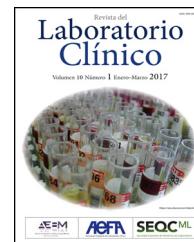


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ORIGINAL ARTICLE

Reference values for serum folate and vitamin B₁₂ in a Spanish population using an electrochemiluminiscent method

José Luis Martín Calderón^{a,*}, Luis Caballero^a, Fernando Solano^b, Fernando Bustos^a

^a Department of Clinical Chemistry, Hospital Nuestra Señora del Prado, Talavera de la Reina, Spain

^b Department of Haematology, Hospital Nuestra Señora del Prado, Talavera de la Reina, Spain

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KEYWORDS

Folate;
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Reference values;
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Abstract

Background: Despite being the most widely used medical decision-making tool, reference intervals are not usually determined by clinical laboratories, due to the highly demanding activities and costly process it involves. However, scientific societies encourage individual clinical laboratories to establish their own reference values. This is especially important in the cases of folate and vitamin B₁₂, due to strong differences in vitamin status among different populations.

Objective: Our aim is to establish reference intervals for folate and vitamin B₁₂ levels in a healthy blood donor population using an electrochemiluminiscent method (ROCHE DIAGNOSTICS).

Method: Folate and vitamin B₁₂ levels were measured in 141 healthy blood donors aged between 18 and 65 years. Biochemical analyses were performed using a Modular E170 analyzer (ROCHE DIAGNOSTICS) and an electrochemiluminiscent method. Reference intervals were calculated with a non-parametric percentile method following the CLSI guidelines.

Results: There were not significant differences in folate or cobalamin levels between age or sex subgroups. The limits of the reference interval for folate were 2.2 and 18 ng/mL (5–40.7 nmol/L), and 213.8 and 763.3 pg/mL (158.2–564.8 pmol/L) for vitamin B₁₂. These intervals differed from those claimed by the manufacturer.

Conclusions: Our results emphasize the convenience of building reference values based on the population served by the laboratory, in order to unequivocally rule out deficiencies of folate or vitamin B₁₂.

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* Corresponding author.

E-mail address: jlmartinc@sescam.jccm.es (J.L. Martín Calderón).

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PALABRAS CLAVE

Folato;
Vitamina B12;
Cobalamina;
Valores de referencia;
Electroquimioluminiscencia

Valores de referencia de ácido fólico y vitamina B12 séricos en una población española, utilizando un método de electroquimioluminiscencia

Resumen

Introducción: A pesar de ser una de las herramientas más usadas en la toma de decisiones médicas, los intervalos de referencia son raramente determinados en los laboratorios, debido a que es una labor compleja en términos de tiempo, esfuerzo y coste. Sin embargo las sociedades científicas recomiendan que los laboratorios establezcan sus propios valores de referencia. Esto es especialmente importante para el folato y la vitamina B12, debido a las grandes diferencias entre poblaciones.

Objetivo: Nuestro propósito es establecer los valores de referencia en una población de donantes sanos, mediante un método electroquimioluminiscente (Roche Diagnostics).

Método: Los niveles de folato y vitamina B12 se midieron en una muestra integrada por 141 donantes sanos de una edad comprendida entre 18 y 65 años. Los análisis bioquímicos se realizaron en un analizador Modular E170 (Roche Diagnostics). Los intervalos de referencia se calcularon siguiendo el método no paramétrico propuesto por las guías del CLSI.

Resultados: No hubo diferencias en los niveles de folato ni vitamina B12 entre sexos ni grupos de edad. Los límites de referencia para el folato fueron 2,2 y 18 ng/ml (5-40,7 nmol/l), y 213,8 y 763,3 pg/ml (158,2-564,8 pmol/l) para la vitamina B₁₂. Estos intervalos difirieron de los propuestos por el fabricante.

Conclusiones: Nuestros resultados subrayan la importancia de obtener valores de referencia en la población a la que da servicio el laboratorio para excluir inequívocamente las deficiencias de folato y vitamina B12.

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Introduction

Folate and vitamin B₁₂ are water soluble vitamins involved in one carbon transfer (methylation) reactions necessary for the production of monoamine neurotransmitters, phospholipids and the nucleotides used in DNA synthesis.¹⁻⁵ Vitamin B₁₂ or folate depletion both increase homocysteine levels, being high circulating levels of homocysteine often consequence of an inadequate status of these vitamins.⁶ Hyperhomocysteinaemia has been associated with cardiovascular, haematological and neurological diseases.^{6,7}

Deficiency of folate or of cobalamin may cause different types of disorders, mainly haematological disorders⁷ such as anaemia and macrocytosis, but also others as memory disturbances, delirium, mood disorders and psychoses.^{8,9} Folate and vitamin B₁₂ deficiencies are still important problems worldwide. The difficulty to establish consensus levels is due to the fact that serum concentrations of these vitamins differ according to age, gender and other variables.⁶ Vitamin B₁₂ levels decrease with age, being clinical and subclinical deficiency more prevalent in the elderly.¹⁰ Epidemiological data suggest an estimated prevalence of at least 10% of individuals over 60 years.^{5,11} Age influence is not so clear-cut in the case of folate levels.¹² In addition diet, habits, and physiological and pathological factors influence cobalamin and folate status. For instance, low vitamin B₁₂ levels are more frequent in vegetarians,^{10,13} in patients with gastrointestinal diseases,¹⁴ and in individuals with high alcohol consumption or with renal failure.^{10,14} Folate status is also negatively influenced by dietary, pathological (alcoholism, depression) and pharmacological (anticonvulsants)

factors.¹⁵ Variability is further enhanced by the compulsory fortification of flour with folate and vitamin B₁₂ dictated by some government agencies.^{5,15} As a consequence, prevalence rates for folate and vitamin B₁₂ deficiencies vary considerably across different studies.¹² Besides the aforementioned factors that promote variability, it is important to take also into account methodological differences, especially when immunoassays are used.

The aim of this work is to define the reference values for serum folate and vitamin B₁₂ in our population. According to Horn and Pesce,¹⁶ reference intervals are the most widely used medical decision-making tools. Ideally, all clinical laboratories should determine their own reference values. However, most of them do not, due to the fact that the establishment of reliable reference intervals is a highly demanding activity,¹⁷ and therefore the usual practice is to adopt the intervals provided in the manufacturer's inserts. We have calculated reference intervals for a population from Central Spain using an electrochemiluminiscent method (Roche Diagnostics) in a Modular E-170 analyzer.

Materials and methods

Patients

One hundred and forty one healthy volunteers aged 18–65 years were selected among blood donors of our hospital. Written informed consent was obtained from all of them. Abuse drug consumers, smokers and alcohol drinkers were excluded from the study. The individuals did not have notice-

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