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Analytical Methods

Voluntary fortification with folic acid in Spain: An updated food composition database

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ABSTRACT

Folic acid (FA) is a key vitamin in the prevention of many diseases including neural tube defects. In Spain, only voluntary FA food fortification is allowed and there is a lack of compositional data to assess the contribution of these products to population's dietary folate intakes. Since 2007, our group has been compiling and updating a FA fortified food composition database. FA levels were obtained from retailers in Madrid and information provided by manufacturers. FA was also quantified by an affinity chromatography-HPLC method. In the present study we recorded 375 products. Our results show a high variability in the declared FA levels amongst different products, and food groups, which is also dependant on the commercial brand. FA overages are commonly added by manufacturers to some fortified products. FA content label claims are missing in 64% of products. This database is a useful tool to manage FA fortified foods data but it is necessary to continuously update it for the sound evaluation and monitoring of population's FA dietary intakes.

1. Introduction

Folic acid (FA) is considered to be a key vitamin in the prevention of many diseases including neural tube defects (NTDs) (Czeizel & Dudas, 1992; MRC, 1991). Women who might become pregnant are recommended to supplement their diet with 400 μg/day of FA (EFSA, 2009). Mandatory fortification of the food supply with FA-mainly wheat four- has been taking place for more than 15 years and, at present, about 77 countries worldwide have implemented this nutritional policy (FFI, 2013). The main objective of this approach is to improve folate intakes of women at a reproductive age. In Europe, however, safety concerns of a population based exposure to high FA intakes have been the main reason to deny or delay mandatory fortification, and for most countries FA fortification is only permitted on a voluntary basis (EFSA, 2009). Hence in Spain, voluntary food fortification with FA is endorsed in accordance with Regulation (EU) No. 1169/2011 (EU, 2011) in which for labelling purposes, FA Daily Reference Intakes (DRI) nutrient reference values is set at 200 μg. The fortified and functional product market in Spain has slowed down the continuous growth that was experiencing since 2006, but it remains a relevant sector in the food industry (Nohr et al., 2014).

Folate intakes and nutritional status of the Spanish population has been assessed in several studies (Aranceta Bartrina & Serra-Majem, 2000; Ruiz Moreno et al., in press; Varela-Moreiras et al., 2012), where insufficiencies have been observed in target and vulnerable population groups such as women of childbearing age, children and the elderly. Average dietary folate intakes in adults were estimated to reach approximately 250 μg per day, that is, ca. 60% of Recommended Intakes for Spanish population (400 μg) (Moreiras, Carvajal, Cabrera, & Cuadrado, 2011). According to these studies, major folate dietary sources were green leafy vegetables and legumes. Nevertheless, until date there is no reliable information on the impact of fortified food consumption and FA intakes are probably underestimated. This is of concern, as researchers emphasize not only the potential deleterious outcomes of insufficient, but also of excessive FA intakes in vulnerable population groups (Fajardo & Varela-Moreiras, 2012; Lucock & Yates, 2009).

Food composition databases (FCDB) and Tables (FCT) are the basic tools for dietary intake assessment (Greenfield & Southgate, 2003). Currently, computerised databases are considered a better option for food composition data management due to the many benefits they provide when compared to printed FCT. European FCDB have been successfully integrated, harmonised and managed by the European food information resource network of excellence (EuroFIR) project, from which Spain was part with the development of the Spanish Food Composition Database Network (Red BEDCA) (Ros, Martínez de Victoria, & Farran, 2009). Spanish FCDB

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and FCT do not always include a complete inventory of fortified foodstuffs. Namely, FA and folate data in European databases was found to be limited as reviewed by a number of researchers (Bouckaert et al., 2010; Verkaik-Kloosterman, 2009). Estimation of population's micronutrient intakes can be therefore hindered, not only for the relative lack of compositional data, but also because of the changing availability of these products for the consumer. Many studies highlight the lack of FA compositional data as a main drawback in order to assess the impact of these products on the population's dietary folate intakes (Sacco & Tarasuk, 2013).

Since 2007, our research group has been actively compiling food folate composition data comprising FA fortified products as well as non-fortified foodstuffs. A database was designed and developed in order to store and retrieve data, including label information declared by manufacturers and data available from other FCDB and Spanish FCT. The first version of our FCDB included data from 277 FA fortified products classified into 6 food groups (Samaniego-Vaesken, Alonso-Aperte, & Varela-Moreiras, 2009). *Cereals and derivatives* was the most commonly fortified food group (52% of total recorded products). We also studied the actual fortification levels by analysing a representative number of these products ($n = 93$), and we found that the addition of FA overages (i.e. adding higher than declared levels) was a common practice amongst manufacturers. This was observed in ready-to-eat breakfast cereals (RTEC) and dairy products, when analysed total folate was compared to label declared FA values (Samaniego-Vaesken, Alonso-Aperte, & Varela-Moreiras, 2010).

Data from this FCDB allowed us to perform FA intake assessments for a vulnerable population group using published food consumption data for children aged 2–13 years from the enKid Study (Aranceta Bartrina & Serra-Majem, 2000; Samaniego-Vaesken, Alonso-Aperte, & Varela-Moreiras, 2013). A comparison between potential FA intakes by fortified vs. unfortified food in women at a reproductive age was also performed (Samaniego-Vaesken, 2013). The first study's main finding was that FA fortified ready-to-eat breakfast cereals (RTEC) and cow's milk could provide, on average and respectively, 6–21% and 54–136% of children's Recommended Dietary Intakes (RDI) per serving. In the second study, FA potential intake was calculated using breakfast menu simulation models as established in accordance with national dietary guidelines (Dapcich et al., 2004). While folate content of unfortified breakfast models was on average 4–23% of women's folate RDI, the inclusion of just one single FA fortified food provided, on average, 20–60% of RDI.

In summary, the Spanish market offers a number of FA voluntarily fortified foods which are commercialised with diverse levels of added vitamins. This widespread availability can lead to a beneficial contribution to folate intakes for some population groups, but could also pose a potential risk of excessive intakes for vulnerable ones, depending on food consumption patterns. Our objective was to update and further improve our food folate FCDB database according to international guidelines, including information on food labels and analytical data, in order to achieve a reliable data compilation for nutritional assessment. A description of the most important aspects of voluntary food fortification practices in our country was also considered.

2. Materials and methods

2.1. Database design and data compilation

A product inventory compilation was conducted by visiting main retail stores in Madrid Region, performing web-based searches and requesting information to manufacturers. Supermarket chains were chosen based on their market share (Mercasa, 2012). Folic acid fortified food products were identified by their label's ingredient list providing it declared the inclusion of one of the following synonyms: folate, folacin, folic acid or vitamin B_9. Label data was recorded in standardized forms that included FA and vitamins B_6 and B_{12} content per 100 g or mL, recommended serving size (g or mL), nutritional and/or health claims and a digital image of the package and label. Microsoft Office Access® software was selected to design and build the database. Product compilation work started in December 2007 and the last update was completed in May 2010. The LanguaL™ food description Thesaurus and EuroFIR guidelines were adopted, including food group codification scheme (Møller & Ireland, 2009).

2.2. Folic acid and folate quantification in fortified food products

A representative percentage of products from each food group was analysed for total folate, FA and (6S)-5-methyltetrahydrofolic acid ((6S)-5-CH3-H4PteGlu). Two batches of randomly selected samples were purchased and independently grounded and stored at −20 °C. Samples were processed and analysed in triplicate under subdued light, minimising contact with air and within a month of their acquisition. Complete analytical procedure has been published elsewhere (Póo-Prieto et al., 2006; Samaniego-Vaesken et al., 2013). Shortly, samples were subjected to a trienzyme extraction method (Martin, Landen, Soliman, & Eitenmiller, 1990), total folate was analysed by microbiological assay with chloramphenicol resistant *Lactobacillus casei* as the assay organism (O'Broin & Kelleher, 1992) and an affinity chromatography followed by HPLC separation and quantification (Póo-Prieto et al., 2006) was used to determine FA and (6S)-5-CH3-H4PteGlu. Methods were compared for concordance, and a Standard Reference Material (SRM 1846) (Sharpless et al., 1997) and an International Standard (Thorpe et al., 2007) were used to test for accuracy. Individual product results from each method of analysis were included in our FCDB under EuroFIR component identifiers for each folate vitamer (Møller et al., 2008).

Shapiro–Wilk test was used to determine normality of data and Mann–Whitney's *U* test for non-parametric samples ($p < 0.001$ significance level) was chosen to compare declared with analysed folate values. Statistical analysis was performed using the SPSS 17.0 statistical package for Windows®.

3. Results and discussion

3.1. Food composition database

A comprehensive database was designed, developed and updated with the use of Access® software. From the initial design which included 277 products (Samaniego-Vaesken et al., 2009) four updates were fulfilled, approximately one per year between December 2007 and May 2010. Improvements in the FCDB included content harmonisation and organization in order to make data readily interchangeable with international databases. First, the LanguaL™ system was adopted for food group and subgroup classification and codification system (Møller & Ireland, 2009). Component identifiers for individual folate vitamers were included to differentiate the label declared folate entities (synthetic FA) and those analysed in the food products (Møller et al., 2008). Database contents were translated from Spanish to English and scientific names for common foodstuffs were added.

A total of 375 FA fortified products classified into 6 food groups and 17 subgroups were collected in the FCDB in accordance with the "*LanguaL™ Thesaurus EuroFIR classification system*" (Møller & Ireland, 2009). Main FA fortified food groups were *Grain or grain products* (46%), *Products for special nutritional uses* (25%) and *Milk,*