Original Paper

Nutrition & Metabolism

Ann Nutr Metab 1998;42:266–273

Received: August 29, 1997
Accepted: April 15, 1998

Production of Interleukin-6 in Cultures of Peripheral Blood Mononuclear Cells from Children with Primary Protein-Calorie Malnutrition and from Eutrophic Controls

Inés Malavé[a]
María A. Vethencourt[a]
Rita Chacón[a]
Damelis Quiñones[a]
Carlos Rebrij[b]
Gonzalo Bolívar[b]

[a] Instituto Venezolano de Investigaciones Científicas,
[b] Centro Clínico de Recuperación Nutricional 'Menca de Leoni', Caracas, Venezuela

Abstract

The spontaneous as well as mitogen-induced in vitro production of interleukin-6 (IL-6) was studied in cultures of peripheral blood mononuclear cells (PBMC) from 14 children with marginal protein-energy malnutrition, 43 children with definite protein-energy malnutrition and 38 eutrophic controls of similar age, sex, race and socioeconomical condition. PBMC were cultured without added mitogen or stimulated with either lipopolysaccharide (LPS) or phytohemagglutinin (PHA). After 48 h incubation, cell-free culture supernatants were collected and stored at –70°C. The amount of IL-6 in the supernatants was determined by a specific bioassay based on the proliferation of B9 hybridoma cells using human rIL-6 as standard. The mean level of IL-6 was significantly increased in supernatants from nonstimulated PBMC cultures from definitely malnourished children as compared with that observed in those of the controls. Stimulation with either LPS or PHA induced a rise in cytokine bioactivity in the supernatants of PBMC cultures from the different nutritional groups tested. Interestingly, IL-6 was significantly increased in the supernatants of PHA-stimulated cultures from malnourished children as compared with those of the controls.

Key Words
Interleukin-6
Protein-energy malnutrition
Cachexia
Th2 cytokines
Malnutrition

Introduction

Children with primary protein-energy malnutrition show increased susceptibility to infections [1, 2]. In turn, infections aggravate the course of primary nutrient deficiency, increasing weight loss and wasting [1–3]. This reiterative cycle of malnutrition and infections with subsequent worsening of the nutritional condition is the main cause of morbidi-

KARGER
Fax + 41 61 306 12 34
E-Mail karger@karger.ch
www.karger.com

0250–6807/98/0425–0266$15.00/0

Accessible online at:
http://BioMedNet.com/karger

Inés Malavé
Instituto Venezolano de Investigaciones Científicas
Apartado 21827, Caracas 1020A (Venezuela)
Tel. +58 2 504 1161/1288, Fax +58 2 504 1086
E-Mail imalave@medicina.ivic.ve

Received: August 29, 1997
Accepted: April 15, 1998

ty and mortality in children from underdeveloped countries [3].

Interleukin-6 (IL-6) is a cytokine with a broad range of biological functions. Among them, it is an important inducer of the acute-phase response, stimulates terminal B lymphocyte differentiation into antibody-secreting plasma cells, promotes the growth of hybridomas, plasmocytomas and myeloma cell lines, activates the proliferation and differentiation of cytotoxic T lymphocytes and induces osteoclastogenesis and bone resorption [reviewed in ref. 4]. IL-6 also exerts metabolic functions, playing a role in the development of the cachectic syndrome as demonstrated in tumor-bearing hosts [4]. It has also been shown that IL-6 inhibits lipoprotein lipase in adipocytes [5] increasing in parallel hepatic lipogenesis, thus contributing to the paradoxical hypertriglyceridemia observed in cachexia [6].

Infectious stimuli are important inducers of IL-6 synthesis [7]. It should also be stressed that even in the absence of acute overt infections, malnourished individuals are exposed to frequent and intense stimulation by microbial antigens which could trigger the production of IL-6. In turn, this cytokine could contribute to mediate the marked deleterious effect of infections on primary malnutrition. On the other hand, alterations in the capacity to produce some cytokines have been reported in malnourished individuals, although the findings have not been consistent [8–11]. In view of the above, in the present work we have studied the production of IL-6 in cultures of peripheral blood mononuclear cells (PBMC) from children with definite protein-energy malnutrition, children at nutritional risk (marginal malnutrition) and from a group of well-nourished controls.

Material and Methods

Study Population

A group of 96 children from either sex (mean age ± SE 28.5 ± 2.3 months) was studied. This population included 44 children with definite protein-energy malnutrition, 14 children diagnosed as bearing subclinical or marginal malnutrition (at nutritional risk), and 38 apparently eutrophic controls according to clinical and anthropometrical evaluation matched for age, sex, race and socioeconomical status with the malnourished groups. Only children without manifestations of overt infections at the time of sampling were included in the study. The socioeconomical condition of each individual was assessed by the procedure of Graffar as modified by Mendez Castellano and Mendez [12]. All the children studied belonged to the status V of Graffar's modified classification living in conditions of critical poverty and extreme poverty. Anthropometrical evaluation of the nutritional condition was performed using a combination of indicators [13]. Among them, weight-for-age (W/A), height-for-age (H/A), and weight-for-height (W/H) were compared with the WHO standard curves. Anthropometrical indicators were also converted into standard deviation units of the NCHS/CDC reference population or Z-scores as recommended by the WHO [14], using an anthropometrical statistical package from the CDC [15].

The control group comprised children with indicators above the 10th percentile (>−1.28 SD) of the reference population [13, 16]. Children with a W/A index equal or below the 10th percentile (≤−1.28 SD) and above the 3rd percentile (>−1.9 SD) of the reference population constituted the group with marginal or subclinical malnutrition [13, 17]. Definite protein-energy malnutrition was diagnosed when the Z-score was equal or below the 3rd percentile (≤−1.9 SD) of the reference population [13, 14, 17, 18]. The whole group of children with definite malnutrition was further divided into two subgroups: (a) the first bearing mild definite malnutrition (with Z-scores <−1.9 SD and >−3 SD) and a second bearing moderate and/or severe protein-energy malnutrition and defined by Z-scores ≤−3 SD [14, 17, 18]. The existence of nutritional edema was considered as an indicator of severe malnutrition as stated by Bengoa et al. [19].

The percentage distribution of the deficit in W/A, H/A and W/H among malnourished children is shown in table 1. Low H/A (stunting) was more prevalent than low W/H (wasting) in the definite malnourished group. This is in coincidence with previous observations in Latin America where chronic nutritional deficiency was more frequent than acute malnutrition [re-

Table 1. Percentage distribution of the deficit in W/A, H/A and W/H in children with either marginal or definite protein-energy malnutrition

Nutritional group	Children with deficit, %		
	W/A	H/A	W/H
Marginal protein-energy malnutrition (cutoff points < the 10th percentile or −1.28 SD)	100	43	63
Definite protein-energy malnutrition (cutoff points < the 3rd percentile or −1.90 SD)	98	89	57

viewed in ref. 18]. However, low W/H indicating acute nutritional deficiency occurred with a relatively high frequency in children with either marginal or definite malnutrition. Presumably diminished food availability due to an economical crisis affecting a country, which has an especially marked impact on populations of low socioeconomical status, could determine a situation of nutritional risk, leading to marginal protein-energy malnutrition. In addition, acute nutritional deficiency could aggravate chronic malnutrition in an important proportion of definitely malnourished children. Low W/A (weight deficit) was observed in all children with marginal and in 98% of children with definite protein-energy malnutrition (except for 1 case with marked nutritional edema; table 1). The high prevalence of low W/A in the malnourished population is in agreement with the fact that it reflects the addition of stunting and wasting [18], and with the high sensitivity of the W/A indicator to nutritional deficiency [17, 20].

The presence or absence of clinical overt infections was assessed by anamnesis, clinical examination and laboratory data, e.g. hematological tests which included leukocyte count and formula as well as erythrocyte sedimentation rate, urine and stool analyses and radiological examination when necessary. Patients with signs or symptoms of overt infections were discarded from the study. Following screening for the absence of clinical overt infections, and to avoid the effect of nutritional rehabilitation, malnourished children were sampled within the first 36 h upon admission to the Center for Nutritional Recovery 'Menca de Leoni' from Caracas, Venezuela, or during the first consultation to the outpatients clinics of that center. Controls were sampled at dispensaries for the control of healthy children located in slum areas of Caracas. The consent of at least one of the parents was obtained before blood sampling in each child. The study was approved by the Ethical Committees of the Instituto Venezolano de Investigaciones Científicas', and of the Center for Nutritional Recovery 'Menca de Leoni'.

Cell Cultures

Samples of peripheral blood were collected into heparinized syringes and PBMC isolated by density gradient centrifugation on Ficoll-Hypaque (D = 1,077). Following 30 min of centrifugation at 400 *g*, cells remaining at the interphase were collected, washed and resuspended at a concentration of 2×10^6/ml in medium RPMI 1640 supplemented with 2 m*M* L-glutamine, 5% fetal calf serum and 1% antibiotic-antimycotic mixture (all from Grand Island Biological Laboratories, Grand Island, N.Y., USA; complete medium). One hundred microliters of the cell suspensions were distributed into the wells of flat-bottomed microculture plates (Linbro, Flow Laboratories, McLean, Va., USA) and cultured in a final volume of 200 µl either without mitogen or stimulated with phytohemagglutinin (PHA) at a concentration of 5 µg/ml or with lipopolysaccharide (LPS) from *Escherichia coli* 055:B5 (both mitogens from Sigma, St. Louis, Mo., USA), also at the concentration of 5 µg/ml. Following 48 h of incubation at 37°C in a humid atmosphere of 5% CO_2 in air, culture supernatants were collected and stored at −70°C for later determination of IL-6.

Quantitation of IL-6

The concentration of IL-6 in the culture supernatants was assessed by a specific bioassay based on the proliferation of the IL-6-dependent B9 hybridoma cell line [21, 22]. Briefly, dilutions of the supernatants to be tested were distributed in duplicate in 96-well microculture plates (Linbro). To build the standard curve, serial twofold dilutions of human rIL-6 (Genzyme, Cambridge, Mass., USA), ranging from 0.39 to 800 pg/ml, were included in each plate. Ten thousand B9 cells

suspended in complete medium were added to each well and the plates incubated at 37 °C for 72 h in a humid atmosphere of CO_2 in air. Proliferation of B9 cells was measured using the colorimetric 3(4,5-dimethylthiazol-2-yl)2,5-diphenyltetrazolium bromide (Sigma) assay described by Mosmann [23]. After addition of 20 µl of this assay solution (at a concentration of 5 mg/ml) to each well, the plates were further incubated for 4 h. Subsequently, 100 µl of a solubilizing solution consisting of 20% SDS (Sigma) prepared in water/N,N-dimethylformamide (1:1; Sigma), pH 4.7, were added, and the plates kept overnight in the darkness. Absorption was measured at 570 nm, and the data calculated by interpolation of the mean optical density obtained with each sample on a standard curve of human rIL-6.

Statistical Analysis

Data are presented as medians and interquartile ranges. Statistical comparison was performed by the nonparametric rank sum two-sample (Mann-Whitney) test.

Results

IL-6 bioactivity was significantly higher in the supernatants of cultures incubated without mitogen from definitely malnourished children than from controls. Children with marginal malnutrition only showed moderately higher levels of IL-6 in the supernatants of nonstimulated cultures than controls, although the difference between these groups was not significant (table 2). It should be mentioned in this regard that spontaneous synthesis of IL-6 in cultures of PBMC has been first described by Aarden et al. [21].

As expected, stimulation with LPS markedly increased IL-6 bioactivity in the supernatants of PBMC cultures from children of the three nutritional groups tested. Even though the concentration of the cytokine tended to be higher in culture supernatants from children with definite malnutrition than from those with marginal malnutrition or from the eutrophic controls, the differences between the groups were not significant (table 2).

Stimulation with PHA induced a rise in the bioactivity of IL-6 in the supernatants of PBMC cultures from the different nutritional groups tested (table 2). Interestingly, IL-6 bioactivity was significantly higher in supernatants of PHA-stimulated PBMC cultures from the group of children with definite protein-energy malnutrition than in supernatants of PHA-stimulated cultures from controls (table 2).

When children with definite malnutrition were analyzed according to the severity of the nutritional deficiency, IL-6 bioactivity was increased in the supernatants of nonstimulated PBMC cultures from the subgroup of children with mild malnutrition and even from the subgroup of moderately plus severely malnourished patients as compared with the eutrophic controls (table 3). Likewise, the concentration of IL-6 was higher in supernatants of PHA-stimulated PBMC cultures from either mildly or moderately plus severely protein-energy malnourished children than in eutrophic controls (table 3). Upon stimulation with PHA, IL-6 was higher in culture supernatants from children with mild than from children with moderate and severe malnutrition, although the difference between these groups was not significant (table 3).

Discussion

This study evidences that the spontaneous production of IL-6 was higher in nonstimulated PBMC cultures from children with definite protein-energy malnutrition than from eutrophic controls. Interestingly, significantly higher IL-6 bioactivity was also found in the supernatants of PHA-stimulated cultures from malnourished children as compared with those from the controls.

These results are in agreement with a recent work demonstrating that the production

Table 2. Concentration of IL-6 in the supernatants of PBMC cultures from children with marginal malnutrition, children with definite protein-energy malnutrition and from their eutrophic controls

Group No.	Nutritional condition	Culture stimulus	Concentration of IL-6, ng/ml		Significance
			median	interquartile range	
1	controls	–	31.85	17.57–82.50	1 vs. 2, NS 1 vs. 3, p = 0.021 1 vs. 4, p = 0.037 1 vs. 7, p = 0.023
2	marginal malnutrition	–	53.81	30.70–75.67	2 vs. 3, NS 2 vs. 5, p = 0.054 2 vs. 8, NS
3	definite malnutrition	–	66.55	26.17–122.10	3 vs. 6, p = 0.032 3 vs. 9, p = 0.014
4	controls	LPS	75.05	56.41–125.80	4 vs. 5, NS 4 vs. 6, NS
5	marginal malnutrition	LPS	87.08	47.53–124.60	5 vs. 6, NS
6	definite malnutrition	LPS	127.20	51.07–199.20	
7	controls	PHA	60.73	28.61–110.50	7 vs. 8, NS 7 vs. 9, p = 0.032
8	marginal malnutrition	PHA	63.42	44.77–117.50	8 vs. 9, NS
9	definite malnutrition	PHA	88.58	46.58–172.70	

Statistical analysis was performed by the nonparametric rank sum two-sample (Mann-Whitney) test (two-tailed).

of IL-6 is increased in cultures of PBMC from aged individuals with protein-energy malnutrition due to chronic noninfectious diseases [24]. In addition, in a previous study, we have found that the proportion of sera with detectable levels of serum IL-6 (≥15 pg/ml) was higher in malnourished than in control children, both groups being without associated clinical infections [25].

Important differences in the procedure and experimental design could explain the apparent disagreement between the results of the present work and those obtained by Doherty et al. [26]. In fact, they performed longitudinal testing of IL-6 production in response to LPS stimulation in whole-blood cultures obtained from undernourished individuals before and after nutritional rehabilitation [26], whereas the cytokine concentrations in PBMC cultures from malnourished children were not compared to those of well-nourished matched controls. Furthermore, in the above-men-

ultures from children with
lnutrition and from their

/ml	Significance
ange	
	1 vs. 2, NS
	1 vs. 3, p = 0.021
	1 vs. 4, p = 0.037
	1 vs. 7, p = 0.023
	2 vs. 3, NS
	2 vs. 5, p = 0.054
	2 vs. 8, NS
	3 vs. 6, p = 0.032
	3 vs. 9, p = 0.014
	4 vs. 5, NS
	4 vs. 6, NS
	5 vs. 6, NS
	7 vs. 8, NS
	7 vs. 9, p = 0.032
	8 vs. 9, NS

sum two-sample (Mann-

between the results of
hose obtained by Do-
they performed longi-
roduction in response
whole-blood cultures
rished individuals be-
al rehabilitation [26],
centrations in PBMC
ied children were not
l-nourished matched
in the above-men-

encourt/Chacón/
brij/Bolívar

Table 3. Concentrations of IL-6 in the supernatants of PBMC cultures from children with either mild or moderate/severe malnutrition and from their eutrophic controls

Group No.	Nutritional condition	Culture stimulus	Concentration of IL-6, ng/ml		Significance
			median	interquartile range	
1	controls	–	31.85	17.57–82.50	1 vs. 2, p = 0.101; p < 0.05[a]
					1 vs. 3, p = 0.022
					1 vs. 4, p = 0.023
2	mild malnutrition	–	63.86	25.73–97.73	2 vs. 3, NS
					2 vs. 5, p = 0.079; p < 0.05[a]
3	moderate/severe malnutrition	–	63.02	24.88–125.20	3 vs. 6, p = 0.079; p < 0.05[a]
4	controls	PHA	60.73	28.61–110.50	4 vs. 5, p = 0.053
					4 vs. 6, p = 0.064; p < 0.05[a]
5	mild malnutrition	PHA	98.37	55.78–611.10	5 vs. 6, NS
6	moderate/severe malnutrition	PHA	82.79	44.29–164.90	

Statistical analysis was performed by the nonparametric rank sum two-sample (Mann-Whitney) test (two-tailed).

[a] One-tailed test.

tioned study, IL-6 was not determined in supernatants of cultures incubated without mitogen or cultures stimulated with PHA [26].

Even though IL-6 is mainly produced by monocytes [7], secretion of this cytokine from other cell sources, e.g. T and B lymphocytes, basophils, fibroblasts and endothelial cells, has been reported [7]. Since IL-6 produced by T lymphocytes is mostly synthesized by TH2 cells [27–29], its significant increase in response to a T cell mitogen like PHA, observed here in PBMC cultures from children with protein-energy malnutrition, could suggest a predominant proportion and/or response of TH2 cells in this population group. Such a fact might be due to the presence of increased antigenic load in the malnourished individuals [3] capable to bias T lymphocytes towards TH2 differentiation [28, 30], and by a higher resistance of TH2 cells to apoptosis [31] as that which could be caused by the nutritional deficiency. On the other hand, it has been shown that IFN-γ exerts an inhibitory effect on the synthesis of IL-6 by macrophages [32]. Thus, the depressed production of IFN-γ previously found in protein-energy malnutrition [8] could result in an increased IL-6 synthesis mediated by cells of the monocyte/macrophage series present in the PBMC cultures. In addition, IL-4 stimulates the synthesis of IL-6 by B lymphocytes [33]. High levels of serum IL-4 have been found in malnourished children [34], suggesting an increased IL-4 production by T lymphocytes. Such a fact could also contribute to the raised IL-6 bioactivity observed here in the supernatants of PHA-stimulated cultures from the nutritionally deficient group.

17 Pirela M: Analisis de mortalidad y predicción de riesgo de morir a partir de indicadores antropométricos en niños menores de 5 años de una población rural de Guatemala. Facultad de Ciencias Médicas, Universidad de San Carlos, Guatemala; thesis, publication INCAP, T-441, 1988.
18 Keller W, Fillmore CM: Prevalence of protein-energy malnutrition. World Health Stat Q 1983;36:129–140.
19 Bengoa JM, Jelliffe DB, Pérez C: Some indicators for a broad assessment of the magnitude of protein-calorie malnutrition in young children in population groups. Am J Clin Nutr 1959;7:714–719.
20 Henriquez de Paredes G, López de Blanco ME, Hernández de Valera Y: Análisis antropométrico en la evaluación de la desnutrición aguda en pre-escolares. Arch Ven Puer Ped 1982;45:3–17.
21 Aarden LA, De Groot ER, Shaap LO, Landsdorp PM: Production of hybridoma growth factor by human monocytes. Eur J Immunol 1987;17: 1411–1416.
22 Helle M, Boeije L, Aarden LA: Functional discrimination between interleukin 6 and interleukin 1. Eur J Immunol 1988;18:1535–1540.
23 Mosmann T: Rapid colorimetric assay for cellular growth and survival: Applications to proliferation and cytotoxicity assays. J Immunol Methods 1983;65:55–63.
24 Cederholm T, Wretlin B, Delstrom KJ, Andersson B, Enström L, Brismar K, Scheynus A, Foshild Palmblad J: Enhanced generation of interleukins-1β and 6 may contribute to the cachexia of chronic disease. Am J Clin Nutr 1997;65:876–882.
25 Malavé I, Vethencourt MA, Pirela M, Cordero R: Serum levels of thyroxin-binding prealbumin, C-reactive protein and interleukin-6 in protein-energy undernourished children and normal controls without or with associated clinical infections. J Trop Pediatr, in press.
26 Doherty JF, Golden MHN, Remick DG, Griffin GE: Production of interleukin 6 and tumor necrosis factor-α in vitro is reduced in whole blood of severely malnourished children. Clin Sci 1994;86:347–351.
27 Mosmann TR, Sad S: The expanding universe of T-cell subsets: TH1, TH2 and more. Immunol Today 1996;17:138–146.
28 Romagnani S: Biology of human TH1 and TH2 cells. J Clin Immunol 1995;15:121–129.
29 McHugh S, Deighton J, Rifkin I, Ewan P: Kinetics and functional implications of TH1 and TH2 cytokine production following activation of peripheral blood mononuclear cells in primary culture. Eur J Immunol 1996;26:1260–1265.
30 Fitch FW, McKisic MD, Lancki DW, Gajewski TF: Differential regulation of murine T lymphocyte subsets. Annu Rev Immunol 1993; 11:29–48.
31 Ramsdell F, Seaman MS, Miller RE, Picha KS, Kennedy MK, Lynch DH: Differential ability of TH1 and TH2 cells to express Fas ligand and to undergo activation-induced cell death. Int Immunol 1994;6:1545–1553.
32 Musso T, Espinoza-Delgado I, Pulkki K, Gusella GL, Longo DL, Varesio L: IL-2 induces IL-6 production in human monocytes. J Immunol 1992;148:795–800.
33 Smeland EB, Blomhoff HK, Funderud S, Shalaby MR, Espevik T: Interleukin 4 induces selective production of interleukin 6 from normal human B lymphocytes. J Exp Med 1989;170:1463–1468.
34 Hagel I, Lynch NR, Di-Prisco MC, Sanchez J, Perez M: Nutritional status and the IgE response against *Ascaris lumbricoides* in children from a tropical slum. Trans R Soc Trop Med Hyg 1995;89:562–565.
35 Gracey MB, Suharjono CP, Sumoto, Stone DE: Microbial contamination of the gut: Another feature of malnutrition. Am J Clin Nutr 1973;26: 1170–1174.
36 Stanfield JP, Auff MSR, Tunnicliffe R: Intestinal biopsy in kwashiorkor. Lancet 1965;ii:519–523.
37 Schneider RE, Viteri FE: Morphological aspects of the duodenojejunal mucosa in protein-calorie malnourished children and during recovery. Am J Clin Nutr 1972;25: 1092–1102.
38 Deitch EA, Ma WJ, Ma L, Berg RD, Specian RD: Protein malnutrition predisposes to inflammatory induced gut-origin septic states. Ann Surg 1990;211:560–567.
39 Alvarado J, Luthringer DG: Serum immunoglobulins in edematous protein-calorie malnourished children. Clin Pediatr 1971;10:174–179.
40 Watson CE, Freesemann D: Immunoglobulins in protein-calorie malnutrition. Arch Dis Child 1970;45: 282–284.
41 Suskind R: Immune status of the malnourished host. J Trop Pediatr 1980;26:1–16.
42 Malavé I, Pocino M: Polyclonal B-cell response to stimulation with *Escherichia coli* lipopolysaccharide in dietary protein restriction. Infect Immun 1982;38:31–34.
43 Pocino M, Malavé I: Enhancement of the in vitro antibody response in dietary protein restriction. Failure in the regulation of antibody synthesis. Immunology 1981;43:235–240.
44 Durán-Chávez C, Sánchez-Herrera G, Cañedo-Solares I, Pérez-Ortíz B: C-Reactive protein in severely malnourished children. Nutr Res 1994; 14:967–975.
45 Strassman G, Fong M, Keney JS, Jacob CD: Evidence for the involvement of interleukin-6 in experimental cancer cachexia. J Clin Invest 1992;89:1681–1684.
46 Ohe Y, Podack ER, Olsen KJ, Miyahara Y, Miura K, Saito H, Koishihara Y, Ohsugi Y, Ohira T, Nishio K, et al: Interleukin 6 transfected Lewis lung carcinoma cells show unaltered net tumor growth rate but cause weight loss and shorten survival in syngeneic mice. Br J Cancer 1993; 67:937–944.
47 Agbedana E, Jhonson A, Oladumi-Taylor G: Selective deficiency of hepatic triglyceride lipase and hypertriglyceridemia in kwashiorkor. Br J Nutr 1979;42:351–356.
48 Carvajal I, Malavé I, Correa C, Castillo C, Pérez M, Hammer S, Camejo G: Alteraciones de las fracciones lipídicas en el suero de niños desnutridos con y sin infección clínica. Hipertrigliceridemia paradójica en desnutrición. Arch Latinoam Nutr 1992;42:250–258.