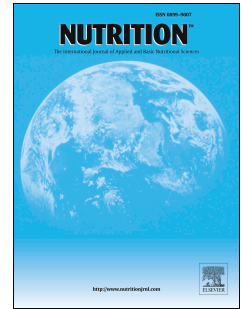


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Vitamin B12 Concentration and Association with Socio-demographic Factors in Colombian Children: Findings From the 2010 National Nutrition Survey (ENSIN)

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**Title:** Vitamin B12 Concentration and Association with Socio-demographic Factors in Colombian Children: Findings From the 2010 National Nutrition Survey (ENSIN)

**Title running:** Vitamin B12 and Socio-demographic Factors in Children

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**ABSTRACT****BACKGROUND**

Rapid changes in dietary patterns, economic development and urbanization in low-to-middle income countries are fueling complex malnutrition states that need better characterization using population-level data. The aim of this study is to describe the key findings related to Vitamin B12 status to identify the prevalence and associated socio-demographic factors in a representative sample of children in Colombia, based on the 2010 National Nutrition Survey.

**METHODS**

We analyzed cross-sectional data from 6,910 Colombian children between 5 and 12 years of age. Serum Vitamin B12 concentration were determined by chemiluminescence and socio-demographic data was assessed by computer-assisted personal interview technology.

**RESULTS**

A total of 2.8% of children had vitamin B12 deficiency, defined as levels lower than 200 pg/mL, and 18.1% had marginal vitamin B12 deficiency, defined as levels between 200 and 300 pg/mL. A multivariate logistic regression analysis revealed increased risks of Vitamin B12 deficiency among children 9 yrs-old and above and for those living in the eastern, western and southern regions of the country, but no significant associations were found for ethnic groups, socio-economic or urbanity levels. Being 11 years of age (OR 2.16; 95% CI 1.56 to 3.00;  $p=0.0001$ ), living in the west (pacific) region of the country (OR 3.92; 95% CI 3.14 to 4.90;  $p=0.0001$ )

and being male (OR 1.41; 95% CI 1.20 to 1.65;  $p=0.0001$ ) were the factors most strongly associated with an increased risk of vitamin B12 deficiency.

## **CONCLUSIONS**

Compared with data from other Latin-American countries Colombian children have a lower prevalence vitamin B12 deficiency but the prevalence of marginal deficiency is substantial. Continued surveillance and implementation of interventions to improve dietary patterns among high-risk groups identified should be considered.

**KEYWORDS:** Nutrition; Children; Vitamin B12; Deficiency; Prevalence

## **INTRODUCTION**

Under-nutrition has traditionally been the focus of nutrition agendas in low and middle income countries [1]. However, rapid economic development and urbanization have given rise to a nutrition transition, where energy-dense diets replace traditional diets and sedentary lifestyles prevail [2]. Nutrition related non-communicable diseases have their origin very early in life, and they develop during childhood and adolescence [3]. Given the risk of food malnutrition in developing countries, it is necessary to measure its prevalence in vulnerable populations, such as children, pregnant women and minorities, to prevent subsequent deterioration of their health [4]. To estimate the magnitude of this problem, the use of direct indicators to assess various nutrient levels, such as iron, vitamin A, zinc and vitamin B12, has been proposed [5]. These biochemical estimators reflects the

nutritional status which influences important biological processes, including cognitive development, physical growth and immune response [6].

Vitamin B12 deficiency, which is common in wealthier countries, principally among the elderly, is even more prevalent in poorer populations worldwide [7]. This deficiency is rare in children and is associated with anemia and strictly vegetarian diets. Vitamin B12 is not synthesized by humans, and the only sources of this nutrient are foods of animal origin, such as meat, fish, eggs and dairy [8]. Vitamin B12 deficiency affects the central nervous system and tissues with high mitotic activity, such as the epithelium of the digestive tract, and hematopoiesis [9]. Therefore, children who are breastfed by vegetarian mothers are at greater risk of serious complications related to vitamin B12 deficiency, which include hematologic [megaloblastic anemia], neurologic [hypotonia, ataxia, developmental delays] and gastrointestinal [inflammatory complications] symptoms [10]. The consequences of anemia are not limited to poor pregnancy outcomes, impaired physical and cognitive development, and increased risk of morbidity in children but also affect national productivity and economics [6].

Currently, there are few global reports on the prevalence of vitamin B12 deficiency in children, in particular for low-to-middle income countries experiencing rapid nutrition transitions such as Latin America. According to the Mexican National Nutrition Survey, the prevalence for vitamin B12 deficiency in school children between 7 and 10 years of age is 25% [10]. Similar data were reported by Casterline et al. [11] in school children in Guatemala. In Colombia, according to estimates from the National Survey of Nutritional Status [12,13], 1 in 5 children

under 5 years of age are at risk for vitamin B12 deficiency. Vitamin B12 deficiency is recognized as an important health problem and different studies involving different age groups and both genders have underscored the magnitude of vitamin B12 deficiency as a major public health challenge for improving the health status especially in refugee camps and among vulnerable at-risk groups. However, to the best of our knowledge, associations of Vitamin B12 deficiency with various socio-demographic factors that could help identify risk groups and offer information to better design interventions has not been investigated in a nationally representative sample in the Americas.

Therefore, the objective of this study is to identify factors associated with vitamin B12 deficiency in a representative sample of Colombian children aged 5 to 12 years.

## **METHODS**

### *Study population*

The Colombian National Nutrition Survey [ENSIN] was conducted in 2010 by the Colombian Institute of Family Welfare [14]. Details of the survey have been published elsewhere [14]. In brief, participants were selected to represent 99 % of the country's population using a multistage stratified sampling scheme. All municipalities from the thirty-two departments in the country were grouped into strata based on similar geographic and socio-demographic characteristics. One municipality was randomly chosen from each stratum, with probability proportional to the population size. Clusters of about ten households each were then randomly

chosen from within these strata and household members were invited to participate.

#### *Data sources*

The survey included 50,670 households, representing 4,987 clusters from 258 strata. Of the 7,266 children aged to 5-12 years, 6,910 [95.1%] were enrolled. This study was approved by the ethics committee of the National Survey of Nutritional Status. This study was reviewed and approved by the Institutional Review Board, INS [Instituto Nacional de Colombia]. Before starting the survey we have guaranteed participants the protection of confidential information obtained from them [Resolución 8430 de 1993; Ministerio de Salud de Colombia] [15]. The children and their parents signed a written informed consent and study procedures were conducted according to the principles expressed in the Declaration of Helsinki.

#### *Vitamin B12 concentrations*

In a random sub-sample of participants, blood was collected by venipuncture and serum was separated into aliquots. Serum vitamin B12 was quantified in these samples using direct chemiluminescence (ADVIA Centaur equipment, Siemens). This method offers high sensitivity and is less costly, easier to implement and safer than microbiological, chromatographic or spectrophotometric assays [14]. The main outcomes of interest were mean serum vitamin B12 concentrations (pg/mL) and the prevalence of vitamin B12 deficiency (serum concentration <200 pg/mL) [16] and marginal depletion (serum concentration 200 to 300 pg/mL) [16].

### *Sociodemographic characteristics*

The following socio-demographic variables were included as potentially associated factors with Vitamin B12 deficiency: age (5 to 5.9, 6 to 6.9, 7 to 7.9, 8 to 8.9, 9 to 9.9, 10 to 10.9, 11 to 11.9 and 12 to 12.9); sex (male and female); urbanicity [grouped as urban and rural]; ethnicity grouped as: a) indigenous, b) black or afro-colombian and c) others (mestizo); geographic region: a) atlantic (north), b) eastern, c) central, d) pacific (west), e] Bogota and f) national territories (south); and social or socio-economic status determined by the System of Identifying Potential Beneficiaries of Social Programs (SISBEN for its Spanish initials) (1 to 3, and 4 or more). The SISBEN is a system designed by the Colombian National Government to identify families who could benefit from social programs. It takes into account sociodemographic characteristics (family composition, employment status, family income, and educational level), living conditions (construction type and materials), and access to public utilities (sewer, electricity, potable water, and garbage collection). Households are classified into 6 levels with 1 being the poorest and 6 being the wealthiest. For this study we classified SISBEN scores into 4 categories (1, 2, 3 and 4 or more).

### *Statistical Analyses*

First, we conducted an exploratory analysis of the frequency distribution [measures of central tendency and dispersion for quantitative variables] and relative frequencies [for qualitative variables] using the *Pearson*  $\chi^2$  test with and without the *Yates* correction. To estimate the relationship between vitamin B12 deficiency and

sociodemographic variables in children (age, sex, urbanicity, geographic region, ethnicity and socioeconomic level-SISBEN), binary logistic regression models were used. The first adjusted model was for age and sex, the second model was based on ethnic group, geographic area, socioeconomic levels and urbanicity; the third model was adjusted by age, sex, ethnic group, geographic area, socioeconomic levels and urbanicity. Odds ratios were considered a confounder if they shifted the model in a constant direction with a proportional increase in the exposure level of at least 10%. All analyses were conducted with the use of the complex survey design routines of the SPSS Statistical software package version 20.

## RESULTS

The study cohort consisted of 6,910 children between 5 and 12 years of age (mean age 8.7 years). The range of vitamin B12 was 45 to 1000 pg/mL (mean 434.9 pg/mL, 95% CI 429.5 to 440.4 pg/mL). A total of 2.8% of children had vitamin B12 levels below 200 pg/mL and 18.1% had levels between 200 and 300 pg/mL. Children that live in the Pacific area, were 11 years old and belonged to SISBEN level II had the highest percentage of vitamin B12 values less than 300 pg/mL (36.2%, 27.0% and 23.0%, respectively). The distribution of the vitamin B12 deficiency and depletion in children aged 5 to 12 years is shown in Table 1.

**\*\* Insert Table 1 \*\***

Table 2 shows the results for a logistic regression analysis. Being 11 years of age OR 2.16 (95% CI 1.56 to 3.00), living in the Pacific (west) OR 3.92 (95% CI 3.14 to 4.90) or national territories (south) OR 1.69 (95% CI 1.32 to 4.90) and being male

OR 1.41 (95% CI 1.20 to 1.65) were predisposing factors for vitamin B12 deficiency.

**\*\* Insert Table 2 \*\***

## **DISCUSSION**

Vitamin B12 deficiency has serious consequences on neurological and functional development in children. In Colombia, vitamin B12 deficiency in children aged 5 to 12 years is relatively low (2.8%). However, a total of 18.1% of children had low values, a factor that should be considered by policy makers in public health. Thus, the results of our study support the conclusion that the prevalence of vitamin B12 deficiency in Colombian children aged 5 to 12 years is low when compared with rates in Latin American countries, including Mexico [10] and Guatemala [11] which on average exceed 20%.

Similarly, it was observed that the risk of vitamin B12 deficiency in Colombian children increases with age, with prevalence ranging from 13.6% at 5 years of age to 22.0% for 12 year-olds. Only one previous study had examined vitamin B12 concentrations in Colombia. Among low-and middle-income children from Bogotá, the combined prevalence of deficiency and marginal deficiency was 17% in 2006 [17], similar to the result found for this age group and city in the present survey.

A logistic regression analysis showed an association with being 11 years old and the risk of vitamin B12 deficiency. No variations were observed for ethnic groups, socioeconomic status or urban and rural area. However, the prevalence was higher in males (20.7%) when compared with females (15.2%). The differences observed

in our study are similar to previously reports by Allen [16] which analyzed vitamin B12 deficiency in the Americas. Colombian children living in the Pacific region showed a higher prevalence for vitamin B12 deficiency (30.4%) while those living in the Central and Caribbean regions showed a lower prevalence (<15%). Living in the Pacific region was considered a factor associated with vitamin B12 deficiency in Colombian children, similar results published by investigators from the nutrition department of the WHO [18]. Although not measured in this study, in Pacific region the school meals are likely to have low phytate levels, because they consist mainly of fast food, eggs and milk [15]. In addition, in Colombia, the Pacific region environments have been associated with aridity, high food prices, limited natural resources, and poor infrastructure development, which often affect the availability of and access to food and health services. Such factors can lead to increased malnutrition among populations residing at this region. Similar challenges can also be found in the Eastern and South (National territories) areas of the country and may explain the higher prevalence of B12 deficiently found in those regions as compared to more developed areas of the country (Bogota, Central and Atlantic regions) that include large urban centers. Previous studies have also found that children living in urban areas and in geographic regions with greater economic and structural development in general had higher serum concentrations than those from rural and poorer areas [19-23].

In another study in Guatemala City, 24% of 127 infants aged 7 to 12 months had deficient (< 200 pg/mL), and 37% had marginally deficient (200 to 300 pg/mL), plasma vitamin B12 concentrations [19]. Plasma vitamin B12 was lower in those

infants who consumed more breastmilk ( $r = -0.33$ ,  $p < 0.0001$  with measured breastmilk intake, kcal/day) but positively associated with B-12 intake from complementary foods. In a different group of infants ( $n = 304$ ) aged 12 months in Guatemala City, deficient and marginal plasma vitamin B12 values were found in 30% and 20% of infants, and 36% and 32% of their mothers, respectively [20].

Vitamin B12 is a cofactor of methionine synthase in the synthesis of methionine, the precursor of the universal methyl donor S-Adenosylmethionine (SAME), involved in different epigenomic regulatory mechanisms and especially in fetal growth [21]. Measurement of the total vitamin B12 concentration in plasma is the usual method for assessing vitamin B12 status. However, neurological and hematological symptoms of depletion can occur in individuals with plasma vitamin B12 concentrations in the low-normal range [22]. Nevertheless, associations between vitamin B12 marginal depletion and adverse health outcomes in observational studies are of concern and urgently need further evaluation in clinical trials [23].

Several factors can affect vitamin B12 concentrations, such as individual genetic variation, disease conditions and prescriptions. Approximately 20% of Latin-American children with low serum vitamin B12 concentrations have neither clinical nor metabolic signs of vitamin B12 depletion [21,24]. One of the important causes for lower status of vitamin B12 could be poor dietary patterns [21]. Based on our dietary survey among indigenous populations in Colombia, the intake of animal protein was very low among children [19]. So enriching the diet and increasing

intake of animal-derived foods might be an important way to improve the status of vitamin B12 of Colombian children.

Measurement procedure errors and other technological problems produce slightly high or low biomarker concentrations. These factors adversely affect the biomarkers' sensitivity and specificity and result in false-positive or false-negative classifications of vitamin B12 depletion. Systematic bias and spurious results affected some measurement procedures used in the past to derive reference ranges [25-27]. The cut-off used in this study affects ENSIN-2010 prevalence estimates for Vitamin B12 deficiency [25].

In this study, one limitation had to be addressed. Our survey was about the health status of Colombian children and only vitamin B12 was measured. Unfortunately, the metabolic precursors of vitamin B12, homocysteine (tHcy), methylmalonic acid (MMA) and holotranscobalamin (holoTC), were not been measured. Studies indicated that these markers increased or decreased when the status of vitamin B12 were low [28]. However, tHcy and MMA started to increase at vitamin B12 levels above the typical cut-off value ( $< 200$  pg/mL), and they increased quickly when vitamin B12 decreased from 400 to 200 pg/mL, which suggests that changes on markers related to nervous system impairment had already begun before the deficiency in vitamin B12 can be determined [29]. The use of serum vitamin B12 as a proxy of vitamin B12 status can benefit from further assessment of its metabolic precursors. A second limitation of the present study was that dietary intake of vitamin B12 was not assessed. Herran et al. [26] determined that the bioavailability of vitamin B12 was greater from dairy products and fish than

from meat. In the dietary study we conducted in Villamor [17], only 13.4% of the protein in the diets of children was derived from eggs and milk, with 40% of protein coming from meat. Colombia is a country that is geographically, climatically and ethnically diverse. Clearly these differences could affect food supply, dietary practices, and consequently vitamin B12 intakes. The pacific, eastern and central parts of Colombia are very diverse, which lead to different dietary patterns and climate. Third, the use of specific cut-off points of serum vitamin B12 to define deficiency and marginal deficiency also deserves comment. We used the conventionally accepted cut-off points of < 200 pg/mL (deficiency) and 200 to 300 pg/mL (marginal deficiency) [21,25]. While these cut-off points may be considered somewhat arbitrary, there is a pathophysiological rationale to their use. MMA and tHcy concentrations associated with anaemia, megaloblastosis and neuropathy decrease substantially when serum vitamin B12 concentrations are greater than < 200 pg/mL, even in the absence of clinical manifestations [26].

In conclusion, Vitamin B12 deficiency remains an area that merits strict monitoring among Colombian children between 5 and 12 years of age. Although its current prevalence is slightly lower than reports from other Latin American countries [10,11,13,17,26,30], the somewhat high prevalence of marginal Vitamin B12 deficiency in this study provides reason to suggest that children could benefit from strategies to supplement vitamin B12, in particular in regions of the country where deficiency is more prevalent [31,32]. This can be done via tailored supplementation in clinical settings or via broader public health supplementation strategies as done in other countries such as Guatemala, Puerto Rico and México [10,20,30]. For

example a sensible strategy may be to include vitamin B12 along with folic acid in flour fortification programs, where a need for additional vitamin B12 has been established. This could have multiple benefits, including further reduction in neural tube defects, improved vitamin B12 levels in breast milk and among mothers and young children, and prevention of cognitive decline in the elderly [30]. In addition, coordination with other dietary interventions and educational strategies can produce viable and sustainable interventions in particular among the high-risk groups identified in this study.

#### **COMPETING INTERESTS**

The authors declare that they have no competing interests.

#### **AUTHORS' CONTRIBUTIONS**

RR-V formulated the research question, carried out the analyses and drafted the initial manuscript. JM-T, JFM-E, JEC-B and FL were involved with the conceptualization and design of the study and analyses of data. All authors critically reviewed the manuscript, and approved the final manuscript as submitted.

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## REFERENCES

1. Popkin BM. The nutrition transition and its health implications in lower-income countries. *Public Health Nutr.* 1998;1:5-21.
2. Moreno LA, Gottrand F, Huybrechts I, Ruiz JR, González-Gross M, DeHenauw S; HELENA Study Group. Nutrition and lifestyle in european adolescents: the HELENA [Healthy Lifestyle in Europe by Nutrition in Adolescence] study. *Adv Nutr.* 2014;5:615S-623S.
3. Fernandez-Twinn DS, Ozanne SE. Early life nutrition and metabolic programming. *Ann N Y Acad Sci* 2010;1212:78–96.
4. López-Sáleme R, Díaz-Montes C, Bravo-Aljuriz L, Londoño-Hio N, Salgado-Pájaro M, et al. Seguridad alimentaria y estado nutricional de las mujeres embarazadas en Cartagena, Colombia, 2011. *Rev. Salud Pública* 2012;14: 200-212.
5. Zemel BS, Riley EM, Stallings VA. Evaluation of methodology for nutritional assessment in children: anthropometry, body composition, and energy expenditure. *Annu Rev Nutr* 1997;17:211–35.
6. Tzioumis E, Adair LS. Childhood dual burden of under- and overnutrition in low-and middle-income countries: a critical review. *Food Nutr Bull.* 2014; 35:230-43.
7. Jadhav M, Webb J, Vaishnava S, Baker S. Vitamin B12 deficiency in Indian infants. *Lancet* 1962;2:903-907.
8. Graham SM, Arvela OM, Wise GA. Long-term neurologic consequences of nutritional vitamin B 12 deficiency in infants. *J Pediatr* 1992;121:710-704.
9. Rasmussen AS, Fernhoff PM, Scanlon KS. Vitamin B12 deficiency in children and adolescents. *J Pediatr* 2001;138:10-17.
10. Black AK, Allen LH, Peltó GH, de Mata MP, Chávez A. Iron, vitamin B-12 and folate status in Mexico: associated factors in men and women and during pregnancy and lactation. *J Nutr* 1994;124:1179-1188.

11. Casterline JE, Allen LH, Ruel MT. Vitamin B12 deficiency is very prevalent in lactating Guatemalan women and their infants at three months postpartum. *J Nutr* 1997;127:1966-1972.
12. Instituto Colombiano de Bienestar Familiar. Encuesta Nacional de la Situación Nutricional en Colombia, 2005; Bogotá: Profamilia.
13. Sarmiento OL, Parra DC, González SA, González-Casanova I, Forero AY, Garcia J. The dual burden of malnutrition in Colombia. *Am J Clin Nutr*. 2014;100:1628S-35S.
14. Instituto Colombiano de Bienestar Familiar. Encuesta Nacional de la Situación Nutricional en Colombia, 2010; Bogotá: Profamilia.
15. República de Colombia. Ministerio de Salud. Resolución N° 008430 DE 1993 [4 de Octubre de 1993].
16. Allen LH. Folate and vitamin B12 status in the Americas. *Nutr Rev* 2004;62:S29-33; discussion S34.
17. Villamor E, Mora-Plazas M, Forero Y, Lopez-Arana S, Baylin A. Vitamin B-12 status is associated with socioeconomic level and adherence, to an animal food dietary pattern in Colombian school children. *J Nutr* 2008;138;1391-1398.
18. McLean E, de Benoist B, Allen LH. Review of the magnitude of folate and vitamin B12 deficiencies worldwide. *Food Nutr Bull* 2008;29:S38-51.
19. Anaya M, Begin F, Brown KH, Peerson JM, Torun B, Allen LH. The high prevalence of vitamin B12 deficiency in Guatemalan infants is associated with a higher intake of breast milk, and with poor quality complementary foods. *FASEB J* 2004;18:A844 [abstract].
20. Jones KM, Ramirez-Zea M, Zuleta C, Allen LH. Prevalent vitamin B12 deficiency in twelve-month-old Guatemalan infants is predicted by maternal B12 deficiency and infant diet. *J Nutr* 2007;137:1307-13.
21. Uwe Gröber, Klaus Kisters, Joachim Schmidt. Neuroenhancement with Vitamin B12—Underestimated Neurological Significance. *Nutrients* 2013;5:5031-5045.

22. Vanderjagt DJ, Ujah IA, Ikeh EI, Bryant J, Pam V, Hilgart A, et al. Assessment of the vitamin B12 status of pregnant women in Nigeria using plasma holotranscobalamin. *ISRN Obstet Gynecol.* 2011;2011:365894.
23. Yetley EA, Pfeiffer CM, Phinney KW, Bailey RL, Blackmore S, Bock JL, et al. Biomarkers of vitamin B-12 status in NHANES: a roundtable summary. *Am J Clin Nutr* 2011;94:313S-321S.
24. Allen LH. Causes of vitamin B12 and folate deficiency. *Food Nutr Bull.* 2008;29:S20-34; discussion S35-7.
25. Carmel R. Biomarkers of cobalamin [vitamin B-12] status in the epidemiologic setting: a critical overview of context, applications, and performance characteristics of cobalamin, methylmalonic acid, and holotranscobalamin II. *Am J Clin Nutr* 2011;94:348S-58S
26. Herrán OF, Ward JB, Villamor E. Vitamin B 12 serostatus in Colombian children and adult women: results from a nationally representative survey. *Public Health Nutr* 2014;27:1-8
27. Mahmoud MS, Yassin MM, Shekhi SE, Lubbad AM. Homocysteine and vitamin B12 status and iron deficiency anemia in female university students from Gaza Strip, Palestine *Rev Bras Hematol Hemoter.* 2014;36:208-2.
28. Herrmann W, Obeid R, Schorr H, Geisel J. Functional vitamin B12 deficiency and determination of holotranscobalamin in populations at risk. *Clin Chem Lab Med.* 2003; 41:1478-88.
29. Allen LH. Vitamin B12 metabolism and status during pregnancy, lactation and infancy. *Adv Exp Med Biol.* 1994;352:173-86.
30. Brito A, Mujica-Coopman MF, Olivares M, López de Romaña D, Cori H, Allen LH. Folate and Vitamin B12 Status in Latin America and the Caribbean: An Update. *Food Nutr Bull.* 2015;36(2 suppl):S109-S118.
31. Ramírez-Vélez R, Martínez-Torres F. Factores asociados a la deficiencia de zinc en niños colombianos; resultados de la Encuesta Nacional de Salud 2010; estudio transversal. *Nutr Hop.* 2014;29(4):832-837.

32. Ramírez-Vélez R, Martínez-Torres F, Meneses-Echavez JF. Prevalence and Demographic Factors Associated with Vitamin A deficiency in children aged 1 to 4 years from Colombia. *Endocrinol Nutr.* 2014;61(9):460-466.

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**Table 1.** Characterization of Vitamin B-12 levels among Colombian children. National Nutrition Survey, 2010

Characteristics	Adequate B12 concentration		Marginal vitamin B12 deficiency		Vitamin B12 deficiency	
	n	%** (95% CI)	n	%** (95% CI)	n	%** (95% CI)
<b>Total</b>	5,371	79.2 (78.7-79.6)	1,304	18.1 (17.2-18.9)	235	2.8 (2.3-3.2)
<b>Age (years) <sup>a</sup></b>						
5 to 5.9	620	85.0 (83.9-85.8)	125	13.6 (11.4-15.3)	21	1.5 (0.6-2.2)
6 to 6.9	623	81.1 (80.0-81.9)	131	16.8 (14.4-18.7)	21	2.2 (0.8-3.3)
7 to 7.9	685	84.4 (83.5-85.1)	125	13.8 (11.4-15.8)	24	1.8 (1.0-2.5)
8 to 8.9	703	81.2 (80.3-81.9)	153	16.1 (13.7-18.1)	30	2.7 (1.5-3.8)
9 to 9.9	679	79.9 (78.7-80.9)	188	19.0 (16.8-20.9)	20	1.1 (0.4-1.7)
10 to 10.9	734	77.2 (76.1-78.2)	188	18.0 (15.9-19.9)	43	4.7 (3.2-6.1)
11 to 11.9	694	73.1 (71.8-74.1)	202	23.5 (21.0-25.6)	39	3.5 (2.1-4.6)
12 to 12.9	633	74.0 (72.8-75.1)	192	22.0 (19.5-24.1)	37	4.0 (2.4-5.2)
<b>Sex</b>						
Male	2,658	76.4 (75.8-77.0)	749	20.7 (19.5-21.8)	126	2.9 (2.2-3.5)
Female	2,713	82.1 (81.6-82.6)	555	15.2 (14.1-16.3)	109	2.7 (2.1-3.2)
<b>Socioeconomic levels by SISBEN</b>						
Level I	3,204	79.5 (78.9-80.0)	787	17.5 (16.3-18.5)	159	3.1 (2.4-3.6)
Level II	664	77.0 (76.0-77.8)	166	20.8 (18.3-22.9)	20	2.2 (1.0-3.3)
Level III	475	79.3 (78.1-80.4)	112	18.4 (15.7-20.5)	13	2.3 (0.7-3.5)
Level IV or more	1,028	79.6 (78.7-80.3)	239	17.8 (16.0-19.3)	43	2.7 (1.8-3.4)
<b>Geographic area</b>						
Atlantic (North) <sup>a</sup>	1,301	86.5 (86.0-86.9)	240	12.5 (11.2-13.7)	25	1.0 (0.6-1.4)
Eastern <sup>a</sup>	732	75.9 (74.9-76.8)	194	20.0 (18.1-21.7)	41	4.1 (2.9-5.1)
Central <sup>a</sup>	1,254	83.9 (83.2-84.5)	230	14.5 (12.9-16.0)	32	1.6 (0.9-2.1)
Pacific (West)	640	63.8 (62.6-64.8)	285	30.4 (28.4-32.2)	59	5.8 (4.4-7.0)
Bogotá <sup>a</sup>	252	82.6 (81.6-83.5)	47	15.4 (12.4-17.8)	6	2.0 (0.4-3.2)
National territories (South) <sup>a</sup>	1,192	79.3 (78.6-79.9)	308	17.4 (15.4-19.2)	72	3.3 (2.1-4.4)
<b>Urbanicity</b>						
Urban	3,296	79.8 (79.3-80.3)	773	17.5 (16.4-18.4)	134	2.7 (2.2-3.2)
Rural	2,075	77.6 (76.9-78.3)	531	19.5 (18.0-20.8)	101	2.9 (2.2-3.5)
<b>Ethnic group*</b>						
Indigenous <sup>a</sup>	640	78.6 (77.0-79.8)	204	19.5 (16.5-21.8)	45	1.9 (0.4-3.1)
Black or Afro-Colombian <sup>a</sup>	558	77.4 (76.3-78.3)	155	18.3 (16.1-20.2)	38	4.3 (2.7-5.6)
Others	4,145	79.4 (78.9-79.9)	927	18.0 (17.0-18.9)	149	2.6 (2.1-3.1)

\* All children analysed by ethnic group were n= 6,861, another 49 appertained to "Raizal del archipiélago" and "Palenquero de San Basilio", who were not analysed because this group did not have a representative sample

\*\* It is not correct to calculate the percentages from the "n" presented in this table; these calculations were taken from weighted values given to each subject

<sup>a</sup> Coefficient of variation is more than 20% deficiency prevalence therefore variation should be used with caution

**Table 2.** Socio-demographic factors associated with Vitamin B-12 deficiency/marginal in Colombian Children. National Nutrition Survey 2010

Characteristics	Bivariate	P value	Adjusted model <sup>a</sup>	P value	Adjusted model <sup>b</sup>	P value	Adjusted model <sup>c</sup>	P value
<b>Age (years)<sup>d</sup></b>								
6 to 6.9	1.32 (0.93-1.86)	0.7945	1.31 (0.93-1.85)	0.7569	1.35 (0.95-1.91)	0.6745	1.34 (0.94-1.90)	0.6909
7 to 7.9	1.05 (0.74-1.49)	0.5558	1.04 (0.73-1.48)	0.5389	1.08 (0.75-1.55)	0.4669	1.07 (0.74-1.54)	0.4648
8 to 8.9	1.31 (0.93-1.83)	0.4378	1.30 (0.93-1.83)	0.4067	1.36 (0.96-1.91)	0.1389	1.35 (0.96-1.91)	0.1230
9 to 9.9	<b>1.42 (1.02-1.98)</b>	<b>0.0410</b>	<b>1.40 (1.01-1.95)</b>	<b>0.0400</b>	<b>1.50 (1.07-2.09)</b>	<b>0.0345</b>	<b>1.48 (1.05-2.07)</b>	<b>0.0378</b>
10 to 10.9	<b>1.67 (1.21-2.30)</b>	<b>0.0201</b>	<b>1.66 (1.21-2.30)</b>	<b>0.0291</b>	<b>1.73 (1.24-2.40)</b>	<b>0.0280</b>	<b>1.73 (1.24-2.41)</b>	<b>0.0278</b>
11 to 11.9	<b>2.08 (1.51-2.87)</b>	<b>0.0001</b>	<b>2.06 (1.49-2.83)</b>	<b>0.0001</b>	<b>2.18 (1.57-3.03)</b>	<b>0.0001</b>	<b>2.16 (1.56-3.00)</b>	<b>0.0001</b>
12 to 12.9	<b>1.98 (1.43-2.74)</b>	<b>0.0001</b>	<b>1.94 (1.40-2.69)</b>	<b>0.0001</b>	<b>2.10 (1.51-2.92)</b>	<b>0.0001</b>	<b>2.06 (1.48-2.87)</b>	<b>0.0001</b>
<b>Sex<sup>e</sup></b>								
Male	<b>1.42 (1.21-1.66)</b>	<b>0.0001</b>	<b>1.40 (1.20-1.64)</b>	<b>0.0001</b>	<b>1.42 (1.21-1.66)</b>	<b>0.0001</b>	<b>1.41 (1.20-1.65)</b>	<b>0.0001</b>
<b>Socioeconomic levels by SISBEN<sup>f</sup></b>								
Level I	1.00 (0.83-1.22)	0.9756	1.04 (0.85-1.27)	0.7156	1.06 (0.87-1.30)	0.5551	1.09 (0.89-1.34)	0.3981
Level II	1.17 (0.89-1.52)	0.2734	1.19 (0.91-1.56)	0.2109	1.21 (0.92-1.60)	0.1890	1.23 (0.93-1.63)	0.1426
Level III	1.01 (0.75-1.37)	0.9321	1.04 (0.77-1.41)	0.8015	1.08 (0.79-1.46)	0.6544	1.10 (0.81-1.50)	0.5350
<b>Geographic area<sup>g</sup></b>								
Eastern	<b>2.03 (1.61-2.56)</b>	<b>0.0001</b>	<b>1.98 (1.56-2.50)</b>	<b>0.0001</b>	<b>1.96 (1.54-2.49)</b>	<b>0.0001</b>	<b>1.93 (1.51-2.45)</b>	<b>0.0001</b>
Central	1.23 (0.97-1.56)	0.1348	1.21 (0.95-1.54)	0.2380	1.20 (0.94-1.53)	0.2250	1.19 (0.93-1.51)	0.3230
Pacific (West)	<b>3.64 (2.93-4.51)</b>	<b>0.0001</b>	<b>3.69 (2.97-4.57)</b>	<b>0.0001</b>	<b>3.86 (3.10-4.81)</b>	<b>0.0001</b>	<b>3.92 (3.14-4.90)</b>	<b>0.0001</b>
Bogotá	1.35 (0.96-1.89)	0.2245	1.31 (0.93-1.83)	0.2400	1.36 (0.97-1.91)	0.1823	1.33 (0.95-1.88)	0.1978
National territories (South)	<b>1.68 (1.32-2.13)</b>	<b>0.0001</b>	<b>1.69 (1.33-2.16)</b>	<b>0.0001</b>	<b>1.67 (1.31-2.14)</b>	<b>0.0001</b>	<b>1.69 (1.32-2.16)</b>	<b>0.0001</b>
<b>Urbanicity<sup>h</sup></b>								
Rural	1.14 (0.97-1.34)	0.1269	1.15 (0.97-1.35)	0.1170	1.11 (0.93-1.32)	0.2581	1.11 (0.93-1.32)	0.2609
<b>Ethnic group<sup>i</sup></b>								
Indigenous	1.05 (0.79-1.40)	0.7450	1.10 (0.82-1.48)	0.5178	0.76 (0.56-1.03)	0.0890	0.79 (0.58-1.07)	0.1311
Black or Afro-colombian	1.13 (0.90-1.40)	0.3078	1.15 (0.92-1.44)	0.2124	0.79 (0.62-1.01)	0.0689	0.79 (0.62-1.01)	0.0647

Odds ratios (95% confidence interval)

<sup>a</sup> adjusted by age and sex<sup>b</sup> adjusted by ethnic group, geographic area, socioeconomic levels and urbanicity<sup>c</sup> adjusted by age, sex, ethnic group, geographic area, socioeconomic levels and urbanicity<sup>d</sup> reference group: 5 to 5.9<sup>e</sup> reference group: Female

<sup>f</sup> reference group: Level 4 or more

<sup>g</sup> reference group: Atlantic (North)

<sup>h</sup> reference group: Urban

<sup>i</sup> reference group: Others

Significant odds ratios are shown in bold

ACCEPTED MANUSCRIPT

## Highlights

Vitamin B12 is not synthesized by humans, and the only sources of this nutrient are foods of animal origin.

Vitamin B12 deficiency is recognized as an important health problem.

We analyzed cross-sectional data from 6,910 Colombian children between 5 and 12 years of age.

Compared with data from other Latin-American countries Colombian children have a lower prevalence vitamin B12 deficiency