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# Biochemical assessment of nutritional status in pre- and post-natal Turkish women and outcome of pregnancy

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Objective: To determine by biochemical methods the nutritional status of pre- and post-natal Turkish women and its relationship with offspring anthropometry.

Design: Longitudinal study.

Setting: Health centres in Istanbul and Izmit, research department and university

hospital laboratories.

Subjects: Randomly selected group of women attending health centres at 13-17 weeks gestation (n = 130); same sample of women at 28-32 weeks gestation (n = 88) and 13-17 weeks post-partum (n = 95); offspring at 13-17 weeks post-partum (n = 90).

Interventions: Blood samples taken from mothers at all three stages and analysed for ferritin, iron, zinc, calcium, alkaline phosphatase, total protein, albumin, vitamins B<sub>2</sub>, B<sub>6</sub>, B<sub>12</sub>, A, E, β-carotene and folate levels; questionnaire completed for recording medical and socio-demographic background. Anthropometric measure-

ments taken from mothers and offspring.

Results: High percentages of subjects were at risk for deficiencies of vitamin  $B_{12}$  (48.8%) and folate (59.7%) in early pregnancy; ferritin (52.3%), zinc (72.3%), vitamin  $B_2$  (38.8%), vitamin  $B_{12}$  (80.9%), and folate (76.4%) during late pregnancy; and ferritin (39.0%), vitamins  $B_2$  (43.1%),  $B_6$  (36.4%),  $B_{12}$  (60.0%), and folate (73.3%) at the post-partum stage. Bone loss was indicated in 55.0% and 80.0% of the subjects in late pregnancy and post-partum respectively. Haematocrit in later pregnancy correlated strongly with prenatal body fat (P < 0.001). Infant anthropometry at 13–17 weeks post-partum was significantly affected by pre-natal weight gain and a number of maternal blood nutrients in pregnancy and post-partum.

Conclusions: Nutrition education programmes and enrichment of the staple food with iron, zinc, calcium, and the B vitamins should be considered.

Sponsorship: Turkish Government and in part Hoffman-La Roche & Co.

Descriptors: blood nutrients, pregnancy, post-partum

#### Introduction

Adequate nutrition during pregnancy and lactation is extremely important to both maternal and fetal health (Zeman & Ney, 1988). The fre-

quencies of low-birth-weight infants, stillbirths, and babies born with congenital disorders are high among malnourished mothers (Leader, Wong & Deitel, 1981; Pitkin, 1981). As the nutritional requirements are high, the effects of

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malnutrition can be severe and long-lasting in pregnant and lactating mothers. Nutritional status during these periods is thus implicated as a contributory factor in maternal and infant morbidity and mortality (Kafatos, Vlachonikolis & Codrington, 1989; Gonzalez-Cossio & Delgado, 1991).

The results of several national nutrition surveys previously conducted in Turkey indicated the prevalence of anaemia and malnutrition among pregnant and lactating women and their offspring (Köksal, 1977; Tönük, 1987). However, these studies did not reflect marginal deficiencies and were not based on biochemical measurements which are considered to be the most objective assessment of nutritional status (Sauberlich, 1978; Brubacher et al., 1979).

The aim of the present longitudinal study was to assess by biochemical methods the blood mineral, protein and vitamin levels of Turkish women at 3-4 and 6.5-7.5 months of pregnancy and 3-4 months post-partum and also to identify the associations of these biochemical indices with the mothers' and infants' anthropometric measurements and socio-demographic status.

# Subjects and methods

Subjects

The study group consisted of 130 women randomly selected from expecting mothers attending 10 different health centres in the Istanbul and Kocaeli provinces of Turkey. Random selection of candidates was achieved by drawing the names of 25 women from a list of all those both registered with the centre and in their second or third months of pregnancy. Of the 250 mothers selected in this manner, 164 responded to our invitation. Those mothers with histories of renal, thyroid, cardiovascular, diabetic and gross obstetric problems and hypertension were excluded from the study. The number of subjects at each centre ranged between 7 and 18. Blood samples were taken from the subjects at 13-17 weeks of pregnancy (1st stage, n = 130), 28–32 weeks of pregnancy (2nd stage, n = 88), and 13–17 weeks postpartum (3rd stage, n = 95). A number of the subjects were not available during the later two stages due to moving house, visiting families in another town, or reluctance to participate fur-

ther. No discernible grouping of social background, area, or medical history was apparent in these women.

During the first stage, the mothers were interviewed individually and a questionnaire form was filled to record obstetric history, smoking and alcohol consumption patterns, supplementation, medication, and food consumption levels and to assess their sociodemographic status based on the subject's education and occupation, husband's occupation, and housing conditions.

The protocol of the study was approved by the Turkish Government's Ministry of Health.

Field studies

Field studies were conducted between February and December, 1991 by calling the subjects to health centres or by house visits. Maternal height (MH) was measured at the first interview and maternal weight (MW), mid-upper-arm circumference (MUAC-M), and triceps skinfold thickness (TST) (using a Harpenden skinfold caliper) were measured at all three stages. It was not possible to get accurate pre-gravid weight records. Birth weight (BW) and birth length (BL) of the offspring were obtained from health centre records (n = 91). At the 3rd stage, infant weight (IW), length (IL), head circumference and mid-upper-arm circumference (MUAC-I) measurements were taken (n =90).

A sample of 20 ml venous blood was drawn from each woman by a nurse into separate vacutainers with and without heparin at least 2.5 h after breakfast or lunch at all three stages. Approximately 3 ml of the heparinised blood was immediately centrifuged and stored at 4°C until the enzyme assays for vitamins B<sub>2</sub> and B<sub>6</sub> could be carried out, at most within 10d of collection. For the determination of haematocrit, 5 ml of heparin-free blood was transferred to another test tube and analysed within an hour of collection. The remaining heparinised blood was centrifuged at 2000 g for 10 min, put into different Eppendorf tubes; frozen immediately, and stored at -30°C until the analyses of zinc, β-carotene, folate, vitamins A, E, and  $B_{12}$ . Unheparinised blood was prepared for the analyses of ferritin, iron, calcium, total protein, albumin, and alkaline phosphatase according to their respective methods.

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Table I C

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1st and 3 2nd stag Ferritin (S) Iron (S) (µ Zinc (P) (µ 1st stage 2nd stage 3rd stage Calcium (S Alkaline ph Total protei Albumin (S α-EGR α-EGOT Vitamin B<sub>1</sub>

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### Biochemical analyses

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Haematocrit was measured by the Cell analyser Ca600 Hemogram Standard Method. Ferritin was determined using a radioimmunoassay kit (Kodak Clinical Diagnostica LTD, Amersham, UK) and iron by using the test-combination ferro kit (Boehringer Mannheim GmbH Diagnostica). Zinc levels were measured by the Perkin Elmer method based on standard atomic absorption determination of zinc in plasma (Butrimovitz & Purdy, 1977). Calcium, total protein, albumin and alkaline phosphatase activity were measured by the Technicon SMA II Model Autoanalyser.

Vitamin B<sub>2</sub> and B<sub>6</sub> levels were determined by co-enzyme stimulation of the erythrocyte enzymes glutathione reductase (EGR) and glutamate oxaloacetate aminotransferase (EGOT) respectively and the results were expressed as activation coefficients  $\alpha$ -EGR and  $\alpha$ -EGOT (Vuilleumier, Keller & Keck, 1990; Wetherilt et al., 1992).

Folate and vitamin B<sub>12</sub> concentrations were measured simultaneously by using a Solid Phase No Boil Dual-count radioassay kit (Diagnostic Products Corporation, Los Angeles, CA). β-Carotene and vitamin A were determined by the high-performance liquid chromatography method of Vuilleumier et al. (1983) and vitamin E by the modified micro method of Emmerie and Engel (Fabianek et al., 1968).

#### Diagnostic criteria

The cut-off points for the classification of the results from biochemical analyses for the different stages are shown in Table 1. Normal and deficient levels of haematocrit, alkaline phosphatase, vitamins  $B_{12}$  and E, total protein, and albumin were obtained from Sauberlich, Dowdy & Skala (1974). Cut-off points for ferritin were obtained from Butte, Calloway & Van Duzen (1981); for iron and vitamin A from Bowering, Lowenberg & Morrison (1980); for zinc from Lehti (1989), Hambidge et al. (1983) and Butte, Calloway & Van Duzen (1981); for calcium from Gibson (1990); for vitamins B<sub>2</sub> and B<sub>6</sub> from Wetherilt et al. (1992); and for folate from Lehti (1989). Bone loss was defined as high alkaline phosphatase levels above the osteomalacia limit as given by Sauberlich, Dowdy & Skala (1974); deficiencies for iron were defined as low serum ferritin levels, for vitamin B<sub>2</sub> and  $B_6$  as high  $\alpha$ -EGR and  $\alpha$ -EGOT values respectively.

Table 1 Criteria for nutrient deficiencies and anaemia in pregnancy and post-partum

	High risk	Moderate risk	Low risk
Haematocrit (%)			
1st and 3rd stages	<30.0	30.0-34.0	≥35.0
2nd stage	<30.0	30.0-32.0	≥33.0
Ferritin (S) (µg/l)			≥9.0
Iron (S) (µmol/l)			≥10.7
Zinc (P) (µmol/l)			
1st stage	<7.9	7.9-10.5	≥10.6
2nd stage	<6.6	6.6-8.9	≥9.0
3rd stage	<9.2	9.2-10.0	≥10.1
Calcium (S) (mmol/l)			≥2.2
Alkaline phosphatase (S) (µkat/l)			≤1.3
Total protein (S) (g/l)	<55	55-59	≥60
Albumin (S) (g/l)	<30	30-34	≥35
α-EGR	>1.52	1.52-1.44	<1.44
α-EGOT	>1.80	1.80-1.70	<1.70
Vitamin B <sub>12</sub> (P) (pmol/l)	<73.8	73.8-110.6	≥110.7
Folate (P) (nmol/l)	<6.8	6.8-13.5	≥13.6
Vitamin A (P) (µmol/l)	<0.35	0.35-1.04	≥1.05
Vitamin E (P) (µmol/l)	<11.6	11.6-16.2	≥16.3

1st stage: 13-17 weeks pregnancy; 2nd stage: 28-32 weeks pregnancy; 3rd stage: 13-17 weeks post-partum. S = serum; P = plasma; EGR = erythrocyte glutathione reductase; EGOT = erythrocyte glutamate oxaloacetate aminotransferase.

Table 2 Anthropometric measurements of pre- and post-natal mothers (mean  $\pm$  s.d.)

	1st stage (n = 130)	2nd stage (n = 88)	3rd stage (n = 95)
MH (cm)	155.8 ± 5.9	_	
MW (kg)	$60.7 \pm 9.9$	65.7 ± 10.9	61.7 ± 11.6
MUAC-M (cm)	$27.4 \pm 3.8$	$27.8 \pm 3.2$	$27.9 \pm 3.4$
TST (mm)	$19.3 \pm 6.5$	$20.2 \pm 7.1$	$20.5 \pm 6.9$

MH = maternal height; MW = maternal weight; MUAC-M = maternal mid-upper-arm circumference; TST = triceps skinfold thickness.

Relative weight of the infants was found by calculating the ratio of the subjects' weight to the desirable weight of a normal child of the same length according to Turkish norms (Neyzi, Binyıldız & Alp, 1978).

# Data analyses

Standard statistical methods of means and standard deviations were used for the evaluation of nutritional indices. Two-tailed Student's t-test for unpaired data was used for differences between the means of biochemical indices at the different stages. Data were subjected to linear regression analysis and analysis of variance to calculate F values for correlations using the NAGFLIB GO2CCF system program (Numerical Algorithms Group, 1984). Results were considered significant for F statistics at the 0.05 level.

For correlation purposes, a socio-demographic status score (SDSS) was compiled for each subject on the basis of her education (0-2), occupation (1-3), husband's occupation (0-3), house ownership (1-3), and household utilities (0-3).

#### Results

Mean maternal age at the first interview was  $23.7 \pm 4.0$  years (range 16-35) and the majority of women (77.0%) were aged between 20 and 29 years. Of the total group, 48% were primiparas, 34% secondiparas, 12% terciparas, and 6% multiparas with four and more pregnancies. Anthropometric measurements of the subjects at all three stages of the study are presented in Table 2. The mean monthly weight gain in the subjects between the 1st and 2nd stages (WG) was  $1.86 \pm 1.16$  kg. BW, BL, and anthropometric measurements of infants at 13–17 weeks post-partum are shown in Table 3. The percentage of infants with BW <2500 g was 3.2%. Three babies were miscarried, two were stillborn, and two died in the first 3 months following birth. Breast-fed, mixed-fed and bottle-fed infants comprised 30.0%, 60.0% and 10.0% of the offspring population, spectively.

The percentages of women with secondary, primary and no education (illiterate) were 19, 78 and 3, respectively. None had attended university. Only 5% worked as labourers or civil servants; the rest were housewives. All were married. Of the husbands, 94% were employed as blue-collar workers, civil servants or small tradesmen/craftsmen and 6% were unemployed. With respect to housing, 41% of the families owned and 55% rented their homes while 4% lived in shanty houses. Most families (91%) had a refrigerator.

Table 3 Infants' anthropometric measurements (mean ± s.d.)

	At birth (n = 91)	At 13–17 weeks post-partum (n = 90)
Length (cm)	50.0 ± 1.3	61.6 ± 3.9
Weight (kg)	$3.51 \pm 0.56$	$6.27 \pm 1.13$
HC (cm)		$40.5 \pm 1.9$
MUAC-I (cm)		$13.6 \pm 1.4$
Relative weight		$98.9 \pm 14.6$

HC = head circumference; MUAC-I = infant mid-upper-arm circumference; relative weight = ratio of infant's weight to the desirable weight of a normal child of the same length according to Turkish norms (Neyzi et al., 1978).

Table

Haemar Ferritin Iron (S) Zinc (P Calciun Alkalin Total pr Albumi

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Haemate Ferritin Iron Zinc Calcium Alkaline Total pr Albumi Vitamin Vitamin Vitamin Folate

<sup>a</sup>Criteria 1st stage

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Vitamin

Table 4 Biochemical nutritional status of pre- and post-natal mothers by stages (mean  $\pm$  s.d.)

	Ist stage	2nd stage	3rd stage
Haematocrit (%)	$36.7 \pm 4.4^{a} (n = 115)$	$35.1 \pm 3.3^{\text{b}} \ (n = 88)$	$36.9 \pm 3.9^{\circ} (n = 86)$
Ferritin (S) (µg/l)	$33.2 \pm 48.7^{a} (n = 108)$	$12.8 \pm 21.2^{\text{b}} (n = 86)$	$19.2 \pm 20.3^{b} (n = 77)$
Iron (S) (µmol/l)	$19.5 \pm 8.2^{a} (n = 126)$	$17.6 \pm 8.6^{a.b} (n = 80)$	$16.3 \pm 7.1^{h} (n = 64)$
Zinc (P) (µmol/l)	$13.1 \pm 4.7^{a} (n = 124)$	$7.7 \pm 2.1^{\rm b} (n = 82)$	$11.3 \pm 2.3^{\circ} (n = 90)$
Calcium (S) (mmol/l)	$2.34 \pm 0.15 \ (n = 125)$	$2.29 \pm 0.30 \ (n = 64)$	$2.32 \pm 0.17 (n = 79)$
Alkaline phosphatase (S) (µkat/l)	$1.06 \pm 0.29^{\circ} (n = 127)$	$1.51 \pm 0.77^{b} (n = 61)$	$2.47 \pm 1.58^{\circ} (n = 79)$
Total protein (S) (g/l)	$68.0 \pm 4.0^{a} (n = 124)$	$65.0 \pm 4.0^{\circ} (n = 69)$	$75.0 \pm 6.0^{\circ} (n = 69)$
Albumin (S) (g/l)	$41.0 \pm 4.0^{a} (n = 126)$	$37.0 \pm 4.0^{\circ} (n = 70)$	$44.0 \pm 4.0^{\circ} (n = 78)$
α-EGR	$1.34 \pm 0.23^{\circ} (n = 110)$	$1.40 \pm 0.28^{a.b} (n = 88)$	$1.46 \pm 0.29^{h} (n = 87)$
α-EGOT	$1.55 \pm 0.24 \ (n = 128)$	$1.53 \pm 0.28 \ (n = 88)$	$1.61 \pm 0.28 \ (n = 86)$
Vitamin B <sub>12</sub> (P) (pmol/l)	$140.8 \pm 105.0^{\circ} (n = 129)$	$94.6 \pm 107.8^{\text{b}} (n = 87)$	$127.1 \pm 83.2^{a} (n = 84)$
Folate (P) (nmol/l)	$13.4 \pm 5.9^{a} (n = 129)$	$10.4 \pm 6.5^{\circ} (n = 88)$	$11.8 \pm 10.1^{a,b} (n = 90)$
β-Carotene (P) (μmol/l)	$0.78 \pm 0.46^{a} (n = 118)$	$1.14 \pm 0.70^{\circ} (n = 87)$	$2.40 \pm 1.53^{\circ} (n = 89)$
Vitamin A (P) (µmol/l)	$2.05 \pm 1.87^{a} (n = 110)$	$1.44 \pm 0.62^{\text{b}} \ (n = 87)$	$2.28 \pm 1.85^{a} (n = 92)$
Vitamin E (P) (µmol/l)	$17.2 \pm 4.0^{a} (n = 126)$	$19.0 \pm 5.8^{\text{b}} \ (n = 88)$	$17.7 \pm 4.6^{\text{a.b}} (n = 95)$

a.b.c.Values for mean  $\pm$  s.d. in the same row with different superscript letters are significantly different (P < 0.05). 1st stage: 13-17 weeks pregnancy; 2nd stage: 28-32 weeks pregnancy; 3rd stage: 13-17 weeks post-partum. Abbreviations: see Table 1.

None of the women took alcohol and 17.7% were light smokers, ie smoked <6 cigarettes/ day. Average tea consumption was 4 cups/day and coffee intake was negligible. A number of the subjects were on supplementation from 6-7 weeks of pregnancy until parturition: 35.4% were regularly taking iron (40-100 mg/day)

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and/or multivitamins and 6.1% took supplementation on a sporadic basis. None of the subjects continued with supplementation after parturition. Medication (pain-killers and antibiotics) was taken by 12.3% of the study population at some stage of gestation. Also, a total of 9 women were prescribed megadoses of

Table 5 Percentage of subjects with high risk (HR)<sup>a</sup> and moderate risk (MR)<sup>a</sup> of anaemia, alkaline phosphatase activity and nutrient deficiencies in pregnancy and post-partum

	1st stage		2nd	stage	3rd stage	
	HR	MR	HR	MR	HR	MR
Haematocrit	7.6	17.8	5.4	22.6	6.8	17.1
Ferritin	25.7		52.3		39.0	
Iron	15.1		16.3		26.6	
Zinc	15.0	14.1	38.6	33.7	15.4	17.6
Calcium	10.3		14.0		15.0	
Alkaline phosphatase	21.1		55.0		80.0	
Total protein	0.0	0.8	4.3	4.3	1.3	0.0
Albumin	0.0	5.6	2.9	18.6	0.0	0.0
Vitamin B <sub>2</sub>	20.0	6.9	23.7	15.1	26.1	17.0
Vitamin B <sub>6</sub>	14.7	6.2	16.1	4.3	28.4	8.0
Vitamin B <sub>12</sub>	27.9	20.9	56.2	24.7	23.5	36.5
Folate	7.0	52.7	23.6	52.8	33.3	40.0
Vitamin A	1.8	22.7	1.2	20.9	1.1	0.0
Vitamin E	6.4	31.8	9.1	20.5	12.6	15.8

<sup>&</sup>lt;sup>a</sup>Criteria for high and moderate risk groups defined in Table 1.

<sup>1</sup>st stage: 13-17 weeks pregnancy; 2nd stage: 28-32 weeks pregnancy; 3rd stage: 13-17 weeks post-partum.

Table 6 Maternal blood nutrients associated with socio-demographic status and anthropometric measurements in pregnancy and post-partum (P value)

			Anthropometric measurements								
				Ist stage			2nd stage		·	3rd stage	
	SDSS	WG	MW	MUAC-M	TST	$\overline{MW}$	MUAC-M	TST	$\overline{MW}$	MUAC-M	TST
Ist stage			_								
Ferritin (S)	÷	0.001	†	†	†	†	†	÷	†	†	÷
Iron (S)	†	-0.05	†	<b>†</b>	0.05	÷	Ť	÷	÷	†	; †
Zinc (P)	†	-0.01	†	†	0.01	†	†	÷	†	, †	; †
Calcium (S)	†	-0.01	†	†	0.05	†	†	+	÷	; †	+
Vitamin B <sub>2</sub>	+	0.05	†	†	†	÷	†	0.05	÷	; †	+
Vitamin B <sub>12</sub> (P)	†	-0.01	†	†	÷	†	†	†	; <del>;</del>	†	† †
Folate (P)	0.05	†	†	ŧ	1	Ť	†	÷	÷	†	†
β-Carotene (P)	0.05	-0.05	†	<b>†</b>	†	†	÷	+	+	†	†
Vitamin E (P)	÷	-0.05	ŧ	†	†	†	†	†	†	†	; †
2nd stage											
Haematocrit	†	÷	0.001	0.01	0.001	0.01	0.01	0.001	0.05	0.05	0.05
Vitamin B <sub>2</sub>	†	†	†	†	†	†	†	†	†	0.05	0.05
Vitamin B <sub>6</sub>	0.05	†	†	†	†	÷	†	+	; †	†	†
Vitamin A (P)	0.05	†	0.05	ŧ	†	†	†	†	†	, †	† †
3rd stage											
Albumin (S)	÷	0.001	÷	0.001	ŧ	†	0.001	÷	÷	0.001	
Vitamin B,	0.05	†	†	. †	÷	†	†	; †	† †	0.001 †	† †
Vitamin B <sub>6</sub>	†	0.001	÷	†	†	; †	†	; †	†	† †	
Vitamin B <sub>12</sub> (P)	†	†	+	; †	†	†	; †	+	, †	-0.05	† †
Folate (P)	0.05	÷	†	÷	4	; †	†	†	†	-0.05 -0.05	
β-Carotene (P)	0.05	†	÷	†	†	†	†	†	†	-0.05 †	† †
Vitamin A (P)	†	÷	0.05	÷	†	†	†	; †	† †	†	₹ †
Vitamin E (P)	0.05	†	†	÷	†	÷	† †	† †	†	†	† †

<sup>†</sup>Not significant for F statistics (P > 0.05).

1st stage: 13-17 weeks pregnancy; 2nd stage: 28-32 weeks pregnancy; 3rd stage: 13-17 weeks post-partum. SDSS = socio-demographic status score; WG = weight gain; MW = maternal weight; MUAC-M = maternal midupper-arm circumference; TST = triceps skinfold thickness; S = serum; P = plasma.

vitamin B<sub>6</sub> (300 mg/tablet) to combat pregnancy nausea.

Means and standard deviations of biochemical nutritional status indicators of the pregnant and post-pregnant women are shown in Table 4 and their percentages in the high and moderate risk categories for deficiencies in Table 5.

The significant associations of biochemical variables with SDSS and maternal anthropometric measurements are shown in Table 6. Parity and smoking habits were not found to correlate significantly with maternal indices or infant anthropometry. Frequency of tea intake did not associate with haematocrit or any other biochemical indices.

SDSS was positively correlated with BW and HC (P < 0.05) but not with any other infant anthropometry. The significant associations elicited between maternal indices and infant anthropometry are given in Table 7. BW correlated positively with BL (P < 0.01), IW (P< 0.01), HC and MUAC-I (P < 0.05) but not with IL. BL did not associate with any of the offspring anthropometry at 13-17 weeks postpartum.

# Discussion

The National Nutrition Health and Food Consumption Survey of Turkey carried out in 1974 revealed that pregnant and lactating women together with their infants represent the groups most vulnerable to nutritional deprivation in Turkey (Köksal, 1977), where the infant mortality rate is 71.0 and 51.6 per thousand in rural and urban areas, respectively (TSIS, 1993). Our

Table 7

Maternal WG All three MW MUAC-TST Ist stage Haemat Calcium Vitamin Vitamin Folate ( 2nd stage Ferritin Zinc (P) Albumii Vitamin Folate ( Vitamin 3rd stage

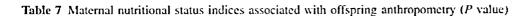
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subjects 90% ha Anatolia Istanbul towns in

The m acceptab mendation of the In there we MUACpregnand

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Maternal indices			Infants' anthrope	ometric measuren	nents	
	BW	BL	IW	IL	НС	MUAC-I
WG	0.05	0.01	0.01	0.01	0.01	0.05
All three stages						
MW	0.001	÷	÷	÷	÷	÷
MUAC-M	0.001	÷	÷	÷	÷	÷
TST	0.01	÷	÷	÷	÷	†
1st stage						
Haematocrit	Ť	†	-0.05	-0.01	÷	-0.05
Calcium (S)	÷	÷	÷	-0.05	<b>†</b>	÷
Vitamin B <sub>2</sub>	÷	÷	0.01	÷	0.05	0.01
Vitamin B <sub>12</sub> (P)	÷	÷	÷	-0.01	÷	†
Folate (P)	÷	÷	÷	-0.01	÷	÷
2nd stage						
Ferritin (S)	÷	÷	-0.05	÷	-0.05	-0.05
Zinc (P)	÷	÷	÷	-0.05	†	+
Albumin (S)	÷	÷	0.05	÷	<u>;</u>	<b>†</b>
Vitamin B <sub>12</sub> (P)	÷	†	†	÷	<del>1</del>	0.01
Folate (P)	<del>*</del>	÷	Ť	÷	1	10.0
Vitamin A (P)	+	†	‡	-0.05	÷	†
3rd stage						
Albumin (S)	÷	†	÷	Ť	0.01	0.01
Vitamin B <sub>6</sub>	÷	÷	-0.05	†	-0.05	ŧ

†Not significant for F statistics (P > 0.05).

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1st stage: 13-17 weeks pregnancy; 2nd stage: 28-32 weeks pregnancy; 3rd stage: 13-17 weeks post-partum. BW = birth weight; BL = birth length; IW = infant weight; IL = infant length; HC = head circumference; MUAC-I = infant mid-upper-arm circumference; WG = weight gain; MW = maternal weight; MUAC-M = maternal mid-upper-arm circumference; TST = triceps skinfold thickness; S = serum; P = plasma.

subjects came from urban areas; however, over 90% had their roots in the rural areas of Anatolia and had migrated in the last decade to Istanbul and Izmit, the two most industrialised towns in Turkey.

The mean WG of the subjects was within the acceptable ranges according to the recommendations of the US Food and Nutrition Board of the Institute of Medicine (1990) even though there were no significant changes in the mean MUAC-M and TST from 3-4 months of pregnancy to 3-4 months post-partum.

The number of women in the high-risk group for haematocrit deficiency was low at all stages and mild anaemia was observed in only about a fifth of the subjects. On the other hand, a high percentage of mothers was found to be at risk for ferritin deficiency, especially at the later stage of pregnancy. It was indeed surprising to observe in this group a high prevalence of storage iron deficiency but only mild anaemia.

Although measurement of serum iron is considered an indirect and non-specific method for the evaluation of iron status (Sauberlich *et al.*, 1974), the number of women at risk for haematocrit deficiency was more in accord with risk groups of this index rather than with those of ferritin.

A prevalence of zinc deficiency was observed during the later stage of pregnancy. Previous studies have shown that low serum concentrations of maternal zinc are associated with an increased incidence of fetal malformations and fetomaternal complications (Breskins *et al.*, 1983; Mukherjee *et al.*, 1984). On the other hand, zinc supplementation is not available in Turkey and as the bioavailability of dietary zinc can be impaired by folate and inorganic iron (Hambidge *et al.*, 1983; Simmer *et al.*, 1987), supplementing Turkish pregnant women with iron and folic acid without monitoring their zinc status could have detrimental effects.

Pregnancy and lactation are periods of major stress on the metabolism of calcium, the major building block of the mineralised phase of bone; however, maternal serum calcium is not considered a good indicator of calcium availability as the serum levels of this mineral are kept at a steady state on account of loss from bones (Gibson, 1990). Indeed, in our study, the percentage of women with low serum calcium levels was much less than that of women at risk with respect to alkaline phosphatase activity. The lack of associations between alkaline phosphatase activity and infant anthropometry confirmed that the stress of calcium and vitamin D deficiency was on the mother rather than the infant. The level of bone loss, which was severe at the later stage of pregnancy and aggravated even further during lactation, calls for immediate measures, possibly in the form of guidance for vitamin D supplementation and a national programme to educate mothers to consume more milk products.

A substantial proportion of the subjects were under high risk of vitamin B2 deficiency, again most likely as a result of insufficient consumption of milk products as well as green leafy vegetables and whole cereals. Considering the low animal product consumption of the study group, vitamin B<sub>6</sub> deficiency was less than expected during pregnancy. One reason may be that some of the women were taking megasupplements of vitamin B<sub>6</sub> for relief from nausea. After birth, all the subjects ceased to take supplementation of any kind which may possibly explain the rise in the number of women with vitamin B<sub>6</sub> deficiency in the postnatal period.

Epidemic levels of vitamin B<sub>12</sub> and folate deficiency were observed during pregnancy and post-partum. As our subjects were largely from low-income groups, they cannot be expected to consume foods of animal origin in sufficient amounts to meet their obviously high vitamin  $B_{12}$  needs. Therefore, if possible, women from similar backgrounds in Turkey should be advised to take vitamin B<sub>12</sub> supplementation under sensible medical guidance. Folate supplementation also appears to be necessary, especially in view of the observed relationship between folate intake and the incidence of neural tube defects (Rush, 1994). In fact, in our study, one of the offspring who was born to a mother at high risk of plasma folate deficiency

suffered from spina bifida and died 1.5 months after birth. Also, dietary intakes can be increased if expecting mothers could be persuaded through nutrition education programmes to consume more green leafy vegetables, which are inexpensive and found in ample amounts at all seasons in Turkey.

In contrast to the trends of all the other nutritional indices studied, levels of \( \beta-carotene and vitamin E, the two fat-soluble nutrients with antioxidant properties, increased significantly from early to late pregnancy. This may possibly be due to the changes in lipid metabolism and oxidation during pregnancy.

According to Turkish norms, calculated relative weights of the 13-17-week-old infants were within normal limits in 69.8% of the boys (n = 46) and 47.8% of the girls (n = 44). Although there were no boys with severe malnutrition, 22.7% of the girls were found to be suffering from serious wasting with respect to relative weight. This difference in nutritional status (P < 0.001) may be due to a relatively closer attention given to the male offspring.

As expected, babies of greater BW were born to mothers with better anthropometry and WG at pregnancy (Leader, Wong & Deitel, 1981; Brown et al., 1986). Smoking habits of mothers did not affect infant anthropometry, possibly because less than a fifth smoked and those who did were careful not to exceed 4-5 cigarettes per day. This is in agreement with the results of a study by Czajka-Narins & Jung (1986) on a group of pregnant women with a similar smoking profile.

Women with better socioeconomic status appeared to have higher folate,  $\beta$ -carotene, and vitamin E and B<sub>2</sub> levels. Interestingly, all these vitamins are found in generous amounts in green leafy vegetables which are possibly consumed in larger amounts by the bettereducated subjects.

Mothers with greater weight, MUAC-M and TST were significantly less anaemic at the later stage of pregnancy, emphasising the importance of pre-natal weight on the haematopoietic system. Women with greater WG had significantly higher ferritin levels in early pregnancy and albumin and vitamin B<sub>6</sub> levels in post-pregnancy, possibly indicating a better protein synthesis. Higher albumin levels were found in the post-partum mothers who had greater MUAC-M at all three stages, implying

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Am. J.Breskin M Z, Plovi that MUAC may be an indicator of long-term protein nutriture in this population. Surprisingly, however, lower levels of vitamin B<sub>12</sub> and folate were found in the post-partum mothers who had greater MUAC-M, and we cannot suggest an explanation for this.

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Subjects with higher vitamin B<sub>2</sub> levels in early pregnancy had greater WG and more body fat in late pregnancy while those with higher vitamin B<sub>2</sub> levels in late pregnancy had more body fat in post-pregnancy. These observations confirm the importance of this vitamin as a co-enzyme in energy utilisation and show its impact during both the pre- and post-natal periods.

Interestingly, subjects with higher levels of iron, zinc and calcium in early pregnancy had more body fat, but put on less weight until the later stage of pregnancy. Women with higher levels of vitamins  $B_{12}$ , E and  $\beta$ -carotene in early pregnancy also put on less weight; however, these biochemical indices did not correlate with body fat. On the other hand, women who were heavier in early pregnancy had better vitamin A status in late pregnancy and post-partum.

The lack of associations between mothers' biochemical nutrient levels and BW or BL may imply that the apparent impact of the mother's nutritional stress was on the mother rather than the baby; however, WG and vitamin B<sub>2</sub> levels in earlier pregnancy, albumin, folate and vitamin B<sub>12</sub> levels at the later stage of pregnancy, and albumin levels in post-pregnancy appeared to affect significantly the growth and development of the 13–17-week-old infants. The effects of maternal post-pregnancy albumin status on HC and MUAC-I were unexpected as no albumin deficiency was found in any of the subjects at this stage.

We cannot surmise any single explanation for the apparently adverse effects of higher blood haematocrit, calcium, vitamin  $B_{12}$ , and folate levels in early pregnancy; ferritin, zinc, and vitamin A levels in late pregnancy; and vitamin B<sub>6</sub> levels in post-pregnancy on the anthropometry of 13–17-week-old infants. A similar inverse relationship was also reported previously between mothers' haemoglobin values after delivery and birth weight of infants (Khan, 1985). These unexpected observations are possibly due to physiological adjustments and other confounding maternal factors rather than direct causative effects.

In conclusion, a prevalence of bone loss and serious deficiencies for ferritin, zinc, vitamins B<sub>2</sub>, B<sub>6</sub>, B<sub>12</sub> and folate are indicated in a crosssection of Turkish pregnant and post-pregnant women. It is unfortunate to find such high levels of B-complex vitamin deficiencies in a country like Turkey where there are ample sources of cereals, legumes, nuts and green vegetables. One main reason could be the extreme refining of the flour used in making bread, the staple food in the country. This is done throughout the country to render bread a white colour to improve its marketability, yet in so doing most of the vitamins in the wheat are lost. Thus our results confirm the necessity of a country-wide nutrition programme to educate the families to prefer brown bread, eat more green leafy vegetables and spare as much of their income as possible to provide milk products for their vulnerable members. Enrichment of bread with the B vitamins, iron, zinc, and calcium should also be considered. In addition, we believe that zinc supplementation should be made available for pregnant women and a survey investigating possible links between megadose intakes of vitamins/ minerals and pregnancy complications should be carried out in this country.

Evaluation of data for dietary intakes and comparison with biochemical nutritional status indicators will be presented in a forthcoming paper.

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