

Stable isotope techniques used to study link between gut health and child growth

By Jeremy Li

A large proportion of the population in low- and middle-income countries lives in an environment characterized by poor water, sanitation and hygiene conditions, which contribute to growth retardation in children. This is due to adverse modification of intestinal processes, which leads to improper absorption of the nutrients necessary for growth and other functions. This disturbance, originally referred to as environmental enteropathy, is now widely called environmental enteric dysfunction (EED) to reflect its multifaceted manifestations and effects.

A new IAEA-coordinated research project, approved in November 2016, is expected to provide a non-invasive, stable-isotope-based tool for diagnosis of EED in order to understand more clearly how this specific gut-related dysfunction affects the growth and health of children over longer periods of time in low- and middle-income countries. Nine countries from both developed and low-and middle-income settings are participating in this project, as technical experts in the case of developed countries and as research implementers in the case of low- and middle-income countries.

“It is of paramount importance to develop accurate, field-based, non-invasive methods to diagnose the condition,” said Victor Owino, a nutrition scientist at the IAEA. Nuclear-based stable isotope techniques offer the advantage that they can be used to assess multiple aspects of EED. (See The Science box.)

The project is studying the effect of EED on child growth and health in specific populations, using a stable isotope technique — the carbon-13 (^{13}C) sucrose breath test. This method has previously been used to assess non-EED-specific intestinal function. The assessment was based on the utilization of naturally ^{13}C -enriched sucrose (from maize).

Since maize and sugarcane are widely consumed in low- and middle-income

countries and already contain a lot of ^{13}C sucrose, natural enrichment may not be adequate. Therefore, the project will develop and test the usability of a more highly enriched ^{13}C sucrose breath test.

The carbon-13 sucrose breath test is based on the simple principle that, in the intestine, sucrose is broken down by a brush border enzyme called sucrase into glucose and fructose. When these are oxidized for use by the body, carbon-13 dioxide ($^{13}\text{CO}_2$) and water are produced. In abnormal circumstances, as in EED, sucrase enzyme activity and therefore $^{13}\text{CO}_2$ production may be reduced. In contrast, in normal circumstances, a strong and early release of $^{13}\text{CO}_2$ in the breath following an oral dose of ^{13}C sucrose indicates a healthy gut function. (See the infographic.)

“One way for this method to be more widely employed is to use commercially available highly enriched sucrose with synthetic ^{13}C stable isotopes,” said Owino.

Four experts from Australia, the United Kingdom of Great Britain and Northern Ireland (UK) and the United States of America (USA) are working on refining the existing ^{13}C sucrose breath test by using highly enriched ^{13}C sucrose, as described above, and validating the test against gut biopsy to identify gut dysfunction in EED.

First-ever EED study to track effect on longer-term growth using stable isotopes

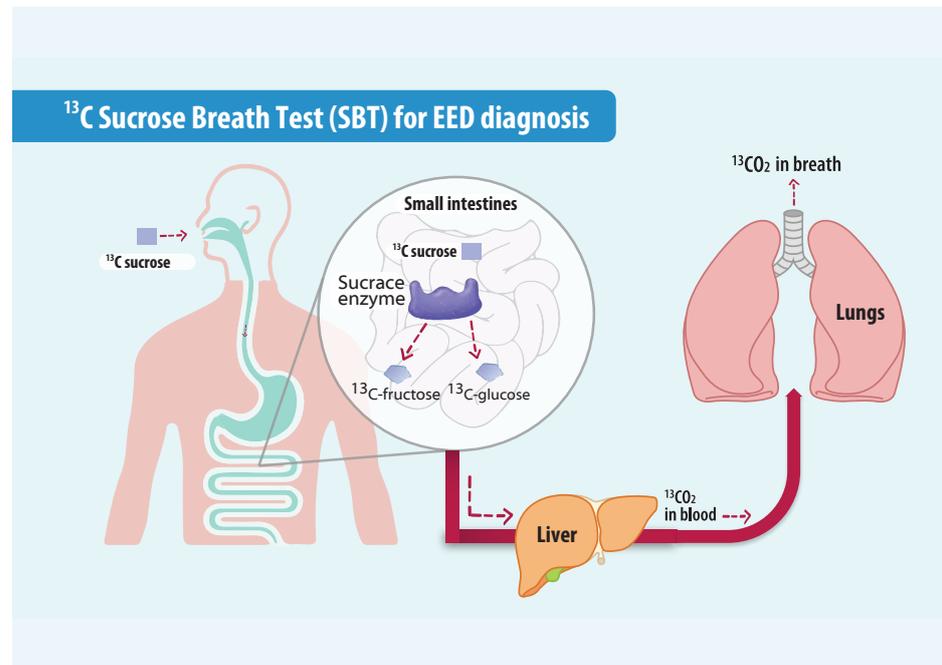
In 2015, the IAEA hosted a technical meeting to consider the current knowledge about EED and discuss interventions to prevent and treat the condition. “One of the conclusions was that more longitudinal studies are needed to better understand the fundamental causes of EED and that there is a need to develop a low-cost and widely applicable test,” Owino said.

The new study will, for the first time, measure how EED affects children over longer periods of time. Children will be retested three and six months after the initial tests to determine their growth during this time, Owino said.

Researchers from Bangladesh, India, Jamaica, Kenya, Peru and Zambia are participating in the study. When the testing technique is refined, they will use it to measure EED in children and assess its association with growth over time, Owino added.

In November this year, the IAEA will hold a meeting for stakeholders, technical contract holders and research contract holders from participating countries to harmonize the protocol, develop concrete plans and discuss the logistical details for the longitudinal studies. The experts from Australia, the UK and the USA will present details of the progress of optimization and validation of the ^{13}C sucrose breath test.

IAEA experts have also co-authored two scientific review papers on EED. The papers highlighted the nature of EED, its impact on child nutritional status and health and ways in which stable isotopes may be used to diagnose and manage the condition and its



(Infographic: F. Nassif/IAEA)

associated health effects. The reviews were published in the world-renowned journals *Pediatrics* (December 2016) and the *Journal of Pediatric Gastroenterology and Nutrition* (February 2017) and both were referenced in a comprehensive review by IAEA staff on use of stable isotopes in nutrition assessments, published in the prestigious *Proceedings of the Nutrition Society* (May 2017).

THE SCIENCE

What is environmental enteric dysfunction?

Environmental enteric dysfunction (EED) is a modification in intestinal function that seems to present in multiple ways that can be measured separately.

Key among these is that the intestine walls become unusually leaky (porous) and the shape of the tissues lining the intestines is altered, making them less fit to absorb food nutrients and prevent bacterial cells passing through.

Inflammation is another major manifestation in EED and is a natural response by the body to external invasion.

Limited nutrient passage or leakages combined with uncontrolled bacterial cell movement form a complex phenomenon that is thought to limit growth. Growth in children is driven by the growth hormone, which acts like a catalyst to trigger the addition of one block — referred to as a growth plate — to another to ensure linear bone growth from birth to puberty and sometimes beyond. Any process that limits the production or functioning of growth hormone leads to linear growth retardation (stunting). EED-related stress leads to reduced expression of growth hormone receptors in the liver, meaning that growth hormone signalling is inhibited.

The entirety of the microbial population in the digestive tract is called the microbiome. The microbiome is fundamental to human host function, immunity and survival. Stress conditions seen in EED result in microbiome immaturity and replacement of beneficial bacteria with harmful ones. This propagates infection that further adversely affects nutrient utilization and growth.

If the mechanisms underpinning growth retardation in EED are to be fully understood and interventions designed to prevent and treat it, sensitive techniques for diagnosis and classification must be developed for use in the field. Nuclear techniques will be a good addition in this endeavour.