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RATIONALE FOR THE SEMINAR: FOOD, INTESTINAL IMMUNE SYSTEM AND GUT MICROBIOTA

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Food components, the gut microflora and the gastrointestinal immune system are in a constant interaction with each other. This continuous interference is generally referred to as the golden triangle as the interaction is considered crucial for intestinal health, proper and efficient immune protection, and physical well-being. This interaction can be boosted by food components that stimulate gut immune defence, and qualitative and quantitative aspects of the gut microflora. Commonly suggested food components with immune enhancing properties are pre- and probiotics, functional foods, nutraceuticals, etc. On the other hand, improper functioning of this golden triangle can result in chronic and persistent disease. Many of such diseases have involvement of an improper functioning gut. Based on such considerations the concept of immunomodulation has become important in recent years. The underlying hypothesis is that via selected food components, the functioning of the golden triangle can be influenced, leading to a persistently better functioning gut. Therefore, immunomodulation has become a leading principle to look for food constituents having such capacities. To better understand the current insights on mechanism of action, the search for useful biomarkers to detect immunomodulatory capacity *in vitro* and *in vivo* and the rational design of food components with these properties that could be added to the diet, we have organized this OHUS 2006 seminar.

IMMUNOMODULATORY FOOD COMPONENTS AS BIOFUNCTIONAL INGREDIENTS

Biofunctional ingredients are compounds that are present in or added to foodstuffs, and that have a health-promoting effect for the human body. The mechanisms underlying these positive effects can be very diverse, but generally they are strongly related to hot-button health issues like prevention of cardiovascular diseases, cancer and cara, enhanced well-being, longevity, *etc.* Examples are isoflavones, lignans, glucosinolates, sterols, and conjugated linoleic acid. For this proposal, biofunctional ingredients are distinguished from pro- and prebiotics. Probiotics are live microorganisms that are considered beneficial for humans. Prebiotics (or non-digestible oligosaccharides) are in principle a food supply for the beneficial bacteria in the human colon. Although pro- and prebiotics also have health-promoting effects for the human body and are added to food systems, we do not consider them as biofunctional ingredients here.

People are moving away from medical solutions to nutritional and lifestyle solutions. Supplementation of biofunctional ingredients in food, or improving the wholesomeness of food

(such as dairy products, breads, beverages, snack foods, condiments), is preferred over taking pills. With the escalating costs of health care due to a rapidly aging population biofunctional ingredients are expected to gain importance. Epidemiological and experimental studies have shown a relationship between important health issues and a high consumption of particularly vegetables and fruits. In some cases, compounds contributing to the health-promoting effect have been identified. However, there is a general belief that not a single component is responsible for this effect, but that the secret is actually in the overall composition of the mixture of components. The interplay between various food components clearly deserves much more attention than it has received until now.

The genetic constitution and age of a person determines to a large extent its health and sensitivity to illness. However, little is known on which food components trigger particular processes in the human body. The complexity of "food signalling" increases tremendously by the presence of the intestinal microflora. The total combined genome size of intestinal bacteria surpasses that of humans many times, indicating the enormous potential to modulate the functionality of food components here (both positively and negatively). An example of this is the conversion of plant isoflavones to equol. Equol is a much more potent ligand for the human oestrogen receptor than plant isoflavones; its estrogenicity is similar to that of (human) estradiol. However, it is important to note that (i) this conversion takes place in only approximately 15% of the human population, and (ii) this bioactive compound can be degraded by (other) microorganisms in the colon. This means that the potential of this bioactive compound in foodstuffs is used very inefficiently. Therefore, it will be advantageous to produce such compounds on an industrial scale, and add them as an ingredient to food stuffs, so that they can be absorbed in the small intestine, before arrival in the colon. It is important to verify that the new biofunctional ingredient is compatible with the food system in which it is incorporated. Solubility, stability, bioavailability and sensoric aspects should be taken into account.

During processing and storage of food (ingredients), many chemical conversions can take place, depending on the conditions used (temperature, pH, oxygen supply, *etc.*). As a consequence, the fine structure of desirable compounds may be altered. In some cases, their functionality may be coincidentally lost. It is then necessary to adjust the processing conditions. In other cases, the compounds are deliberately altered to enhance their functionality. Often, this is done by application of commercially available enzyme preparations (*e.g.* removal of sugar residues to improve bioavailability) or functional fermentations. With respect to improving health, the latter hold an enormous potential; health-promoting substances like arachidonic acid and β-carotene can be produced very efficiently using microorganisms. Fermentation of whole foods is also used to improve digestibility and utilization. Different kinds of analytical techniques (including HPLC, mass spectroscopy, *etc.*) will be employed to monitor structural changes during processing. With respect to functional fermentations, high-throughput techniques will be used to screen for microorganisms capable of performing desirable conversions, and to identify (and ultimately improve) the key enzymes involved in these reactions. Facilities to purify large amounts of the desired compounds are available.

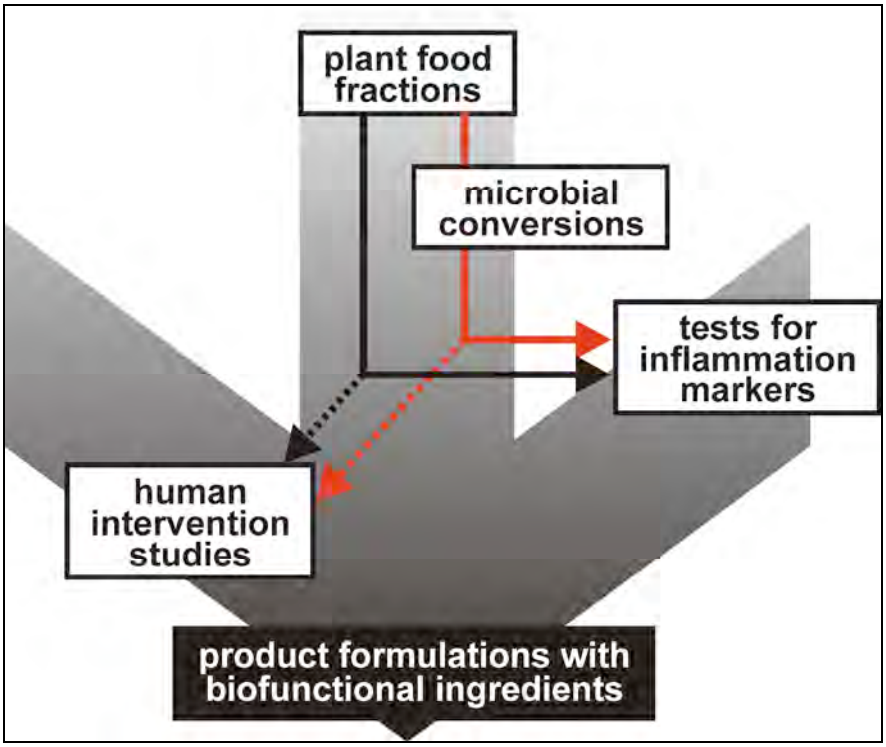

Figure 1: Schematic representation to arrive at immunomodulatory components for use in the treatment of human diseases.

Intake of vegetables and fruits is generally regarded as beneficial for human health, but the intake in Western European countries is low as compared to *e.g.* Southern Europe. Many of the Western diseases or discomforts, such as heart disease, cancer, inflammatory bowel disease, rheumatic arthritis are in one way or another related to chronic inflammation in specific target organs, like lungs (asthma), pancreas (diabetes), peripheral nervous system (multiple sclerosis), gut (morbus Crohn), skin (psoriasis), *etc.* Fruits and vegetables contain numerous beneficial components, and therefore the population in the western world is advised to increase their consumption. Over the years, the success of this advice has lost its persuasive force, and opportunities to improve health and prevent disease are sought more and more in supplementation of foods (such as dairy products, breads, beverages, snack foods, condiments) with these beneficial components as biofunctional ingredients. With the escalating costs of health care due to a rapidly aging population biofunctional ingredients are expected to gain importance. The causal factors for health-promoting effects of fruits and vegetables remain in many cases unknown, and it has been postulated that the mixture of compounds is responsible, rather than any one single component. Research strategies to identify mixtures of compounds, and to convert these to functional ingredients in product development, are currently lacking for this important group of plant foods. The intention of this project is to iden-

tify which specific compounds, or which combination of compounds, in vegetables and fruits play a protective role in persisting inflammatory reactions.

It is recognized that food components can be subjected to extensive microbial conversions during their passage through the intestinal tract. The total combined genome size of intestinal bacteria surpasses that of humans many times, underlining the enormous potential to modulate the functionality of food components. Examples showing that plant components can be converted by microbiota to more bioactive molecules are known, and their number is increasing rapidly. On the other hand, there are also examples indicating that the bioactive potential of plant components can be destroyed by intestinal bacteria. Therefore, specific focus will be on microbial conversions of plant-derived fractions, i.e. all fractions will be incubated with faecal slurries and tested for bioactivity as described for the untreated fractions.

Most research on the health-protecting properties of biofunctional ingredients is based on analyses with more or less purified compounds. However, their effectiveness in health protection is strongly dependent on so-called matrix-interactions: i.e. their interaction with the entire of other compounds in the food product and the physico-chemical environment in which they are embedded, in particular proteins, carbohydrates, fats/oils. The effect of processing conditions on the activity of biofunctional ingredients from processed food has been investigated to only a limited extent. For instance, it has been found that commercially processed seaweed had a lower antioxidant activity than fresh samples. This indicates that it is very important to investigate the remaining functionality after processing of food. Nevertheless, the number of interaction studies of biofunctional ingredients, particularly in relation to processing, is very limited. To our knowledge, no information is available on the interaction of such ingredients with components in beverages and milk products, as well as in more severe processed snacks. Another important aspect is that supplementation of food products with biofunctional ingredients should not negatively influence the sensory attributes of the products. The food industry requires insight in the so-called matrix-interaction in order to market attractive products with the appropriate biofunctional ingredients in its essential form.