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Erythrocyte fatty acid status in a convenience sample of residents of the Guatemalan Pacific coastal plain

Noel W. Solomons [a], Eileen Bailey [b], María José Soto Méndéz [a], Raquel Campos [a], Klaus Kraemer [c,d], Norman Salem Jr. [b,*]

[a] Center for Studies of Sensory Impairment, Aging and Metabolism (CeSSIAM), Guatemala City, Guatemala
[b] Nutritional Lipids, DSM Nutritional Products, Columbia, MD, USA
[c] Sight and Life, Basel, Switzerland
[d] Department of International Health, Johns Hopkins Bloomberg School of Public Health, Baltimore, MD, USA

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ABSTRACT

We report the fatty acid composition, and in particular, the n-3 and n-6 long-chain polyunsaturated fatty acids (LCPUFA), in erythrocytes from a convenience sample of 158 women and 135 schoolchildren residing in the southern Pacific Coast of Guatemala. Erythrocyte fatty acids were analyzed by gas-liquid chromatography with flame ionization detection and the profiles were expressed as a weight percent; the Omega-3 Index values were also determined. Schoolchildren had significantly higher mean ARA and total n-6 fatty acid levels than the women. Women had significantly higher EPA fatty acid levels than schoolchildren, but the reverse was true for DHA. For mean total n-3 fatty acid concentration, women and schoolchildren had similar values. The red cell weight percentages of selected fatty acids were also similar in women and schoolchildren. As compared with erythrocyte fatty acid data from developed countries, Guatemalan women and schoolchildren had consistently lower LCPUFA values. The traditional diet of Guatemalans living in the Pacific coastal region provided a worse erythrocyte fatty acid profile than that typically obtained from a Western diet. Additional fatty acid composition studies with associated dietary intake data in other inland locations may be useful for the interpretation of the nutritional status of Guatemalan children and adults.

1. Introduction

The World Health Organization (WHO) attributes one-third of all yearly global deaths (15.3 million) to cardiovascular disease [1]. In low- and middle income countries, the prevalence of cardiovascular disease is increasing as populations shift from consuming traditional, healthy diets to those that are unhealthy, unbalanced, and characterized by high intakes of saturated fats, salt, refined carbohydrates, and low intakes of fruits, vegetable, and dietary fiber [1]. The widespread use of partial hydrogenation to produce *trans* fatty acids has also contributed to an even greater risk of developing cardiovascular disease. *Trans* fatty acids elevate low-density lipoprotein cholesterol levels and decrease high-density lipoprotein cholesterol levels [2,3]. The effects of this dietary shift or "nutritional transition" in low- and middle income countries will continue to worsen until unbalanced diets, obesity, and lack of physical activity are addressed, prevented and/or remedied [1].

Not all dietary fats and fatty acids have negative health consequences. It is important for both children and adults to consume adequate amounts and types of fats, as they serve important physiological functions [4,5]. The polyunsaturated fatty acids (PUFA) play key roles in cardiovascular health, normal brain development and immune function. These include alpha-linolenic acid (18:3n-3) and linoleic acid (18:2n-6), and especially the long-chain PUFA (LCPUFA) of the n-3 series, eicosapentaenoic acid (EPA, 20:5n-3) and docosahexaenoic acid (DHA, 22:6n-3), and of the n-6 series, arachidonic acid (ARA, 20:4n-6). In particular, these LCPUFA play a major role in prostaglandin and thromboxane metabolism, vision at the level of the retina and visual cortex, and the integrity of neuronal membranes [6]. They also act as important substrates,

Abbreviations: ARA, arachidonic acid; DHA, docosahexaenoic acid; EFA, essential fatty acid; EFSA, European Food Safety Authority; EPA, eicosapentaenoic acid; INCAP, Institute of Nutrition of Central America and Panama; LCPUFA, long-chain polyunsaturated fatty acids; PUFA, polyunsaturated fatty acids; WHO, World Health Organization; CeSSIAM, Center for Studies of Sensory Impairment, Aging and Metabolism

* Correspondence to: Nutritional Lipids, DSM Nutritional Products, 6480 Dobbin Road Columbia, MD, 21045, USA. Tel.: +1 443 542 2370.
E-mail address: norman.salem@dsm.com (N. Salem Jr.).

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structural elements, transporters, and signaling molecules [6]. Because humans cannot efficiently produce the specific molecular desaturation configurations for LCPUFA metabolism, the respective 18-carbon dietary precursors, alpha-linolenic and linoleic, and their respective longer-chain derivatives of the n-3 series, EPA and DHA, and of the n-6 series, ARA, are all classified as essential fatty acids (EFA) by many researchers [7].

Most of the epidemiological evidence related to the cardiovascular benefits of LCPUFA has been derived from long-term population studies of fish consumption and from randomized, controlled intervention clinical trials using fish oil with LCPUFA [1]. Many studies have shown clear evidence that consumption of LCPUFA is beneficial for heart health [8,9]. These studies have demonstrated that LCPUFA not only lower triglyceride levels but also provide modest lowering of blood pressure and heart rate in normotensive and mildly hypertensive individuals [10,11], reduce the risk of sudden death [12], reduce inflammation [13], and improve endothelial and vascular function [14]. The cardio-protective effect of LCPUFA is particularly strong among high-risk populations with a previous myocardial infarction [12]. Adequate nutritional status of LCPUFA is, therefore, important to reduce the morbidity of cardiovascular disease.

To determine LCPUFA status in individuals, bloodstream fatty acid composition is typically measured. Erythrocyte membrane fatty acid composition accurately reflects dietary fat intake in adults and children [15–17]. LCPUFA in erythrocytes are especially strongly correlated with dietary intake [18]. However, only a few studies have described erythrocyte fatty acid composition in children and adults. Many of these studies considered erythrocyte fatty acid data from individuals living in high-income countries [18,19,21,22] or those with medical conditions [23–25], and some studies have only small sample sizes [20].

In 2011–2012, CeSSIAM (The Center for Studies of Sensory Impairment, Aging and Metabolism) participated in a 10-month multi-micronutrient intervention study on the southern coastal plain of Guatemala (Province of Retalhuleu) using a fortified juice-flavored beverage. The fortified drink or an unfortified placebo was administered in a randomized fashion to adult women and schoolchildren residing in several rural communities. The nutritional beverage contained several vitamins and minerals, but neither formulation contained lipids of any kind. Blood samples were collected only at the end of the trial for erythrocyte fatty acid compositional analyses.

Nutritional studies in Guatemala began in the early 1950s with the establishment of the Institute of Nutrition of Central America and Panama (INCAP) [26]. INCAP was developed as an institution dedicated to research in clinical nutrition deficiencies in food and nutrition [26]. The initial studies of INCAP indicated that rural Guatemalans consumed a diet low in animal protein and calories from fat (7.5–12%) [27]. The traditional diet of Guatemala was based on maize derivatives (tortillas, tamales, gruels), black beans, rice, coffee, sugar and squashes [27]. As the result of their traditional dietary practices, Guatemalans had much lower serum cholesterol values than those observed in populations living in high-income countries [28,29] who consumed a much higher percentage of calories from fat.

Over the years, the results from dietary surveys in Guatemala continued to document relatively low fat contributions to total energy, in the 15–27% range [30,31]. In recent years, urbanization and globalization in Guatemala have contributed to nutrition transition [32]. Based on recent dietary data from a convenience sample of public and private schoolchildren in the western highlands city of Quetzaltenango, 29% of energy, on average, was obtained from fat [32]. Alpha-linolenic, EPA and DHA were the most limiting fatty acids, with less than 1% of total dietary lipids derived from LCPUFA [32].

The present study describes the erythrocyte fatty acid composition in 158 adult women and 135 schoolchildren from 12 Guatemalan communities located in the Province of Retalhuleu. Comparisons are made with erythrocyte fatty acid composition data from previous studies in other populations and with fatty acid levels in Guatemalan children from Quetzaltenango based on estimates from dietary intake data [32].

2. Methods

2.1. Setting and population

The collections for this study were conducted in March, 2012 in the Province of Retalhuleu on the Pacific Coast of Guatemala. This is a hot and humid location with maximum daily temperatures oscillating between 36° and 22 °C across the seasons. The mean altitude is 200 m above sea-level. The area is traversed by many rivers originating in the Guatemalan highlands. The province has about a quarter of a million inhabitants mostly residing in rural towns and hamlets dedicated to agriculture. Sugar cane, coffee, rubber, palm fruit are the principal cash crops while rice, beans and corn are grown for food.

The study locations included small, rural communities of families primarily dedicated to sugar cane cultivation and the associated sugar milling industries. A total of 18 communities provided subjects to the survey; most were participants in a service and assistance program run by FUNDAZUCAR (Guatemalan Sugar Grower's Foundation), a charity of the sugar industry. FUNDAZUCAR was established over 24 years ago to help develop and execute educational, health, and municipal strengthening programs and projects [33].

2.2. Subjects

The original candidate population for the multi-micronutrient intervention study included 1214 women (18–49 years old) and school-aged children (6–11 years old). Exclusion criteria included: evidence of a serious illness, regularly consuming dietary (vitamin and mineral) supplements, being pregnant or lactating at the time of blood sampling, or having diabetes or glucose intolerance. Subjects were prescreened with capillary blood glucometer readings ACCU-CHEK® Performa, (Roche Diagnostics, Indianapolis, IN, USA) in the fasting state. Women with a blood glucose of $\geq$ 126 mg/dL were excluded from the multi-micronutrient intervention study because the associated intervention involved daily consumption of a nutrient-fortified sugar-sweetened beverage.

After the prescreening for high glucose levels, a directed recruitment strategy was used to identify at least 240 adult women and 150 schoolchildren to meet the inclusion/exclusion criteria to participate in the main study. Eligible women in 12 of the initial 18 communities were identified, while eligible school-aged children were identified in 9 communities. Subjects were stratified and randomized first for inclusion, and later for assignment to the intervention treatment group. Initially randomized were 399 subjects, 243 adult women and 153 schoolchildren. At the end of the trial, there remained 293 individuals: 158 adult women and 135 schoolchildren. This represented an attrition of 26.5%, 34.9% for women and 11.7% for children. Viewed another way, this meant 153 subjects from the fortification arm (81 women and 72 children) and 140 subjects from the non-fortification arm (79 women and 61 children) who provided a satisfactory blood sample for fatty acid assay and inclusion in the analysis.

The study was approved by the Independent Human Subjects Committee “ZUGUEME” and authorized by local health and community authorities. The nature, intention, advantages and risks of