

Nutritional, immunological and health status of the elderly population living in poor neighbourhoods of Quito, Ecuador

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The number of elderly people is increasing in less-developed countries. Although nutritional deficiencies and infectious diseases are generally more prevalent in resource-poor countries, the health and nutritional status of the elderly in South America in general, and in Ecuador, in particular, remains largely unstudied. The objective of the present study was to assess the nutritional, immunological and health status of elderly Ecuadorians. A cross-sectional study was conducted to evaluate a sample of elderly Ecuadorians with 24 h dietary recalls, biochemical and anthropometric measurements, delayed type hypersensitivity skin response and a health questionnaire. The 145 elders who enrolled had a mean age of 74.3 (SD 6.9) years. Of the subjects, 52% exhibited BMI ≥ 25 kg/m², whereas 9.1% had BMI ≤ 20 kg/m². Means of dietary intakes were below recommendations for most nutrients; exceptions were carbohydrate, fat, Fe and Se. Serum nutrient levels indicated that 50, 44, 43, 19 and 18% of participants had deficiencies of Zn, Fe, vitamins B₁₂ and D, and folate, respectively. The mean number of positive responses to seven recall antigens was 2.1 (SD 1.7) with an induration diameter of 9.9 (SD 7) mm, which are substantially lower than those reported for elders in developed countries. During the previous 6 months, 54 and 21% of subjects reported at least one episode of respiratory infection or diarrhoea, respectively. Of these, 47% sought care at a hospital or from a physician and 96% from a relative or friend. In conclusion, while few elderly Ecuadorians were underweight, obesity was common. Micronutrient deficiencies were prevalent and may contribute to reduced immunological responses in this population.

Elderly: Immune senescence: Less-developed countries: Micronutrients: Body mass index

The number of elderly individuals is increasing in both developed and less-developed countries. Populations in less-developed countries are currently undergoing rapid and unprecedented changes in their age structure, which will have a dramatic impact on the number of aged in this century. The percentage of the total worldwide population over 60 years of age is expected to more than double from 8 to 19% by 2050 (United Nations, 2001). The increase in the proportion of elderly in less-developed countries is projected to grow 1.5 times faster than that in developed countries, and by 2050, approximately 80% of all individuals over the age of 60 years will be living in less-developed countries (United Nations, 2004). For the last two decades, developed nations have gradually increased the proportion of their national health resources designated for the elderly, but very little of the limited and already strained health resources of

less-developed countries have been devoted to ageing members of their societies. Because the elderly have a decreased capacity to fight disease without adequate nutritional, economical, and social support, they face the risk of living their lives burdened by disease and dysfunction. This threatens to put an even greater strain on their countries' resources.

Age-associated physiological, psychological, social, and economic changes may adversely affect the nutritional and immunological status of older individuals. These changes are often reflected in poor health and a reduced quality of life. The incidence of infections (Crossley & Peterson, 1996) and diseases such as cancer, atherosclerosis and autoimmunity (Cross *et al.* 1987) have been shown to increase with age, reflecting, in part, alterations in immune function seen in the elderly. The elderly are at greater risk for low consumption

Abbreviations: DTH, delayed type hypersensitivity; PLP, pyridoxal-5-phosphate.

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of several micronutrients such as vitamins E, C, and B₆, folate, Se, Fe and Zn, which have been shown to play important regulatory roles in maintaining the function of the immune system (Meydani & Santos, 2000). Malnutrition in the elderly has been shown to be associated with impairment of the immune response and decreased functional status, and this has been shown to respond to nutrient repletion (Lesourd, 1995; Meydani & Santos, 2000; High, 2001).

The health and nutritional status of the elderly in South America, in general, and in Ecuador, in particular, remains largely unevaluated. According to estimates based on a census conducted in 2000 (Instituto Nacional de Estadística y Censos, 2001), the total population of Ecuador is approximately 12 million with an estimated 6.7% (813 624) of inhabitants over the age of 65 years. The elderly population of Quito, the capital of Ecuador, is estimated to be 122 417, which represents a significant number of urban inhabitants whose health and nutritional status remains largely uncharacterised. This cross-sectional study of an independently living elderly population in a low-income urban area of Quito presents, for the first time, detailed anthropometric, dietary recall, nutritional, biochemical and immunological parameters in elderly Ecuadorians.

Methods

Study site and population

The present study was conducted in elderly Ecuadorians who live in poor urban areas in northwestern Quito (Ecuador), 2800 m above sea level. The neighbourhoods are located on a hill and are structurally similar. There is only one principal paved road and electricity is present in all homes. Some households have a municipal source of potable water and sewerage. Most houses consist of two rooms of cement block construction. Inhabitants are mainly poor immigrants from small cities and rural areas of Ecuador. To identify eligible elderly individuals, we carried out a census in three neighbourhoods. During household visits we provided potential study candidates with detailed information on the study and invited those who were willing to participate. A total of 145 elderly individuals aged 63–92 years, verified with their national identification card, who were willing to provide written informed consent, were enrolled.

The Ethical Committee of the Corporación Ecuatoriana de Biotecnología approved the protocol and the informed consent forms. During meetings at the fieldwork station, each subject was given detailed information on the study objectives and the procedures to be performed. In the instance of a subject having a health condition such as deafness, we used the assistance of a relative to help the subject to understand the study purpose and procedures. After potential participants had had an opportunity to ask questions, written informed consent was obtained. Biochemical analysis of serum or plasma samples was approved by the Tufts – New England Medical Center Institutional Review Board.

Nutritional profile

Individual dietary intake was estimated with a modified 24 h recall–weighing method (Zamora & Valverde, 1983). The

interview was carried out in each household. Each subject was given an explanation on the importance of answering as truthfully and accurately as possible. In order to help the subject recall the previous day, we asked her/him about her/his activities, such as the time of awakening, daytime activities and when he or she went to bed. This approach helped the participants to remember the foods ingested. During the interview, the amounts of food consumed were verified by asking the subject the size of the household measures used to prepare the consumed food. The recall questionnaire was applied once per week to each subject. Household measures and the weights of food most frequently consumed in the neighbourhood were standardised. The questionnaire had two parts: (1) general information on health status, regular daily activities, special diet, type of lipids, use of salt in cooking and at the table, consumption of vitamins, supplements and alcohol; (2) specific information on food ingested at different times of the day (breakfast, lunch and dinner, and between meals). These data were collected with a questionnaire standardised during a pilot phase in a neighbourhood close to those where the present study was conducted. Prepared food and the ingredients used in each preparation were recorded, including their weight. Generally, recipes were obtained for foods served to the entire family, from which the amount of food ingested for each member was calculated. We made adjustments based on the portion he/she effectively consumed from what they recalled. Finally, each food and its ingredients were coded according to United States Department of Agriculture Food Codes and, when available, for specific Latin American Foods (United States Department of Agriculture Agricultural Research Service, 2004). We used release 17, which is no longer available, though release 18 can now be accessed (United States Department of Agriculture Agricultural Research Service, 2006). Foods not included in this file were coded according to the Ecuadorian Table of Foods (Instituto Nacional de Nutrición, 1965).

When available, we used the estimated average requirement (Food and Nutrition Board & Institute of Medicine, 2000; Institute of Medicine, 2000b) to determine the percentage of subjects consuming below or above the recommendation of a nutrient, and the adequate intake when an estimated average requirement was not available. The acceptable macronutrient distribution range (Food and Nutrition Board & Institute of Medicine of the National Academies, 2002) was used to determine adequacy of macronutrient intakes. It is recommended as the best method for assessment of population-based macronutrient adequacy, based on intervention and epidemiological evidence of reducing risk of chronic diseases (Food and Nutrition Board & Institute of Medicine of the National Academies, 2002).

Standard anthropometric measurements were performed including weight, height, knee height, skinfold thickness, and waist circumference for each participant according to the procedures outlined by Gross (1997) as follows. Weight was recorded to the nearest 0.1 kg using a Detecto scale (Detecto®, Webb City, MO, USA) with the least amount of clothes possible in all elderly participants, except one who could not stand on the scale and another one who refused to take her clothes off. The scale was calibrated daily with standard weights. Height was measured in all subjects but one who was unable to stand. A fibreglass tape measure right next to a wood structure was used. Measurements were recorded to the

nearest cm. Knee height was measured in all subjects using a fibreglass tape while the participant was sitting comfortably in a chair, with the knee bent at 90° and with the foot at 90° relative to the leg (Blaum *et al.* 1995). The procedure was repeated and recorded twice, to the nearest cm. Waist circumference was obtained between the border of the right anterior superior iliac crest and the umbilicus. It was recorded twice and measured to the nearest 0.1 cm with a non-stretchable measuring tape. Triceps skinfold was measured with a Caliper® (Beta Technology Incorporated, Cambridge, MD, USA) in a vertical fold at the midline posterior over the triceps muscle, between the acromion and the olecranon, with the elbow extended and the arm relaxed. Two measurements were taken, and recorded to the nearest mm.

Nutrient analysis and clinical chemistries were assessed in serum or plasma samples of a random sub-sample of sixty-five subjects. A 10 ml venous blood sample was drawn from each subject, after an overnight fast, into an EDTA-treated vacutainer tube and a vacutainer tube without anticoagulant. The samples were immediately transported to the laboratory and centrifuged. Serum or plasma was collected in plastic tubes, frozen at -20°C, and then shipped to Boston for analysis. Lipids, glucose, homocysteine, B vitamins, vitamin E, vitamin D, Ca, Se, Zn, Fe and Cu were analysed according to standard procedures used at the Nutritional Evaluation Laboratory of the Jean Mayer US Department of Agriculture Human Nutrition Research Center on Aging at Tufts University as previously described (Meydani *et al.* 1994). Reference ranges for vitamins A, D and E, pyridoxal-5-phosphate (PLP), Fe, Zn and Se were based on those recommended in the *Textbook of Clinical Chemistry* (Tietz, 1986) or by the manufacturer of the kits used to analyse for the nutrients (vitamin B₁₂, folate, Na, K, Ca, P, Mg, Cl), and verified by the Nutritional Evaluation Laboratory at the Jean Mayer US Department of Agriculture Human Nutrition Research Center on Aging, using data generated in healthy elderly people over the last 25 years. Modifications to these recommendations were made for some of the nutrients, based on new reports related to serum level of nutrients and risk for prevention of chronic diseases. These include vitamins B₆ and B₁₂ and folate. Although clinical cut-off points have been set at 20 nmol/l, it has been argued that plasma PLP below 30 nmol/l may be a better indicator of low vitamin B₆ (Driskell, 1994; Bailey *et al.* 1997). The sensitivity and specificity for cut-offs for vitamin B₁₂ concentrations are relatively poor. Although a clinical cut-off point has often been set at < 148 pmol/l (200 pg/ml), vitamin B₁₂ deficiency has been documented with serum concentrations as high as 258 pmol/l (350 pg/ml) (Lindenbaum *et al.* 1994). Therefore, for estimates of population deficiency, the cut-off of 185 pmol/l (250 pg/ml) has been used (Tucker *et al.* 2000). Cut-off points for folate have also varied considerably. Sauberlich (1990) defines plasma levels of > 5 ng/ml as low risk for deficiency, 3–5 as moderate risk and < 3 as high risk. Selhub & Rosenberg (1996) refer to these same cut-off levels as adequate, low and deficient in the 7th edition of *Present Knowledge in Nutrition*. Thus, we used 5 ng/ml as a cut-off point for folate adequacy.

Health profile

An interview was performed using a questionnaire to obtain information on diagnosed non-communicable diseases,

respiratory tract infections, and diarrhoeal diseases within the previous 6 months.

Immunological assessment

Delayed type hypersensitivity (DTH) was assessed using a cell-mediated immunity kit (Multitest CMI; Pasteur-Merieux, Paris, France), a single-use, disposable applicator of acrylic resin with eight heads loaded with a glycerine control and the following seven recall antigens: tetanus toxoid, diphtheria toxoid, *Streptococcus* (group C), *Mycobacterium tuberculosis*, *Candida albicans*, *Trichophyton mentagrophytes* and *Proteus mirabilis*. We followed the product's technical recommendations in applying and evaluating the results, including cleanliness and supervision of the puncture area, time and pressure needed at the application site, and reading of the local reaction at 24 and 48 h after the test was applied. The DTH test was administered on the volar surface of the right arm and evaluated by the same investigator (F. S.) for all subjects. To read the DTH responses, we used a circular ruler calibrated in mm. We measured the vertical and horizontal diameters of induration, and considered the reaction positive when the mean value was ≥ 2 mm. Averages for each individual antigen were calculated and a composite score based on the results of all of the antigens in each subject was determined.

Statistical analysis

Data entry and management were done with Epi-Info software, version 6.04d (CDC, Atlanta, GA, USA). Statistical analyses were performed with SPSS, version 11.5 (Lead Technologies Inc., Haddonfield, NJ, USA; SPSS Inc., Chicago, IL, USA). The mean intake and SD of energy, macronutrients, vitamins and minerals were calculated from the 24 h dietary recall survey at the Jean Mayer US Department of Agriculture Human Nutrition Research Center on Aging, Dietary Assessment and Epidemiology Research Program using the compiled database described earlier. Mean intakes were compared with estimated average requirements or adequate intakes for micronutrients and to the acceptable macronutrient distribution range reference values for macronutrients (Institute of Medicine, 1997, 1998, 2000*a,b*; Food and Nutrition Board & Institute of Medicine, 2000; Food and Nutrition Board & Institute of Medicine of the National Academies, 2002; Food and Nutrition Board, 2004).

Descriptive statistics for anthropometric measurements, BMI, DTH response, and health profile survey responses were calculated globally and by sex. BMI was calculated as the weight in kg divided by height in metres squared (Garrow & Webster, 1985). Weight was classified as normal (BMI 20–24.9 kg/m²), underweight (BMI < 20 kg/m²) and overweight (BMI ≥ 25 kg/m²). Differences in means and percentages were evaluated by Student's *t* test and the χ^2 test, respectively. A linear multiple regression analysis was done to test the correlation between plasma micronutrient concentrations and DTH response at 48 h. The model included age, sex and BMI as confounders. The models for vitamins A and E were also adjusted for cholesterol concentrations. To evaluate the association between dietary micronutrients and serum micronutrient concentrations, a similar model was applied. The energy intake was also included as confounder.

A logistic regression model was developed to evaluate the association between plasma micronutrient concentrations and respiratory and diarrhoeal infection during the previous 6 months. The model included age, sex and BMI as potential confounders. A similar model was used to evaluate the association between dietary micronutrients and respiratory and diarrhoeal infection. The energy intake was also included as confounder. The model for dietary Fe intake also included dietary Ca, vitamin C and fibre as confounders (Fleming *et al.* 1998). Regression model diagnostics were done and the adequacy of covariate functional forms was examined.

Results

Nutritional profile

One hundred and forty-three elderly subjects were enrolled, of whom ninety-six (67%) were female (Table 1). There was no substantially significant difference in weight between men and women (58 (SD 10) v. 53 (SD 9) kg). Female subjects had significantly lower values for height and knee height, and higher triceps skinfold values than men. Of the subjects, 39% had normal BMI, while 9.1% were underweight and 51.7% were overweight. The mean BMI for women was higher than that of men ($P=0.007$). Dietary recall assessments in fifty-two participants showed that most participants consumed less than the recommended estimated average requirement or adequate intake for vitamins A, D, E, K, B₂, B₆ and B₁₂, folate, Zn, Ca, Mg and K. Although the mean total energy intake was lower than generally recommended (7840–9226 kJ) (Food and Nutrition Board & Institute of Medicine of the National Academies, 2002), there was a wide range of reported intakes (899–16 677 kJ). Furthermore, in the absence of information on the level of physical activity, it is difficult to determine adequacy of energy intake. Carbohydrate consumption was higher than recommended in 86.5% of subjects, while protein and fat intakes were low in 53.8 and 92.3%, respectively (Table 2). Laboratory micronutrient analysis in sixty-five subjects revealed that about 50% of these elderly Ecuadorians had low concentrations of plasma vitamin B₁₂, Zn and Fe, 30% had low PLP (vitamin B₆) and 19% had low folate and vitamin D (Table 3).

Multiple linear regression analysis in a sub-group of subjects for whom both dietary intake and serum nutrient concentrations were available ($n=52$ and $n=65$, respectively) showed a positive correlation between dietary Zn and PLP levels and their corresponding serum level ($P<0.01$ and $P<0.05$, respectively). A significant correlation between dietary Fe and serum Fe levels was found ($P=0.007$), but the significance was lost after correcting for sex, age, BMI, energy intake, Ca, vitamin C and fibre levels (Fleming *et al.* 1998). The dietary Fe levels were split into haeme and non-haeme Fe, but no significant correlation between serum Fe levels and either dietary form was observed.

About 20% of the subjects had high triacylglycerol and VLDL concentrations, while 19% had low HDL concentrations; however, the LDL:HDL ratio was within the reference range. Consistent with the observation of low B vitamin concentrations, homocysteine concentrations were higher than the reference value in 25% of the subjects (Table 4).

Health profile

The most frequent non-communicable diseases reported by the subjects were hypertension (19%), arthritis (19%), heart disease (12%) and osteoporosis (10%). Other medical problems included impaired vision, cerebrovascular accident, and type 2 diabetes (6, 5 and 4%, respectively). The most common infectious diseases recalled during the previous 6 months were upper respiratory infections (54%), diarrhoea (21%) and bronchitis (7%). Approximately 47% of those with infections had visited a physician or hospital emergency room, and 96.2% had asked for help from relatives or friends. Women suffered more frequently from diarrhoea ($P<0.02$) and sought care at emergency rooms more often than men ($P<0.01$).

Logistic regression analysis in a sub-group of subjects for whom dietary and serum nutrient levels were available ($n=52$ and $n=65$, respectively) showed no significant correlation between plasma nutrient concentration and respiratory infections or diarrhoea. Similar analysis showed that only dietary vitamin C was significantly correlated with respiratory infections (relative risk 1.101 (95% CI 1.004, 1.208)). There was no significant association between dietary nutrients and diarrhoea. These results, however, need to be interpreted with caution due to the low

Table 1. Anthropometric measurements in elderly Ecuadorians
(Mean values and standard deviations)

Variable	Men ($n=47$)		Women ($n=96$)		<i>P</i>
	Mean	SD	Mean	SD	
Weight (kg)	58	10	53	9	0.09
Height (cm)	155	8	143	4	<0.0001
Knee height (cm)	40	2	35	3	<0.0001
Waist (cm)	86	8	88	12	0.327
Triceps skinfold (mm)	6	2	11	4	<0.0001
BMI (kg/m ²)	24	3	26	4	0.007
Underweight (BMI < 20 kg/m ² ; %)	13		7		0.285
Normal (BMI 20–24.9 kg/m ² ; %)	47		36		0.190
Overweight (BMI ≥ 25 kg/m ² ; %)	40		57		0.058
High waist circumference*	4.3		44.9		<0.0001

* Men > 102 cm; women > 88 cm.

Table 2. Dietary macronutrient and micronutrient intake of elderly Ecuadorians (*n* 52) (Mean values and ranges)

Nutrient	Mean	Range	EAR, AI or AMDR reference values	Below reference range (%)	Above reference range (%)
Energy (kJ)	5862	899–16 677			
Total fat (% energy)	14.1	0.71–34.1	25–35*	92.3	
Carbohydrate (% energy)	76.7	47–100	45–65*		86.5
Protein (% energy)	10.3	0.76–20.81	10–30*	53.8	
Total vitamin A (μg RE)	406	0.1–5031	500–625†	88.5	
Vitamin D (calciferol) (μg)	1	0.00–12	10–15‡	98.1	
Total vitamin E activity (mg)	2	0.04–5	12†	100	
Vitamin K (phylloquinone) (μg)	32	0.1–226	90–120‡	92.3	
Riboflavin (mg)	0.78	0.01–2.8	0.9–1.1†	67.3	
Vitamin B ₆ (mg)	1	0.1–3	1.3–1.4†	53.9	
Folate (μg)	359	2–859	320†	50.0	
Vitamin B ₁₂ (μg)	1.4	0.00–30	2.0†	82.7	
Ca (mg)	238	2–1667	1200‡	98.1	
P (mg)	553	12–1638	580†	61.5	
Mg (mg)	174	5–367	265–350†	92.3	
Fe (mg)	10	0.1–23	5.0–6.0†	19.2	
Zn (mg)	5	0.1–14	6.8–9.4†	76.9	
Cu (mg)	1	0.1–3	700†	34.6	
Se (μg)	64	0.4–163	45†	28.9	
Na (mg)	2686	244–11 654	1500‡	30.8	69.2
K (mg)	1651	83–4548	4700‡	100	

EAR, estimated average requirement; AI, adequate intake; AMDR, acceptable macronutrient distribution range; RE, retinol equivalents.

* AMDR (Food and Nutrition Board & Institute of Medicine of the National Academies, 2002).

† EAR (Food and Nutrition Board & Institute of Medicine, 2000; Institute of Medicine, 2000b).

‡ AI; EAR not available (Institute of Medicine, 2000b; Food and Nutrition Board, 2004).

number of subjects for whom all data were available, as well as the recall nature of data collected for infection.

Immunological assessment

DTH skin tests were performed on fifty-two randomly selected participants. The majority (69%) had positive responses to tuberculin, 41% to diphtheria and less than 40% to the other antigens at 48 h. The composite score and the mean number of positive responses were similar at 24 and 48 h. Similar patterns were seen in both women and men. The response to tetanus toxoid was significantly higher at 48 h than at 24 h, whereas the response to *Proteus* was significantly lower at 48 h than at 24 h (Table 5). Logistic regression analysis in a sub-group of subjects for whom dietary and serum nutrient concentrations as well as DTH were available (*n* 52) showed a positive correlation between plasma PLP concentration and DTH score at 48 h after correcting for sex, age and BMI ($P=0.002$), and a positive correlation between dietary intake of vitamin B₆ ($P=0.005$), vitamin A ($P=0.02$), Fe ($P=0.02$) and Cu ($P=0.01$) after correcting for energy intake, BMI, sex and age. There was also a trend for correlation between dietary vitamin D levels and DTH ($P=0.057$). These results, however, need to be interpreted with caution because of the limited number of subjects for whom all values were available.

Discussion

This sample of elderly Ecuadorians had multiple nutritional deficiencies, as assessed by both dietary recall and laboratory

analyses. This population had lower than recommended dietary intake of vitamins A, D, E, B₂, B₆ and B₁₂, and folate, Zn, Ca and Mg. Although the mean total energy intake appeared to be low, total energy intake varied widely. Furthermore, in the absence of information on physical activity, it is difficult to determine adequacy of energy intake. In addition, despite several quality-control measures included in the collection of these dietary recall data, the possibility that some subjects under-reported their intake could not be ruled out. Notably, carbohydrate intake was higher than recommended (Food and Nutrition Board & Institute of Medicine of the National Academies, 2002) in the majority of subjects. In addition, 69.2% had higher than recommended intake of Na. It is, however, difficult to obtain accurate estimates of salt intake from the dietary intake data. Thus, these results should be confirmed by 24 h urinary Na excretion analysis. Although the proportion of overweight and obese elderly people in this peri-urban, poor community was less than that in the USA (Flegal *et al.* 1998), the fact that 51.7% had BMI greater than 25 kg/m², in the presence of low concentrations of several micronutrients, suggests that these elderly Ecuadorians suffer from the increasingly common double burden of diseases associated with nutrition transition in less-developed countries and consumption of diets with poor quality.

We did not specifically evaluate the metabolic syndrome in the present study, but based on triacylglycerol and HDL levels, approximately 20% of these Ecuadorian elders met the proposed limit of these indicators of the metabolic syndrome (Hall *et al.* 2003). This proportion is similar to those reported in overweight American adults (Park *et al.* 2003). Of the women, 45% had waist circumference that was

Table 3. Blood vitamin and trace element concentrations in elderly Ecuadorians (*n* 65)
(Mean values, standard deviations and ranges)

Variable	Mean	SD	Reference range	Reference method	Below reference range (%)
Vitamin A ($\mu\text{g/l}$)	57.8	15.0	30.0–90.0*	Bieri <i>et al.</i> (1979)	3
Vitamin D (ng/ml)	19.03	5.6	15–38*	Anonymous (2001)	19
Vitamin E ($\mu\text{g/l}$)	11 621.1	330.0	500.0–1800.0*	Bieri <i>et al.</i> (1979)	0
PLP (nmol/l)	42.49	23	30–117†	Hamfelt (1962); Maruyama & Coursin (1968); Sundaresan & Coursin (1970); Reynolds (1987)	31
Vitamin B ₁₂ (pg/ml)	323.44	256	250–1200†	‡	43
Folate (ng/ml)	6.57	2	5–30†	‡	18
Cu ($\mu\text{g/l}$)	128.11	22.0	55.0–175.0*	Dawson <i>et al.</i> (1968)	0
Fe ($\mu\text{g/l}$)	64.34	34.0			39
Females			50.0–170.0*	Olson & Hamlin (1969)	
Males			65.0–175.0*		
Zn ($\mu\text{g/l}$)	70.38	13.0	70.0–150.0*	Smith <i>et al.</i> (1979)	50.0
Se (ng/ml)	142.24	17.81	33–180*	Brumbaugh & Walther (1991)	0
Na (meq/l)	141.93	3	135–150§	Anonymous (1988)	0
K (meq/l)	4.69	0.5	3.5–5.3§	Tietz (1986); Anonymous (1988)	0
Ca (mg/l)	9.34	0.3	8.3–10.2§	Michaylova & Ilkova (1971)	0
P (mg/l)	3.30	0.4	2.3–4.7§	Daly & Ertingshausen (1972)	1.5
Mg (mg/l)	1.70	0.2	1.3–2.6§	Ingman & Ringbom (1966); Gindler & Helth (1971)	0
Cl (mmol/l)	105.64	3	96–110§	Anonymous (1988)	0

PLP, pyridoxal-5-phosphate.

* Tietz (1986).

† Tietz (1986), modified; see p. 847.

‡ Quantafase II B₁₂/Folate Radioassay (Bio-Rad Laboratories Inc., Hercules, CA, USA).

§ Reference ranges recommended by the kit used for analysis or as indicated (Cobas Mira Chemical System; Roche Diagnostics, Nutley, NJ, USA).

higher than the limit value (Hall *et al.* 2003), whereas only 4.3% of men had values above the limit, suggesting that elderly women in Quito are at higher risk for the metabolic syndrome than men.

A substantial proportion of the subjects had low plasma Zn and Fe concentrations. Consistent with this, 76.9% of the elderly had a low dietary intake of Zn due to the limited consumption of food of animal origin in this population. A positive correlation between dietary intake and plasma Zn concentration was observed even after adjustment for Cu. Although only 19% of

subjects had low daily intake of total Fe according to the dietary recall survey, most of the consumed foods are grains and vegetables, which are not a good source of bioavailable Fe. Similar results have been found in Ecuadorian children (Sempertegui *et al.* 1996). In addition, high consumption of phytates from barley and other local cereals, which reduces mineral absorption, has been reported in this region (Holt & Brown, 2004). Logistic regression analysis indicated significant positive correlation between dietary Fe and serum Fe levels, but the significance was lost after correcting for sex, age, BMI, energy

Table 4. Fasting blood lipid, glucose and homocysteine concentrations in elderly Ecuadorians (*n* 65)
(Mean values and standard deviations)

Variable	Total		Above upper limit (%)	Reference range	Reference method
	Mean	SD			
Cholesterol (mg/l)	190.0	32.0	2	130.0–278.0	Allain <i>et al.</i> (1974)
Triacylglycerol (mg/l)	167.0	83.0	21	41.0–204.0	Trinder (1969); Spayd <i>et al.</i> (1978); Esders & Michrina (1979)
HDL (mg/l)	52.0	10.0	0*	37.0–82.0	Trinder (1969); Allain <i>et al.</i> (1974); Spayd <i>et al.</i> (1978); Esders & Michrina (1979); Warnick <i>et al.</i> (1982)
LDL (mg/l)	106.0	29.0	2	62.0–189.0	Friedewald <i>et al.</i> (1972)
VLDL (mg/l)	34.0	15.0	20	5.0–31.0 or 3.0–34.0	Friedewald <i>et al.</i> (1972)
LDL:HDL	2	0.6	0	1.1–5.9 or 0.8–4.9	–
Glucose (mg/l)	86.0	14.0	3	64.0–112.0	Barthelmaï & Czok (1962)†
Homocysteine (mol/l)	12	4	25	5.5–15.0	Araki & Yoshiyasu (1987)

* 19% Below limit.

† As modified by Roche Diagnostic Systems, Inc. (Nutley, NJ, USA); technical procedure number 44 557 (1985).

Table 5. Delayed type hypersensitivity response to multitest CMI (Pasteur-Merieux, Paris, France) at 24 and 48 h in elderly Ecuadorians (*n* 50)
(Mean values and standard deviations)

Antigen	Diameter of induration (mm)				Positive (%)	
	24 h		48 h		24 h	48 h
	Mean	SD	Mean	SD		
Tetanus toxoid	0.70	1.5	1.43*	2.3	20	30
Diphtheria toxoid	2.02	2.3	2.30	2.7	47	41
<i>Streptococcus</i>	0.70	1.2	0.71	1.2	24	20
Tuberculin	2.90	2.4	3.30	2.5	65	69
Glycerin	0.02	0.14	0.02	0.14	0	0
<i>Candida</i>	0.62	1.0	0.92	1.3	12	28
<i>Trichophyton</i>	0.18	0.52	0.23	0.58	2	4
<i>Proteus</i>	2.25	1.9	0.88*	1.2	51	22
Total diameter of induration	9.68	6.09	9.94	7.03		
Total number of positive responses	2.20	1.41	2.12	1.69		

Mean value was significantly different from that at 24 h * $P < 0.01$.

intake, Ca, vitamin C and fibre levels (Fleming *et al.* 1998). The dietary Fe levels were split into haeme and non-haeme Fe, but no significant correlation between serum Fe levels and either dietary form was observed. Given the small number of subjects in whom both measurements were available, it is likely that a larger number of subjects are needed to observe significant correlation when confounders are included. Alternatively, since the diet of the elderly is low in Zn and B₆, and they have low animal protein consumption, we suggest that the low Fe status is mainly due to the poor quality of their diet; however, the elderly Ecuadorians might have large day-to-day variation in haeme intake not well captured in a few days represented by dietary records.

Of these elders, 43% had low plasma vitamin B₁₂ concentrations and 18% had low folate. This could be related to low dietary intake, although vitamin B₁₂ deficiency might also have been secondary to diminished absorption due to gastric atrophy associated with ageing (Wolters *et al.* 2004). Approximately 25% of our cohort had high homocysteine concentrations. Plasma homocysteine is related to the cellular availability of both vitamin B₁₂ and folate (Wolters *et al.* 2004). The plasma PLP was low in 31% of these elders, which might be due to poor consumption of vitamin B₆ (53% had below recommended dietary intake of B₆). This is further supported by the observation that dietary vitamin B₆ levels positively correlated with serum level of PLP ($P < 0.05$). To our surprise, and despite numerous sunny days throughout the year, 19% of these Ecuadorian elders had low vitamin D concentrations. It is possible that those subjects who were deficient tended to stay inside their households most of the time.

About 20% of these elders had high triacylglycerol and VLDL concentrations. This could be related to the high intake of carbohydrates, which are converted into lipids in the liver. A similar percentage had low HDL concentrations. Underlying dyslipidaemias in this population may contribute to the self-reported moderate levels of hypertension and heart disease (19 and 12%, respectively) of the studied subjects. These figures are lower than those reported for the US population (Lethbridge-Cejku *et al.* 2004; National Center for Health Statistics, 2004), but this could be related to under-reporting and infrequent visits to health providers.

DTH reaction to seven recall antigens showed an average diameter of induration, which was about one-third of that reported in developed countries (Meydani *et al.* 1997). Although a positive association was found only between plasma PLP and dietary PLP, Fe, Cu, vitamin A, and DTH, and between dietary vitamin C and respiratory infection, the impaired cellular immunity might also be due to other critical deficiencies such as that of Zn, protein (Peppersack *et al.* 2001) and/or metabolic changes related to being overweight. However, a larger sample size than that of the present study as well as interventional clinical trials would be needed before these associations can be established. Several factors contribute to resistance to infectious diseases. However, immune response is an important determinant. Thus, the low immunological status of these elderly Ecuadorians could contribute to the high frequency of upper respiratory and diarrhoeal infections reported (54 and 21%, respectively) during the previous 6 months. However, these figures are based on a 6-month recall survey focused on infectious diseases and thus should be interpreted with caution, given potential problems with recall bias.

In conclusion, micronutrient deficiencies and inadequate consumption of protein and fat are common in elderly Ecuadorians and might contribute to their low immunological status and high prevalence of infectious diseases. These findings need to be confirmed in larger cross-sectional and interventional studies. Nevertheless, this initial report of nutrition and health status in elderly Ecuadorians suggests that an argument similar to those used in support of increasing children's nutrition in the less-developed countries could begin to be made for elderly people in the less-developed countries. Furthermore, these results suggest that nutrition intervention to address both under- and over-nutrition in older individuals in Ecuador may play a crucial role in preventing death and disability, promoting successful ageing, and reducing the burden on the limited healthcare resources of the country.

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