Food metabolomics: META-PHOR
A new European research initiative

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ABSTRACT

There is an ever-growing demand for improved food-stuffs which better meet the needs of both industry and the consumer. With increasing emphasis being placed on health and safety-related aspects of our food, there is also a strong desire to develop new tools to monitor and improve the quality of what we eat. A new technology, metabolomics, has the potential to become one such tool of major value in this field. Although still in its infancy, metabolomics-based strategies are already being designed for a diversity of applications in, for example, food processing and quality control, plant breeding for improved crop varieties and in the development of novel foodstuffs. The importance of this technology has also recently been recognized by the EU Research division which now funds a multi-national metabolomics initiative focused on plant applications.

INTRODUCTION

The quality of our food in terms such as nutritional value, taste, fragrance and appearance is essentially determined by its biochemical composition (1). This biochemical profile is also a key factor in determining important food properties such as shelf life, nutritional stability and market value. European and global food policies continually demand even stricter monitoring and control of food quality (and safety) and there is a growing need for new tools to help us define and understand what we actually mean by 'quality' and how this can be effectively monitored.

Our pattern of eating is changing, not just in the Western world but now also in key developing countries such as China and India. This, combined with a generally more sessile working environment, increased amount of free time and a general reduction in manual labour is resulting in a phenomenal redirection of our general lifestyle in comparison to e.g. just 20 years ago. The increasing incidence of 'over-nutrition' (2) has been widely covered in the literature. Many of us are now living in what can be called the 'convenience era' where time is considered to be of the essence. Regarding food supply, growing demands for oven-ready meals and other ready-made food products (sauces, soups, puddings etc) have also led to an important new sector in the food industry. Furthermore, year-round demand for most if not all (fresh) products has also led to continual global redistribution of (out of season) food materials in every direction.

Consumers are however, becoming increasingly more aware of the importance of healthier eating and are becoming more critical of what they are being offered in shops and restaurants. Consumers are now also generally better informed and have a wealth of electronic information at their fingertips. Their expectations and demands are growing, particularly concerning the quality and safety of their food. Consequently, packaging information is taking on an increasingly important role and, to pay homage to consumer demands, producers and suppliers are looking to new ways to provide the required information in the most marketable way.

A new technology is emerging which, while still in development, has already demonstrated its potential as a valuable tool in helping us analyse the biochemical composition of our foodstuffs. This technology, metabolomics (see information box), allows us to analyse the biochemistry of complex mixtures and to filter out the key information of greatest relevance (1, 3). Metabolomics is currently gaining considerable attention in the medical world where it is under assessment as a tool for disease diagnosis as well as one of novel value in the development of innovative strategies for disease prevention and intervention (4). However, the greatest technological challenges are to be found in the plant field where we are faced with crops which are renowned for their capacity to be biochemically highly complex and to contain unique metabolite mixtures which change with development, age, environment etc (5).

What is metabolomics?

Metabolomics is a newly emerging science which can be seen as an advanced, specialized form of analytical biochemistry. This technology is centred around the detection of small molecules and, by definition, excludes the organic biopolymers such as proteins and fatty acids. Important small metabolites include e.g. amino and other organic acids, sugars, volatile metabolites and most of the diverse secondary metabolites found in plants such as alkaloids, phenolic components and coloured metabolites such as carotenoids and anthocyanins. Key to any metabolomics approach is the aim to gain the broadest overview possible of the biochemical composition of complex biological samples in just one or a small number of analyses. Liquid or gas chromatography (LC or GC) are usually used to separate the individual components in complex organic extracts after which Mass Spectrometry (MS) is employed to detect the metabolites present. Alternatively, Nuclear Magnetic Resonance (NMR) may be used. Characteristic to the technology is the large scale nature of the analyses performed - involving not only the semi-automated production of a large amount of complex data per analysis but also performing these analyses sequentially, on large numbers of samples. Highly complex data matrices are obtained - often of many Gigabytes. Consequently, metabolomics analyses can only be performed when all the necessary computing and bioinformatics tools are in place to allow automated data storage and efficient non-labour intensive data analysis. Metabolomics is usually used either for 'fingerprinting' samples to perform comparative analyses to detect differences of for 'profiling' where individual differential metabolites are identified for further analysis.

FOOD AND HEALTH

In most diets, plant products provide the main part of human food intake. The link between food and health (or rather, poor health) is also becoming increasingly clear. As reported by the

FAO/WHO in 2003, diets rich in (fresh) fruits and vegetables are generally associated with a reduced risk of developing several of the most important non-communicable diseases (NCDs) such as Type 2 diabetes, obesity, cardio-vascular disease and many forms of cancer (6). Emenaker & Milner also recently reported on the concept of "Eating for cancer" (7). There is also growing evidence indicating that diets which have a high plant component are associated with increased longevity (8). While it is sometimes difficult to prove scientifically and definitively that certain specific chemical components in our food are health-beneficial, there is growing evidence and belief that, for example, compounds such as phenolics (flavonoids, anthocyanins) which are present in most crop plants and are known to have anti-oxidant properties, induce effects conducive to reducing risk of CVD (9, 10). Food-related allergies are also greatly on the increase, especially in the western world and, while there may be different issues playing a role here, a direct link with food components (proteins, peptide ligands etc) has clearly been shown on many occasions (11). So-called bio-active compounds therefore have great potential influence on our (future) health and consequently, the desire to understand this better and to correlate physiological effects with biochemical composition continues to grow (Figure 1).

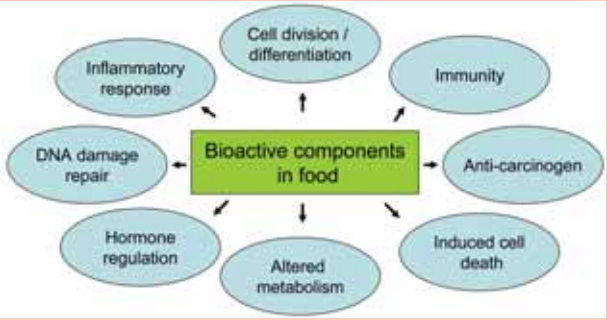

Figure 1. Certain components in our food have been shown to have a direct or indirect effect on certain events and processes in the human body related to health and disease (modified from (16))

With such knowledge we would be in a much better position to develop new targeted strategies for improving plant products either through directed breeding approaches or through improved food processing methods.

WHERE DOES METABOLOMICS FIT INTO A MODERN FOOD PRODUCTION STRATEGY?

Morris and Sands discussed recently the 'breeder's dilemma'. This dilemma has arisen because the demands driving western food production are switching in emphasis. While crop production has traditionally, almost exclusively been focused on yield, this strategy is generally considered to be counter-productive for improving food quality and human nutrition (12). Goff and Klee also proposed that current breeding strategies, employed for fruit crops such as strawberry and tomato, have lead to a major loss in key secondary metabolite volatile (fragrance / flavour) components (13). As many of these volatiles are derived from essential nutrient and health-promoting compounds such as fatty acids and carotenoids, this has led to a concomitant loss of information which these volatiles may provide regarding nutritional make-up and may detrimentally influence our eating pattern. Food processing strategies have also not given full attention to nutritional/quality aspects which is now required. Consequently, there is a clear role for new approaches such as metabolomics both as an information generating tool as well as a one for directing our activities towards new food and nutrition goals.
Metabolomics has a double role to play. Firstly, on the medical side where it can help guide future dietetic advice towards individualized health (14). Metabolomics has the potential to generate a comprehensive set of biomarkers which can be used to understand and monitor the interaction between food intake / uptake and human health and thus provide essential indicators as to how food should be produced and offered to meet modern health needs. Metabolomics is predicted to become a cornerstone in this field (2). Secondly, metabolomics shall be employed to direct breeding strategies to enhance specific desired balances of food components in fresh food which have been identified as being more optimal. In addition, the technology shall prove pivotal to the further optimization of food processing methods to help generate or preserve the desired nutritional balance in longer shelf-life strategies. However, before all this is possible in the most optimal way, the technology requires further development and fine-tuning.

META-PHOR - A NEW RESEARCH PROJECT FINANCED BY THE EUROPEAN COMMISSION

The tools needed to bring metabolomics technology to full maturity for application in the context of food science are highly diverse in nature. Successful development demands collaboration between a multitude of disciplines - analytical chemistry, food chemistry, plant biology, nutrition, computational science, bioinformatics and statistics (15). Initiating and facilitating such multi-disciplinary approaches is a primary goal of the Research division of the European Commission. Establishing an international consortium to bundle expertises and agree on common strategies and standards has also clear added value. The EU fund many such projects and one started in October 2006 is called META-PHOR (Metabolomics for Plants, Health and OutReach). The META-PHOR remit has been defined as follows: "To generate knowledge on those metabolites in our food which determine key characteristics such as nutritional value, quality and health by developing the advanced tools required for their detection. This knowledge will facilitate better monitoring of the food production chain and will create new opportunities for targeted strategies for breeding, storage and processing. Dedicated strategies for the biochemical fingerprinting and profiling of plant metabolites will be created. Broad application of metabolomics is currently restricted by a poor ability to generate, process and store data in a standardised manner. Incomplete compound databases also limit metabolite identification. META-PHOR will therefore bring together key experts with proven track records, in a coordinated, multi-disciplinary approach to deliver a phenotyping platform with potential for high throughput analysis. Statistics, bioinformatics and software tools will be developed and a sustainable database of plant metabolites will be initiated. META-PHOR shall focus on three crops - Brassica (Broccoli), melon and

Crop	Key Phytonutrients	Relevance	Profiling technology / Identification
Broccoli	Phytosterols Glucosinolates Flavonoids Vitamins Folate	Health applications Potentially anti-cancer Antioxidants, Health Nutritional value Health benefits	GC-MS / NMR LC-MS / FT-MS / NMR LC-MS / FT-MS / NMR LC-PDA-MS / NMR Bioassays / LC-MS / NMR
Melon	Isoprenoids Flavonoids Sugars Volatiles	Antioxidants / quality Antioxidants, Health Flavour / Taste Fragrance	LC-PDA-MS / NMR LC-MS / FT-MS / NMR LC-MS / GC-MS / NMR GC-MS
Rice	Micronutrients Volatiles Vitamins	Nutritional value Quality / market value Nutritional value	LC-ICP-MS GC-MS / NMR LCMS / GCMS / NMR

GC-MS: Gas Chromatography-Mass Spectrometry; NMR: Nuclear Magnetic Resonance; LC-MS: Liquid Chromatography-Mass Spectrometry; FT-MS: Fourier Transform-Ion Cyclotron-Mass Spectrometry; LC-ICP-MS: Liquid Chromatography-Inductively Coupled Plasma-Mass Spectrometry.

Table 1. A summary of the key groups of metabolites present in the META-PHOR crops chosen which are linked to nutrition, health promoting effects and quality

rice to enable us to cover the full range of key metabolite groups and micronutrients of importance to plant biology, crop quality and nutrition and which are key components regarding e.g. biofortification and nutrigenomics issues (Table 1).
The protocols and strategies defined shall be done so in a way which is broadly applicable to all other crops and food products beyond the three chosen core crops. Through an outreach programme we will demonstrate the opportunities of metabolomics to the general and scientific public."

WHAT CAN WE EXPECT FROM META-PHOR?

META-PHOR is dedicated to solving current limitations to the application of metabolomics technologies for research into food quality and safety. While the consortium has a strong technological orientation, we shall be working closely with crop scientists and nutritionists to enable the design of those technologies most needed to answer the questions and needs of both science and industry in the quest to meet consumer demands. This initiative has a strong European focus but not exclusively so. A deliberate choice was made to look at broader global nutrition issues and, in this regard, research partners in The Philippines (The International Rice Research Institute) and Laos shall play a crucial role in helping develop metabolomics specifically for rice applications. In addition, META-PHOR is also linked to the global 'Harvest Plus' initiative where great emphasis is being placed e.g. on issues concerning food micronutrients and biofortification for the developing world (www.harvestplus.org). The project is strongly research-focused but is fully committed to developing technologies for broad application. META-PHOR scientists will regularly appear at conferences to promote the technology and inform a broader audience of recent advances. Those partner scientists developing the analytical tools shall also interact with the nutrition partners to produce position papers on the value of the technologies for both quality control issues and for application in modern strategies to develop novel or improved food products for the fresh and processed markets which better meet the needs of a modified life-style and the demands of the modern health-conscious consumer.
Furthermore, the extra attention paid to food quality issues of particular importance to developing countries, either in terms of important local nutritional demands (e.g. aspects of micronutrient fortification) or regarding quality-related market value issues, such as rice fragrance, shall create new opportunities in this area. Metabolomics is anticipated to become a broadly applicable approach and further developments shall only increase its value and importance as a research and implementation tool.

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Figure 2. Metabolomics technology is based upon the application of advanced separation / detection equipment to produce highly complex datasets which are then analyzed and mined using dedicated software tools to extract the most relevant information correlated to the research goals in question

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