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Irish Phytochemical Food Network (IPFN)

Irish Phytochemical Food Network

Key external stakeholders:

Food manufacturers, Food Ingredient companies, pharmaceutical and functional food manufacturers, Growers, Consumers, Scientific community, government authorities, plant breeders

Practical implications for stakeholders:

The overall objective of the Irish Phytochemical Food Network (IPFN) was to monitor levels of important phytochemicals along the food chain from '*farm to fork*' thereby gaining holistic knowledge of how each process affects the levels of these compounds. Phytochemicals are naturally occurring chemical compounds in plants, which may have protective or disease preventative prosperities when consumed. A network was formed to develop a critical mass of work in the area of plant based bioactive compounds, while coordinating and grouping existing knowledge and work in this area into a single, focused group. The project focused on the entire food production chain, from crop growing to new product development. As the IPFN brought together scientists with complimentary and diverse skills, this created synergies with expertise, sharing of skills, resources and equipment.

Main results:

- The selection of variety is as important as processing in the retention and overall quantity of bioactive compounds present
- The ability to detect phytochemicals in primary food products has been optimised for a number of important vegetable based bioactive substances
- Extraction of phytochemicals from the primary material has been optimised, as has their reincorporation into a number of food products
- The positive impact of these vegetable derived bioactive compounds on gut microflora has been observed in a model system
- Minimal processing and packaging can be optimised to retain phytochemicals, while a modeling technique has been developed to monitor the fate of these compounds from '*Farm to Fork*'

Opportunity / Benefit:

The work completed on the IPFN has identified and removed many barriers to the creation of food products based on plant derived bioactive compounds. Advances in the quantification of these compounds, the ability to synthesis phytochemicals to produce quantities required for biological tests, the optimisation of packaging and processing techniques will significantly aid the commercial sector to create new food products.

Collaborating Institutions:

UCD, UCC, DIT, UL, NUIG

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1. Project background:

Phytochemicals are compounds found in plants, that are not required for normal functioning (non-nutritive) of the body, but have a beneficial effect on health, either through their action as antioxidants or enzyme inhibitors. It is important to gain a greater understanding of the action and quantity of these compounds in our foods, as the nutrient content of vegetables does not fully explain the benefit to the body of their consumption. Therefore it's likely that the retention of naturally occurring levels of phytochemicals in whole, semi and full processed foods will have a positive impact on human health and therefore there may be commercial opportunities for companies in adopting strategies to retain these compounds in food stuffs. International trends point to an increased focus on developing varieties or rediscovering old varieties with favourable phytochemical profiles with the subsequent development of products which will be marketed on the basis of their phytochemical content.

The primary work of the IPFN focused on 3 phytochemical groups, the glucosinolates in brassicas, polyactylenes in apaciae and flavonoids in alliums. There were a number of significant gaps in knowledge and analysis which needed to be addressed, which included (1) the impact of variety and agronomy on the accumulation of these compounds, (2) the lack of rapid analytical methods to quantify these compounds within the food matrix, (3) a lack of understanding of the impact of consumption of gut microflora, (4) the impact of processing on the retention of these compounds, (5) modeling techniques to track these compounds through the food, (6) consumer perception to the supplementation of these compounds into food stuffs and (7) the ability and critical parameters to incorporate these compounds into food products. The IPFN focused on addressing these and other issues across five experimental work packages.

2. Questions addressed by the project:

- Can current techniques for the detection and quantification of phytochemicals from plant materials be improved?
- What impact does variety and agronomy have on the phytochemical content of vegetables?
- Does minimal processing impact on vegetable phytochemical content?
- How can phytochemicals be retained within processed foods?
- Can phytochemicals be supplemented into food stuffs in a manner acceptable to the consumer?
- What impact do phytochemicals have on gut microflora?
- Is it possible to model the fate of phytochemicals through the food chain?

3. The experimental studies:

To address the knowledge gaps identified in the project rationale a number of research approaches were taken. In order to address the gaps in the knowledge of agronomy and variety selection, a wide variety of broccoli, carrot and onions were grown in replicate, over a 2 year period at the same location, to negate for the impact of soil type and climate. A split-plot comparative study of organic and conventional agronomy, where soil management and crop protection, was run over a five year period. This allowed for a comparison of the major elements of agronomy on phytochemical accumulation to be separate and evaluated (WP2). Given the number of samples generated and the need for rapid analysis for other work packages it was necessary to develop and optimise phytochemical techniques. New methods for the detection of glucosinolates, polyactylenes and flavonoids were developed, significantly decreasing analysis time (i.e. reduction in analysis time for glucosinolates) (WP2). Once the phytochemicals of interest were extracted and

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quantified, they were screened against microorganisms of relevance to gut health, either in as extracted compounds, or as a whole plant extract (WP2).

The impact of processing on the fate of these compounds was assessed, which included minimal processing techniques, such as peeling or chopping, to more full processing techniques such as cooking. (WP4) Further studies identified the critical control points of retention of phytochemicals, which would facilitate their incorporation into prepared food products. To demonstrate this, the glucosinolate, sulforaphane was incorporated successfully into three prepared consumer foods (WP6). Probalistic models were used to map these different processes in the food chain supply from a farm to fork approach (WP5).

4. Main results:

- A range of improved validated analytical methods for assessing key phytochemicals, such as polyactylenes, glucosinolates and polyphenols were developed, significantly improving the quantification of these compounds but also reducing analysis time
- Technologies for the extraction and purification of polyactylenes have been developed, as have methods for the direct recovery of isothiocyanates from broccoli. The synthesis of racemic falcarinol, its oxidised version falcarinone, and its reduced version panaxjapyne A has been completed.
- The variety of vegetable had a significant impact on the quantities of phytochemicals recovered. Overall agronomic practice had a minimal impact on the quantity and range of phytochemicals recovered, however in some instances, such as in broccoli, there was a significantly higher accumulation of the glucosinolates glucobrassicin and neo-glucobrassicin in organically grown rather than in conventionally grown broccoli.
- Levels of polyactylenes in minimally processed carrots were affected by mechanical operations, primarily during abrasive peeling. Even if these operations are essential, there is a scope for optimisation of the depth of exposure of outer parts of carrots to carborundum drums, or alternate peeling methods such as rubbing/brushing operations could reduce the loss of polyactylenes.
- A probabilistic simulation model was developed to estimate the level of polyactylenes in carrots. Monte Carlo simulation techniques were used to simulate the effects of pre and post-harvest processing factors at various stages (farm to fork approach) and evaluate subsequent human exposure. The sensitivity analysis of the model highlights to food producers the importance of cultivar selection and agronomic factors which are equally as important as processing conditions on influencing phytochemical content.
- Minimal Processing research focused on critical control points for enhancement and retention of phytochemical anti-oxidants in fresh-cut vegetables and fruits. The results demonstrated that levels varied greatly among raw materials, notably in leafy salads and fresh-cut fruit salads, and that product formulation was the most effective step in controlling anti-oxidant levels.
- The effects of storage atmosphere were examined by investigating different levels of oxygen within modified atmosphere packages during storage. Use of low levels of oxygen from Day 1 retained much higher levels of ascorbic acid, but slowed the increase in phenols to maximum levels until about Day 7. It is concluded that the critical points for enhancing and retaining phytochemical anti-oxidants in fresh-cut products were raw material selection, mild processing (in most products), and storage under optimal modified atmospheres.
- Qualitative research, in the form of focus groups, was undertaken to explore consumer preferences for the inclusion of phytochemicals in foods was predicated on the perceived need for the product and belief in the benefit (delivered by the phytochemical). For example, parents of younger children viewed the product concept favorably for children who would not eat vegetables. Some also thought that the functional cheese would be of benefit to older consumers and those with an established illness. Younger groups did not perceive a personal need to consume disease preventing foods. For some participants, especially females, the preference was to consume the beneficial ingredient as a supplement. Others showed a strong preference to consume the natural ingredient source (e.g. broccoli), rather than from any other source.
- Overall it appears that functional foods containing phytochemicals may be met with hesitation and caution by some consumers. Using established and accepted functional food concepts may improve acceptance, e.g. such as the addition of phytochemicals to fortified milk. Furthermore, clear unambiguous communication of less familiar concepts such as phytochemicals and endorsement by trusted establishments will be central to consumer acceptance.
- The results revealed some new anti-microbials as well as phytochemicals (or extracts thereof) with positive effects on beneficial gut bacterial yet negative effects on the growth of the strains of opportunistic pathogens screened. Although strain-specific effects were noted for beneficial bacteria

(Lactocbacillus sp. tested), most of the polyacetylene, glucosinolate and isothiocyanate extracts tested either moderately enhanced growth of these bacteria or did not affect growth adversely. In fact, specific polyacetylenes (e.g. the S-stereoisomer of falcarinol), glucosinolates, isothiocyanates and bioflavonoids (e.g. quercetin-rich fractions) enhanced or promoted growth of most lactobacilli screened.

- Specific phytochemical fractions (e.g. polyactylenes, glucosinolates and isothiocyanates) inhibited the growth of bacteria regarded as opportunistic bacterial pathogens (e.g. strains of *Escherichia coli* and *Pseudomonas aeruginosa* tested) and most of the glucosinolate fractions were fungicidal to the *Candida albicans* strain tested. Specific glucosinolates (e.g. glucoiberin) were potently inhibitory to the growth of E. coli yet had little effect on the growth of most strains of lactobacilli at the concentrations tested.
- Broccoli seeds were used to develop wholegrain mustard and consumer sensory analysis found these products highly acceptable for all attributes, except texture which should be thickened slightly. Broccoli florets and the processing by-products stalks are an excellent source of vitamin C & A, protein, fibre & glucosinolates and their inclusion as freeze-dried powders in ready-soup formulations resulted in food products rich in bioactive isothiocyanates & sulforaphane. The sensory panel found both soups acceptable when compared to a commercial control and only found that colour was significantly affected.

5. Opportunity/Benefit:

The work completed on the IPFN has identified and removed many barriers to the creation of food products based on plant derived bioactive compounds. Advances in the quantification of these compounds, the ability to synthesis phytochemicals to produce quantities required for biological tests, the optimisation of packaging and processing techniques will significantly aid the commercial sector to create new food products.

6. Dissemination:

The Irish Phytochemical Food Network focused strongly on dissemination, with 4 individual workshops held during the project, attended by representatives of over 65 individual companies and over 40 primary producers. The dedicated project website had over 10,000 dedicated visits during the project. Over the course of the project, over 30 different researchers, at varying stages of their career contributed to the IPFN project, presenting their work at dozens of international and national conferences and workshops.

Main publications:

To date, over 30 peer reviewed articles have been published by the IPFN consortium. The following listed publications are indicative of the publications generated.

1. Alvarez-Jubete L., Valverde J., Kehoe K., Rai D., Barry-Ryan C. (2014). Development of a novel functional soup rich in bioactive sulforaphane using broccoli (Brassica oleracea L. ssp. italica) florets and byproducts, Food and Bioprocess Technology, 7, 1310-1321.
2. Tiwari U. and Cummins E (2013). Factors influencing levels of phytochemicals in selected fruit and vegetables during pre and post food processing operations. Food Research International, 50 (2), 497-506.
3. Tiwari U, Sheehy E, Rai D, Gaffney M, Evans P and Cummins E. (2015). Quantitative human exposure model to assess the level of glucosinolates upon thermal processing of cruciferous vegetables. LWT - Food Science and Technology , 63 (1), 253-261.
4. Reilly, K., Valverde., Finn, L., Gaffney, M., Rai, D & Brunton, N. (2015) A note of the feasibility of selenium supplementation of Irish grown Allium crops. Irish Journal of Agricultural and Food Research. 53(1): 91-99
5. Valverde, J., Reilly, K., Villacreces, S., Gaffney, M., Grant, J & Brunton, N. (2014) Variation in bioactive content in broccoli (Brassica oleracea var. italic) grown under conventional and organic production systems. Journal of the Science of Food and Agriculture. DOI: 10.1002/jsfa.6804
6. Rawson A, Brunton NP, Rai DK, McLoughlin P, Tiwari BK, Tuohy MG (2013) Stability of falcarinol type polyacetylenes during processing of Apiaceae vegetables. Trends in Food Science and Technology 30(2), 133-141.
7. Harris S, Brunton N, Tiwari U. and Cummins E. (2015). Human exposure modelling of quercetin in onions (Allium cepa L.) following thermal processing. Food Chemistry, 187 , 135-139

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