

Highlights of Analytical Chemistry in Switzerland

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Metabolite Profiling Reveals that Dark Chocolate May Beneficially Modulate the Stress-related Metabolism in Humans

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Many studies have demonstrated the potential health implications of dark chocolate constituents, but rarely as a whole product. For instance, cocoa is rich in flavonoids, mainly flavan-3-ols, which are associated with benefits for cardiovascular health by maintaining low blood pressure, improving endothelial function, and by reducing thrombotic, oxidative and inflammatory states. Other cocoa-containing bioactive molecules include theobromine and amines (phenylethylamine, N-oleoyl- and N-linoleoyl-ethanolamine), which are reported to reduce blood pressure and act on the central nervous system metabolism, respectively. There is thus growing evidence on the health benefits associated with chocolate.

We have sought to capture a global view of the metabolic changes associated with chocolate consumption in healthy men and women using metabonomics. Nutrimetabonomics provides a system approach to assess the systemic metabolic status of an individual, which encapsulates information on genetic and environ-

mental factors, gut microbiota activity, lifestyle and food habits. We have used proton nuclear magnetic resonance (^1H NMR) spectroscopy and mass spectrometry (MS) as complementary analytical platforms to monitor metabolic changes associated with a daily intake of 40 g of dark chocolate over a period of two weeks in the urine and blood plasma of 30 individuals classified according to their self-reported anxiety trait.

Human subjects with higher anxiety trait showed a distinct metabolic profile indicative of a different energy homeostasis (lactate, citrate, succinate, trans-aconitate, urea, proline), hormonal metabolism (adrenaline, DOPA, 3-methoxy-tyrosine) and gut microbial activity (methylamines, *p*-cresol sulfate, hippurate). Dark chocolate reduced the urinary excretion of the stress hormone cortisol and catecholamines and partially normalized stress-related differences in energy metabolism (glycine, citrate, *trans*-aconitate, proline, β -alanine) and gut microbial activities (hippurate and *p*-cresol sulfate). **The study provides evidence that a daily consumption of 40 g of dark chocolate over a period of two weeks is sufficient to modify the metabolism of healthy human subjects.** Therefore, subtle changes in dietary habits are likely to modulate the metabolic status that might be associated with long-term health consequences, in particular *via* the activity of the symbiotic bacterial partners.

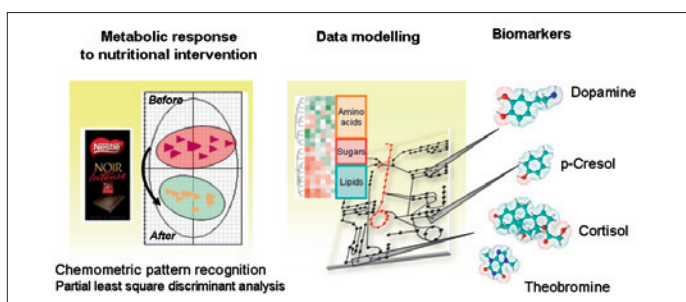
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Reference

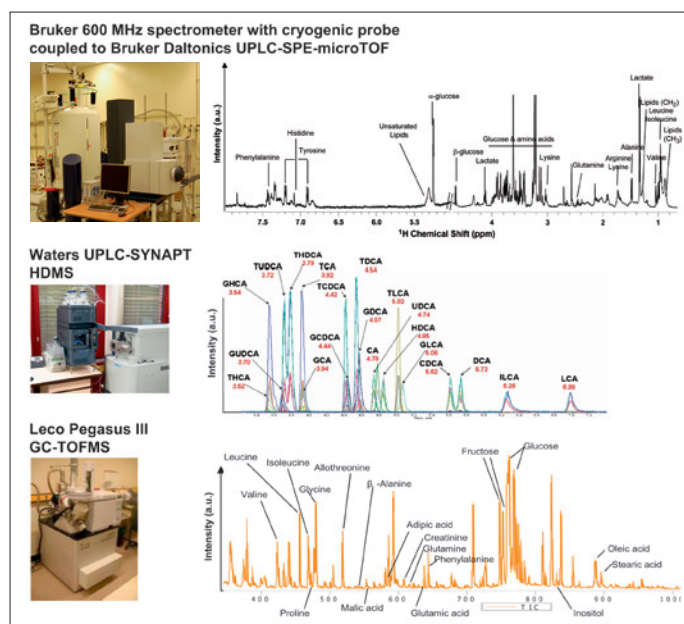
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The world of chocolate.



Flowchart of nutritional metabonomics. Chemometric and bioinformatic analyses allow modelling of nutrition-induced metabolic changes and identification of biomarkers associated with food habits and health benefits.



Typical NMR- and MS-based metabonomic analytical platforms. NMR offers a holistic profile of hundreds of metabolites with no *a priori* selection, while MS methods are commonly employed for global and targeted profiling. Both techniques are jointly employed to provide a snapshot of the metabolism and for biomarker identification.

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