

PIVOTAL ASSESSMENT OF THE EFFECTS OF BIOACTIVES ON HEALTH AND WELLBEING. FROM HUMAN GENOMA TO FOOD INDUSTRY.

The PATHWAY-27 project (<http://pathway27.eu>) started in 2013 with the aim to better understand the physiological effects and mechanism of action of certain bioactive compounds and to develop bioactive-enriched foods (BEFs) with scientifically demonstrated impact on health, especially on metabolic syndrome.

Here we are, five years later and we have achieved a lot thanks to the close collaboration of the 25 partners in the PATHWAY-27 Consortium.

Three bioactive compounds (oat β -glucans - OBG, anthocyanins - AC and docosahexaenoic acid - DHA) alone or in combinations were used for enriching foods from three different food matrices (dairy, bakery and egg-based products). In each food matrix, three products were selected: The dairy products were milkshakes, puddings and combined desserts, the bakery matrix included biscuits, buns and breadsticks, while as egg-based products, pancakes, omelettes and egg beverages were chosen. In total, forty-five BEFs (3 categories * 3 products * 5 bioactive combinations) were developed and produced, which provided satisfactory chemical, physical, microbiological and sensory properties. BEFs were further prioritized based on sensory attributes, results of consumer acceptance tests, microbiological quality, and bioaccessibility and chemical stability.

The selected products (milkshakes, biscuits, pancakes) enriched with five different bioactive combinations were used in three 4-week human pilot studies run in three European clinical centres. The aim of the pilot studies was to identify the enrichments with the greatest beneficial physiological effect on volunteers. BEFs with the largest impact on blood lipids were then used in a 3-month multicentric, double-blinded, randomised, placebo-controlled human intervention study run in Italy, France, Germany and the United Kingdom, involving more than 300 subjects. Careful planning and close collaboration between the partners enabled smooth production and delivery activities of BEFs and placebo products for the pilot and 3-month human intervention study.

The dietary trial allowed not only the evaluation of clinical outcomes; various omics techniques (metabolomics, microbiomics, epigenomics, genotyping, and integrative analyses) were also applied on plasma, urine, stool, blood cells and fat biopsies collected from volunteers, allowing a comprehensive vision of the effects of BEF administration.*

Bioaccessibility and bioavailability of the bioactive compounds were investigated using a pig model. Effect and mechanisms of action of the bioactive compounds or their primary metabolites (propionate as metabolite of β -glucan and protocatechuic acid as metabolite of anthocyanin) were also studied at the cellular and molecular level. Liver and fat cell cultures served as representative models of the main organs involved in regulating certain risk factors of metabolic syndrome. Results showed that all bioactives decrease basal lipolytic activity and secretion of inflammatory markers in fat cells. In hepatocytes, they showed potential anti-diabetic effects and decreased lipid accumulation.

At the molecular level, the bioactive compounds induced epigenetic effects in preadipocytes, and docosahexaenoic acid altered the transcriptomes of both liver and fat cells decreasing the expression of genes related to fat and cholesterol synthesis.



PIVOTAL ASSESSMENT OF THE EFFECTS OF BIOACTIVES ON HEALTH AND WELLBEING. FROM HUMAN GENOMA TO FOOD INDUSTRY.

Activities performed in PATHWAY-27, especially the industry survey, enabled the project partners to identify the weak points and gaps in the knowledge associated with specific aspects of developing products with health claims. This information was used together with published evidence, guidance documents from the European Food Safety Authority, and the feedback from several consultations with key stakeholders from industry, academia and regulatory bodies, to develop two sets of Guidelines. The Guidelines for the food industry, mainly SMEs, are practical product development guidelines highlighting the special aspects of developing products with health claims. The Guidelines for the scientific community help to understand and apply the relevant steps of the health claim substantiation process (including how to design, conduct, report and interpret human dietary interventions) and are addressed to scientists from both academia and food industry. Both Guidelines are freely available from the PATHWAY-27 project website at www.pathway27.eu.

For optimal consortium performance, internal knowledge exchange was fostered through a series of webinars and a dedicated staff mobility programme. Last, practical training sessions and workshops organized by the consortium helped enhance external communication of the project results to all target audiences.

PATHWAY-27 contributed to a number of International events:

- International Conference on Foodomics -5th edition- Foodomics 2018 - from Data to Knowledge- 11-12 January 2018, Cesena, Italy;
- PATHWAY-27 Guidance Paper Workshops- 21-22 September 2017, Brussels, Belgium;
- International Conference on Food Science and Technology-13-15 November 2017, Rome, Italy;
- 1st International Conference on Food Bioactives and Health- 13-15 September 2016, Norwich, United Kingdom;
- PATHWAY-27 workshop -Next step in the development of products with health claims- Practical supportive tools for the development of the product with health claims- 5 May 2016.

PIVOTAL ASSESSMENT OF THE EFFECTS OF BIOACTIVES ON HEALTH AND WELLBEING. FROM HUMAN GENOMA TO FOOD INDUSTRY.

Several publications were prepared by the project members:

- Björk C, Wilhelm U, Mandrup S, Larsen BD, Bordonni A, Hedén P, Rydén M, Arner P, Laurencikienė J: Effects of selected bioactive food compounds on human white adipocyte function. *Nutr Metab (Lond)*. 2016, 13: 4
- Pineda-Vadillo C, Nau, F, Dubiard CG, Cheynier V, Meudec E, Sanz-Buenhombre M, Guadarrama A, Tóth T, Csavajda E, Hingyi H, Karakaya S, Sibakov J, Capozzi F, Bordonni, A., Dupont, D. In vitro digestion of dairy and egg products enriched with grape extracts: Effect of the food matrix on polyphenol bioaccessibility and antioxidant activity. *Food Res Int* 2016, 88: 284.
- Di Nunzio M, Valli V, Bordonni A (2016) PUFA and Oxidative Stress. Differential Modulation of the Cell Response by DHA. *Int J Food Sci Nutr* 67, 834-843
- Karakaya S, Simsek S, Tolga Eker A, Pineda-Vadillo C, Dupont D, Perez B, Viadel B, Sanz-Buenhombre M, Guadarrama Rodriguez A, Kertesz Z, Hegyi A, Bordonni A, Nehir Ela S Evaluation of stability and bioaccessibility of anthocyanins in bakery products enriched with anthocyanins. *Food Function* 2016, 7: 3488
- Pineda-Vadillo C, Nau F, Guerin-Dubiard C, Jardin J, Lechevalier V, Sanz-Buenhombre M, Guadarrama A, Tóth T, Csavajda É, Hingyi H, Karakaya S, Sibakov J, Capozzi F, Bordonni A, Dupont D: The food matrix affects the anthocyanin profile of fortified egg and dairy matrices during processing and in vitro digestion. *Food Chem* 2017, 214: 486
- Di Nunzio M, Valli V, Tomás-Cobos L, Tomás-Chisbert T, Murgui-Bosch L, Danesi F, Bordonni A. Is cytotoxicity a determinant of the different in vitro and in vivo effects of bioactives? *BMC Complement Altern Med* 2017, 17: 453
- Ghini V, Di Nunzio M, Tenori L, Valli V, Danesi F, Capozzi F, Luchinat C, Bordonni A. Evidence of a DHA Signature in the Lipidome and Metabolome of Human Hepatocytes. *Int J Mol Sci*. 2017, 18(2). pii: E359.
- Sebők, A. Hegyi, Zs. Kertész, Al. Bordonni: Importance of coordinated interactions of multiple stakeholders for developing products with health claims. *International Journal on Food System Dynamics*
- Valli V, Heilmann K, Danesi F, Bordonni A, Gerhäuser C. Modulation of adipocyte differentiation and proadipogenic gene expression by sulforaphane, genistein, and docosahexaenoic acid as a first step to counteract obesity. *Oxid Med Cell Longev* (2018), in press

and additional manuscripts are in preparation.

All project activities and outcomes can be found at the project's website (<http://pathway27.eu/>).

* At the time of publication of this leaflet, the results of the 3-month dietary randomised controlled trial were still being analysed please check the project website for updates.

Further information: <http://www.pathway27.eu/>

Dr. Alessandra Bordonni (project coordinator)
Alma Mater Studiorum UNIBO (Italy)
Tel: +39 51209 7861
E-mail: alessandra.bordonni@unibo.it

Dr. András Sebők (WP8 leader)
Campden BRI Magyarország Nonprofit Kft.(Hungary)
Tel: +36 1 433 1470
E-mail: a.sebok@campdenkht.com