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Research paper

Status and interrelationship of toenail elements in Pacific children

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ABSTRACT

Objective: Elemental deficiencies or in excess effects growth and development. Pacific population are at a disadvantage due to food insecurity as compared to New Zealand European households. This study aims to evaluate the status and interrelationship of elements (essential, non-essential and toxic) in nine-year-old Pacific children who were part of the Pacific Island Families Study living in New Zealand.

Materials and

Methods: This observational study included 278 eligible nine-year-old children. Essential elements (including calcium, chromium, cobalt, copper, iodine, iron, magnesium, manganese, selenium, zinc, molybdenum), non-essential and toxic elements (arsenic, aluminum, antimony, boron, cadmium, lead, mercury, nickel,) were determined in toenails and after acid digestion, analysed using inductively coupled plasma mass spectrometry. Principal component analysis and multivariate analysis of covariance was used to identify differences in the groups of elements and the inter-correlations between elements.

Results: The mean calcium (868 μg/g Ca), selenium (0.35 μg/g Se) and zinc (129 μg/g Zn) concentrations were lower while the mean cadmium (0.21 μg/g Cd) lead (0.86 μg/g Pb) and mercury (0.72 μg/g Hg) concentrations were higher than the optimal health requirements. Ethnic differences in relation to toenail elemental concentrations were observed for aluminium and iron. Gender differences were observed for aluminium, antimony, arsenic and lead. Selenium and molybdenum were inversely associated with mercury. Manganese, zinc and calcium were positively associated.

Conclusions: This research contributes to the understanding of the elemental concentrations for Pacific children by using tissue samples from toenails, which improves the completeness of sampling than other tissues and provides a longer exposure time frame. The study also reports several inter-correlations between essential, non-essential and toxic elements in Pacific Island population.

1. Introduction

In New Zealand (NZ), there are large inequities in child health status especially within the Pacific population due to socio-economic disparities [1], and limited food security [2]. Food insecurity has been observed in up to 50% of the Pacific people's households in NZ compared to 23% of New Zealand European and Other (NZEO) households [2,3].

Food is the principal pathway for sources of nutrition which helps keep nutritional balance in the body. However, changes in nutritional patterns are dependent on several cultural, socio-economic, climatic and ecologic factors which determine the types of food available and consumed by the community [4]. For certain population groups (such as Pacific Island people), due to their economic circumstances, they may not have the required nutrition to maintain an optimum level of health. Nutritious diet plays a role in maintaining the required essential elements to support its physiological system. Due to better sample preparation procedures, sophisticated analytical techniques, and quality control validation [5], it is now possible to determine the concentration of most naturally occurring elements (especially essential) in the food chain and ultimately the human body [6]. With a greater understanding of how the human body uses these elements, comes an appreciation of their relationship with health and disease [7].

Human toenails are a specialised keratinious skin appendages (as in hair) that grow approximately 1 mm per month and the growth cycle is completed at 6 to 9 months [8]. Keratins are fibrous proteins that contain disulfide bridges [9] which are thought to chelate elements present during formation [10]. Depending on age, gender, health

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conditions and metabolic rates, nail growth rates differ [5]. A wide variety of elements are found in human toenails and many tend to exist at approximately the same order of magnitude in each type of toenail [11]. Elements in nails provide a time-integrated measure of body intake, especially toenails clippings which reflect a long exposure time frame given the relatively slow growth rate [12]. A 1.62 mm of nail sample corresponds to roughly 1 month of body nutritional status [5]. For example, the mean Se intake in time spans from one month to a year [13]. In addition, concentrations of toxic elements in nail tissues have been reported in order of magnitude higher than those of body fluids and other accessible tissues [14,15].

Nail samples, particularly toenails, are also less exposed to external contamination [16]. Just like hair, they are simple to collect, easy to analyse and store well [17]. Also, elements deposited into the nails are not subject to additional metabolic processes and many elements are present in the nail at substantially larger concentrations than in urine or blood [18]. Studies have examined the reliability of toenail measurements [19,20]. Although attenuation in measures of association may occur, the toenail concentrations of most elements are suggested to be useful biomarkers of exposure in which a single sample is assumed to represent long-term exposure [20].

Humans encounter exposures (some beneficial and some harmful) to elements in various chemical forms on a daily basis. As mentioned earlier, sources of exposure can be through ingested foods, multivitamins, and beverages, inhaled airborne particulates and absorbed compounds. The advantage of biomarkers is their potential for integration of all absorption pathways, yielding information on what is purported to be the most important actual body burden or status of the toxin or nutrient.

The concentrations of elements in nails have been studied in relation to health problems such as skin diseases, hypertension, diabetes, obesity, diet, smoking and drinking habits [21–25].

Therefore this study aimed to determine the concentration of essential elements [calcium (Ca), magnesium (Mg), manganese (Mn), molybdenum (Mo), copper (Cu), iron (Fe), zinc (Zn) selenium (Se) Nickel (Ni) iodine (I), antimony (Sb), chromium (Cr), cobalt (Co)], and non-essential/toxic elements (include Arsenic (As), Boron (B), Aluminum (Al) cadmium (Cd), lead (Pb), mercury (Hg)) in toenails of Pacific Island children living in Auckland, NZ as an indicator of their nutritional status.

2. Materials and Methods

An observational study was undertaken between July 2010 and July 2011 involving a sub-sample of nine-year-old Pacific children within a Pacific Island Families (PIF) longitudinal cohort in which these individuals have been followed since birth [26]. These children were born in Middlemore hospital, South Auckland, New Zealand to mothers in the year 2000 [26]. The eligibility criteria for the core PIF study was that at least one parent of the new born child identified themselves as being of one of the Pacific Island ethnicity and was a permanent resident of New Zealand. South Auckland has the highest number of Pacific Island population in New Zealand [1] from which this cohort was selected. The participants who were healthy were enrolled into this study at the same time as they were recruited into the PIF core study. Consent and assent was sought from children and their mothers before commencing this study. Children who provided assent but had short nails (typically less than 0.05 grams (g) of cut material) were excluded. This study was approved by the NZ health and disability ethics committee (NTX/07/05/050).

Toenail sample collections were conducted in the school setting. Approximately > 50 milligram (mg) of toenail clippings from feet (big toes and little toes) were placed, as they were clipped and collected onto a tissue paper. These were then placed in a sealed zip-lock polythene bag and each sample was identified with a unique code identification number to ensure anonymity when analysed. The samples were then stored at room temperature until laboratory analysis.

2.1. Toenail sample laboratory analysis

All toenail samples were washed prior to digestion to remove potential exogenous elemental contaminants derived from cosmetic treatments, 'dirt', etc. The washing procedure involved: (1) five steps using acetone, deionised distilled water (DDW, 18.2 MΩ) (x3) then acetone again; (2) at each washing step enough liquid was added to cover the sample and sonication (ranssonic water bath (T460/H)) for 5–10 minutes; and (3) decantation. Following the washing procedure the nail samples were dried overnight at 60 °C in a drying oven (LTE Scientific). Once dried, the sample was weighed (four decimal place analytical balance) and transferred to a pre-acid/DDW washed/dried Kjeldhal™ tube for digestion where 0.5 ml of concentrated nitric acid (Fisher Scientific, Trace Analysis Grade nitric acid) was added and the tube sealed with PVC Clingfilm. The Kjeldhal™ tube was placed in a hot block (Tecator 2012 Digestor) and heated at 160 °C for 30–60 minutes. Once the digestate was visibly clear, the Kjeldhal™ tube was removed from the heat and cooled and the digest solution transferred to a clean, weighed 15 ml centrifuge tube. The digested sample was weighed again (to four decimal places) and diluted 250 times (volume/weight) using DDW based on a dilution factor of 250. Due to some small toenail sample masses the 250-dilution factor was not sufficient to ensure there was enough volume of sample (4 ml) to be analysed, thus for some samples a higher dilution factor was used and noted. Before analysis the digest was filtered using a 0.22 µm syringe-driven filter unit (Millex®-GP, Millipore, Bedford, USA). Analysis of all the washed and digested nail samples was carried out by the Agilent 7700 x ICP-MS instrument (Agilent Technology, UK). The certified reference material (CRM) used were NIST SRM 1643e (National Institute of Standards and Technology, USA) and TMDA-54.4 (National Water Research Institute, Canada). The recovery rate range for all the elements was between 76% to 101.3%. All instrumental data for each element (according to the isotope selected) was reported as counts per second. The value was corrected for a reagent blank signal (to correct for any contribution from the digestion procedure) and ratioed with the internal standard isotope value (to correct any instrumental drift or signal enhancement/depression caused by the matrix). Data for the calibration standards was handled in the same manner and an Excel™ calibration curve produced for each element, with ratio signal (y-axis) and concentration of five standards (x-axis), from which the calibration equation was determined for calculation of the unknown toenail sample elemental concentration. The elemental values for each toenail sample were corrected for the dilution factor and the final values used in data analysis.

2.2. Questionnaires

Validated and reliable questionnaires were administered at the nine-year phase to both mother and child. These questionnaires were interviewer administered regarding socio-demographic, cultural, environmental, child development, family and household dynamics, lifestyle and health issues. Participant characteristics and demographic variables which were of interest for this study were included in the analysis:

• child's gender (girls, boys) and child Pacific ethnicity (Samoans, Tongan, Cook Island, and Others (which included Tokelau and Niuean), as determined by the mother at age two years.

2.3. Anthropometric measurement

Obesity was measured by standardized Body Mass Index (BMI), height to waist ratio and percentage body fat was also measured in this sample. Prior to data collection equipment was standardised, procedures documented in an operations manual and assessors trained. The average weight and height was calculated. Average weight and height