THE VOICE OF THE **INC FOUNDATION**

NUTFRUIT

FOR THE **NUT AND DRIED FRUIT** WORLD

Edition 77. N° 2 **July 2019**

XXXVIII INC WORLD CONGRESS

Boca Raton Leads the Way for the Future of the Global Nut and Dried Fruit Industry

p. 42

Dr. Oz on How to Communicate the Message of Health

p. 49

www.nutfruit.org

INC international Nut&DriedFruit

Multi-Metabolite Biomarker Panels to Study Nut Intake

PROF. JORDI SALAS-SALVADÓ

PHD STUDENT ALESSANDRO ATZENI, DR. MÒNICA BULLÓ AND PROF. JORDI SALAS-SALVADÓ

Department of Biochemistry and Biotechnology, Human Nutrition Unit. Faculty of Medicine and Health Sciences, Institut d'Investigació Sanitària Pere Virgili (IISPV), Universitat Rovira i Virgili, Reus (Spain). CIBER Obesity and Nutrition Network (CIBEROBN), Carlos III Health Institute, Madrid (Spain).

In the future, multi-metabolite panels could be used in conjunction with classical methods of evaluating food consumption, to reduce measurement error when analyzing the associations between nut consumption and the risk of disease.

Nuts are one of the natural plant foods with the highest unsaturated fat content. They also contain protein and fiber, minerals such as magnesium, some vitamins (e.g., vitamin E), and other bioactive compounds, phytosterols and phenolic compounds (1).

Several cross-sectional and longitudinal studies have confirmed that nut intake is beneficially associated with some cardiovascular risk factors, a lower risk of coronary heart disease, cardiovascular disease and total mortality. Also, some epidemiological studies have found an inverse association between the frequency of nut consumption and some types of cancer or cancer mortality (2). In addition, some studies have demonstrated that the consumption of nuts may play a role in the prevention of metabolic diseases, such as type 2 diabetes, acting at the molecular level by modulating glucose and insulin metabolism (3).

Dietary assessment methods, such as food frequency questionnaires (FFQs), present several limitations related to self-reporting, e.g., the lack and omission of useful information about food sources, food processing, or storage conditions. These errors produce inconsistent results, especially when studying the links between food intake and disease causation. A complementary approach, totally independent of all the limitations mentioned above, is necessary in order to better understand the associations between nut intake and health outcomes (2).

Biomarkers of food intake (BFIs) are generally single, or a combination of, metabolites which indicate the consumption of specific foods in terms of time-response and dose-response after intake. Metabolomics is considered the most effective method to identify metabolites in biofluids with high sensitivity, and also represents a promising tool for food-intake biomarker discovery (4).

A recent review based on a comprehensive literature inquiry of both observational and human intervention studies, describes the principal BFIs of the most commonly consumed nuts (5). The results of this literature review reported information about the associations between nut intake and potential candidate BFIs according to the types of nuts, the study design, analytical method, sample type, and discriminating metabolites.

ALA (α-linolenic acid) represents 11% of the total fatty acid composition of walnuts and can be considered a potential candidate biomarker of walnuts (6). Urolithins are the product of polymeric ellagitannins (ETs) metabolized by gut microbiota and its presence in biofluids has been reported after walnut intake (7).

Non-targeted metabolomic approaches revealed for the first time an increased excretion of serotonin metabolites in urine associated with nut consumption (8). As walnuts have a higher serotonin content than other food sources, 5-hydroxyindole-3-acetic acid (5-HIAA), a serotonin metabolite, can be also used as a BFI for walnuts (9).

However, since these compounds are contained in many other food sources, none of them can be considered specific for walnuts when used alone. Therefore, a combination of these biomarkers can represent a more specific measurement of walnut intake to be used in the future in epidemiological studies.

Almonds and hazelnuts are high in α-tocopherol, which is one of the principal nutrient compounds of vitamin E. The combination with other molecules, such as flavan-3-ol-derived metabolites, can be used to obtain more specific information about almond intake (10).

As for pistachios, we have a very specific metabolite (the N-methyl-trams-hydroxy-L-Proline) that can be used as a marker of pistachio intake. This metabolite, contained in pistachios, has been identified in urine in the EPIDERM (Effect of Pistachio Intake on Insulin Resistance and Type 2 Diabetes Mellitus) controlled, crossover trial study (11). In this trial, changes in urine metabolome were detected in individuals exposed to a diet supplemented with pistachios, especially related to a modulation of some microbiota-derived compounds and other metabolites related with the tricarboxylic acid (TCA) cycle (11). However, not all the metabolites identified can be used as markers of intake.

Other studies reported the measure of lutein, zeaxanthin, β-sitosterol, and α-tocopherols to verify compliance with diets rich in pistachios, but as these compounds are common in many other foods, they cannot be considered a specific metabolite for pistachio intake (12).

Brazil nuts are one of the highest food sources of selenium and high levels of this compound have been reported after intake (13), but again, as this mineral can be found in several dietary sources and many factors can interfere in the measurement in biofluids, the use of this marker alone cannot provide a reliable intake assessment.

In conclusion, classic dietary assessment methods including FFQs are associated with measurement errors, and for this reason the use of dietary biomarkers has emerged as a complementary approach. The presence of distinct biomarkers for nuts cannot be considered specific due to the overlaps in the measurements. To establish a multi-metabolite biomarker panel in the future, it may be necessary to address a more specific measurement of intake. However, untargeted metabolomics is necessary in order to identify other compounds to include. The identified multi-metabolite panel could be used in the future in combination with classical food consumption methods to reduce measurement error when analyzing the associations between food intake and the risk of disease.

References

1. Ros E. Health benefits of nut consumption. *Nutrients*. 2010; 652–82. **2.** Garcia-Aloy M, Andres-Lacueva C. Food intake biomarkers for increasing the efficiency of dietary pattern assessment through the use of metabolomics: Unforeseen research requirements for addressing current gaps. *Journal of Agricultural and Food Chemistry*. 2018. **3.** Hernández-Alonso P, Camacho-Barcia L, Bulló M, Salas-Salvadó J. Nuts and dried fruits: An update of their beneficial effects on type 2 diabetes. *Nutrients*. 2017;9:1-34. **4.** Collins C, Mcnamara AE, Brennan L. Conference on 'Targeted approaches to tackling current nutritional issues' Symposium 2 : Developing and using novel health and nutritional biomarkers. Role of metabolomics in identification of biomarkers related to food intake. *Proceedings of the Nutritition Society*. 2019; 1-8. **5.** Garcia-Aloy M, Hulshof PJM, Estruel-Amades S, Osté MCJ, Lankinen M, Geleijnse JM, de Goede J, Ulaszewska M, Mattivi F, Bakker SJL, et al. Biomarkers of food intake for nuts and vegetable oils: An extensive literature search. *Genes & Nutrition*. 2019;14:7. **6.** Maguire LS, Sullivan SMO, Galvin K, Connor TPO, Brien NMO. Fatty acid profile, tocopherol, squalene and phytosterol content of walnuts, almonds, peanuts, hazelnuts and the macadamia nut. *International Journal of Food Sciences and Nutrition*. 2004;55(3), 171-178. **7.** Selma M V., González-Sarrías A, Salas-Salvadó J, Andrés-Lacueva C, Alasalvar C, Örem A, Tomás-Barberán FA, Espín JC. The gut microbiota metabolism of pomegranate or walnut ellagitannins yields two urolithin-metabotypes that correlate with cardiometabolic risk biomarkers: Comparison between normoweight, overweight-obesity and metabolic syndrome. *Clinical Nutrition*. 2018;37(3), 897-905. **8.** Tulipani S, Llorach R, Jáuregui O, López-Uriarte P, Garcia-Aloy M, Bullo M, Salas-Salvadó J, Andrés-Lacueva C. Metabolomics unveils urinary changes in subjects with metabolic syndrome following 12-week nut consumption. *Journal of Proteome Research*. 2011;10:5047-5058. **9.** Tohmola N, Johansson A, Sane T, Renkonen R, Hämäläinen E, Itkonen O. Transient elevation of serum 5-HIAA by dietary serotonin and distribution of 5-HIAA in serum protein fractions. *Annals of Clinical Biochemistry*. 2015;52(4), 428-433. **10.** Garcia-Aloy M, Rabassa M, Casas-Agustench P, Hidalgo-Liberona N, Llorach R, Andres-Lacueva C. Novel strategies for improving dietary exposure assessment: Multiple-data fusion is a more accurate measure than the traditional single-biomarker approach. *Trends in Food Science and Technology*. 2017;69, 220-229. **11.** Hernández-Alonso P, Cañueto D, Giardina S, Salas-Salvadó J, Cañellas N, Correig X, Bulló M. Effect of pistachio consumption on the modulation of urinary gut microbiota-related metabolites in pre-diabetic subjects. *The Journal of Nutritional Biochemistry*. 2017;45, 48-53. **12.** Hernández-Alonso P, Salas-Salvadó J, Baldrich-Mora M, Juanola-Falgarona M, Bulló M. Beneficial effect of pistachio consumption on glucose metabolism, insulin resistance, inflammation, and related metabolic risk markers: A randomized clinical trial. *Diabetes Care*. 2014;37(11), 3098-3105. **13.** Dumont E, Ogra Y, Suzuki KT, Vanhaecke F, Cornelis R. Liquid chromatography-electrospray ionization tandem mass spectrometry for on-line characterization, monitoring and isotopic profiling of the main selenium-metabolite in human urine after consumption of Se-rich and Se-enriched food. *Analytica Chimica Acta*. 2006;555(1), 25-33.