.01$, 95% CI: 1.011-1.061). The importance of this finding is difficult to assess, however intervention trials using vitamin A supplements have not shown any significant benefit,[58] and in our study there was a positive association between dietary vitamin A and indices of LDL oxidation.

Conclusions

Classical coronary risk factors, diabetes, hypertension, positive smoking habits and dyslipidaemia were found to be more common among Saudi patients with established CVD. We have also demonstrated that in a non-Caucasian population, dietary factors contribute to coronary risk. Multivariate analysis showed that dietary total fat and vitamin A were independent coronary risk factors. This is the first report of a potentially deleterious role in CVD of dietary vitamin A in a non-Caucasian population. Unidentified, residual confounding factors, however may account for this finding. It is possible that the differences in anti-

oxidant vitamin status are a consequence of the disease rather than contributory determinants. Prospective cohort studies in non-Caucasian populations are needed to confirm our observations.

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