

Project title **Control of soil-borne diseases by the biofumigation effect of *Brassic***

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Background

Over the last years, in the search for a more environmental friendly pest and disease control, efforts have been directed towards a better understanding of the biological effects of natural products and the transformation products thereof. Plant extracts and crop residues have also been considered to have a potential role in sustainable agriculture, controlling pests and diseases, while minimizing dependence on artificial inputs such as pesticides. Special focus has been made on members of the *Brassicaceae* family, which have been shown to act as suppressors of pests and disease organisms. This effect is generally attributed to a range of biocidal compounds released into the soil when glucosinolates are transformed into bioactive compounds. Isothiocyanates have often been regarded as the most toxic among the various types of glucosinolate derived products, and they have been found to have nematocidal, fungicidal, insecticidal and herbicidal properties. However, the toxic properties of isothiocyanates can be seriously questioned when their high reactivity is taken into account. Disease control by Brassica species (termed as “biofumigation”) has normally been achieved by either including these plants as sole crops in the rotation (“break-crop” effect) or by using them as green manure, while their use as extracts has not been broadly investigated. However, before Brassica species could be used effectively, either as a sole crop in a rotation, as green manure or as extracts in formulations, more knowledge is required about their composition, the factors influencing their effectiveness in disease-control and their optimal frequency, temporal sequence and spatial distribution when included in crop rotations.

Objective

This PhD project is aimed at the achievement of knowledge on the processes involved in the biofumigation potential of Brassicas. Its overall approach is to study the glucosinolates produced by the Brassica plant, the glucosinolate-hydrolysis products and the effect of these compounds on soil-borne pathogens. The specific objectives can be defined as follows:

- To study the timing and the compartmentalization of the production of glucosinolates in certain Brassica species.
- To determine the type of products produced in enzyme catalysed hydrolysis of *Brassica* glucosinolates using active enzymes from these plants and evaluating the physico-chemical properties of products produced at different assay conditions.
- To study the biofumigation potential of the selected Brassica cultivars on specific pathogen targets using different methods.
- To determine the relative contribution and toxicity thresholds of the different glucosinolate-hydrolysis compounds in the biofumigation effect of the selected Brassica cultivars.
- To check the real applicability of biofumigation in an already established sustainable rotation (in collaboration with a Spanish agricultural institute).

Progress - 2006

PhD Courses:

Globalisation: threat or opportunity for Organic Farming? SOAR Summer School. September 2005. 4 ECTS.

Biochemistry and Natural Product Chemistry. PhD course at IGV, KVL. March 2006. 30 ECTS.

Potential of Organic Farming to contribute to Rural Development in Europe. SOAR Summer School. June 2006. 4 ECTS.

Experiments:

Glucosinolates and degradation products (cont.): Experiments were conducted by Capillary Electrophoresis in order to identify the type of compounds produced in enzyme catalysed hydrolysis of different glucosinolates from *Brassica* species at different degradation conditions.

Field experiments: Disease incidence in wheat plants following *Brassica* crops (*B. napus* and incorporated *B. carinata*) was determined in the crop rotation managed by ITGA in Navarra (Spain) as part of the collaboration established between this institution and KVL through this PhD project.

Conferences:

Bellostas, N., Mortensen, K. Sørensen, J. C., Sørensen, H. 2006. Structure properties of myrosinase isoenzymes from cruciferous plants defined by glycoside structures and small subunits. Poster at the First Glucosinolate Congress, 10-14 September 2006. Jena (Germany).

Bellostas, N., Hansen, L. M., Jørgensen, L. N., Kudsk, P., Madsen, P. H., Sørensen, J. C., Sørensen, H. 2006. Glucosinolate hydrolysis products for weed, fungi and insect control. Poster at the First Glucosinolate Congress, 10-14 September 2006. Jena (Germany).

Sørensen, H., Bellostas, N., Sørensen, J. C. 2006. Glucosinolates/myrosinase: analytical techniques. Workshop at the 2nd International Biofumigation Symposium. 25-29 June 2006. Moscow, Idaho (USA).

Bellostas, N., Sørensen, J. C., Sørensen, H. 2006. Effect of ascorbic acid and glutathione on the production of nitriles by myrosinase. Oral communication at the 2nd International Biofumigation Symposium. 25-29 June 2006. Moscow, Idaho (USA).

Bellostas, N., Casanova, E., Garcia-Mina, J.M. Sørensen, J. C., Sørensen, H. 2006. In Vitro screening of the effect of three glucosinolate derived nitriles on soil-borne fungi. Poster at the 2nd International Biofumigation Symposium. 25-29 June 2006. Moscow, Idaho (USA).

Bellostas, N., Kudsk, P. Sørensen, J. C., Sørensen, H. 2006. Glucosinolate hydrolysis products for weed control. Poster at the Joint Organic Congress, 30-31 May 2006. Odense (Denmark).

Bellostas, N., Sørensen, J. C., Sørensen, H. MECC-synchronous monitoring of substrate and products in the myrosinase catalysed hydrolysis of glucosinolates. Poster at the 20th International Symposium on Microscale Bioseparations (formerly HPCE Conference Series). 22-26 January, 2006. Amsterdam (The Netherlands).

Bellostas, N., Kachlicki, P., Sørensen, J. C., Sørensen, H. 2005. Glucosinolate types and concentrations in seedlings of different Brassica plants used for food. Poster at EuroFood Chem XIII, 21-23 September 2005. Hamburg, Germany.

Teaching:

Supervision of one bachelor student (September-January) in the project "Isothiocyanates from broccoli glucosinolates and their conjugation with Glutathione". Supervision during the experimental part and the writing process.

Supervision of one bachelor student (April-June) in the project "Study of the Conjugation of isothiocyanates and nucleophiles by MECC". Supervision during the experimental part and the writing process.

Plans – 2007

Experiments:

Bioassays: The study of the biofumigation potential of different glucosinolate degradation compounds on wheat and pea pathogens will be conducted in collaboration with Inabonos, Spain.

Field experiments (Continued): Disease incidence in wheat plants following *Brassica* crops (*B. napus* and incorporated *B. carinata*) will be determined again in spring in the crop rotation managed by ITGA in Navarra (Spain).

Publications

Bellostas, Natalia; Nielsen Kudsk, Per; Sørensen, Jens C. and Sørensen, Hilmer (2006) [Glucosinolate hydrolysis compounds for weed control](#). Paper presented at Joint Organic Congress, Odense, Denmark, May 30-31, 2006..

Bellostas, Natalia; Sorensen, Jens Christian and Sorensen, Hilmer (2006) [Micellar electrokinetic capillary chromatography—Synchronous monitoring of substrate and products in the myrosinase catalysed hydrolysis of glucosinolates](#). *Journal of Chromatography A*.

Bellostas, Natalia; Casanova, Esther; Garcia-Mina, Jose Maria; Sorensen, Jens Christian and Sorensen, Hilmer (2006) [In vitro screening of the effect of three glucosinolate derived nitriles on soil-borne fungi](#). Poster presented at Second International Biofumigation Symposium, Moscow, Idaho, USA, 26-29 June 2006.

Bellostas, Natalia; Sorensen, Jens Christian and Sorensen, Hilmer (2006) [Effect of ascorbic acid and glutathione on the production of nitriles by myrosinase](#). [oral] Presentation at *Second International Biofumigation Symposium*, Moscow, Idaho, USA, 26-29 June 2006.

Bellostas, Natalia; Kudsk, Per; Sorensen, Jens Christian and Sorensen, Hilmer (2006) [Glucosinolate hydrolysis products for weed control](#). Paper presented at Joint Organic Congress, Odense, Denmark, 30-31 May 2006; Published in *Proceedings of the Joint Organic Congress*.

Bellostas, Natalia; Kachlicki, Piotr; Sorensen, Jens Christian and Sorensen, Hilmer (2005) [Glucosinolate types and concentrations in seedlings of different Brassica species used for food](#). Poster presented at EuroFoodChem XIII, Hamburg, Germany, September 2005.