

Biofumigation 7 Symposium 2021

BOOK OF ABSTRACTS, Version 2



Editors: Vincent Michel, Aurélie Gfeller, Matthias Lutz, Matthew Back, Mohamed Besri



Biofumigation 7
plants for soil health



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INTERNATIONAL SYMPOSIUM ON BIOCIDAL AND NON-BIOCIDAL PLANTS TO IMPROVE SOIL HEALTH

22 - 25 MARCH 2021



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INTRODUCTION

Dear participants, dear readers

We are delighted to present you the 2nd version of the **Book of abstracts** of the **Biofumigation 7 Symposium**.

In this version of the Book of abstracts you will not only the abstracts of all the contributions but also the **questions** asked during the symposium and the **answers** of the authors! We would like to thank all the authors for the very complete and comprehensive answers, completed with numerous references, links to websites, figures and images!

This Book of abstract could also be called "**Video proceedings**"! In fact, nearly all the presentations (19 out of 23!) can be watched on the Agroscope Youtube channel! The link to the video is listed just below the abstract of the oral presentation.

To facilitate the navigation within the Book of abstracts, we added hyperlinks (blue and underlined). Therefore, you can rapidly access the presentations or posters you are interested in.

The seventh international Biofumigation Symposium was split into two events: Event-1 treated plants or plant-derived products used for biofumigation. Event-2 was on the use of plants that are used for other soil health related purposes. Based on the definition of J. Kirkegaard¹ plants in Event-1 were designated as biocidal, such in Event-2 as non-biocidal.

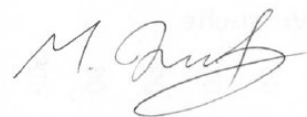
We hope you enjoy this final version of the Book of abstracts and are looking forward to meeting you possibly at the next Biofumigation Symposium which will take place in 2024 in Argentina.

The Organizers

Dr. Vincent Michel

Dr Aurélie Gfeller

Dr Matthias Lutz



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¹ John Kirkegaard. 2009. Biofumigation for plant disease control – from fundamentals to the farming system. In: D. Walters (ed.). Disease Control in Crops: Biological and Environmentally Friendly Approaches. Wiley-Blackwell, UK)

COMMITTEES

Organising Committee

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Aurélie Gfeller, Agroscope, Switzerland
Matthias Lutz, Agroscope, Switzerland
Rausis Chantal, Agroscope, Switzerland, Secretary

Scientific Committee

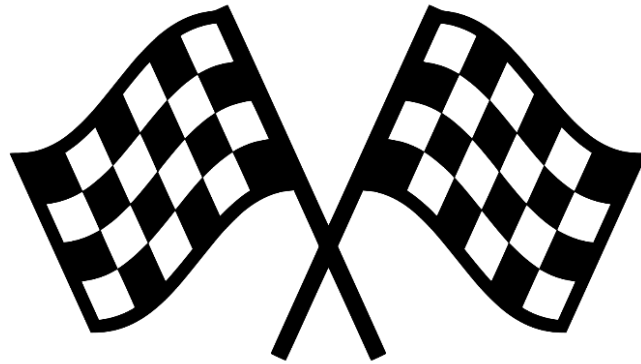
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Conference Secretariat

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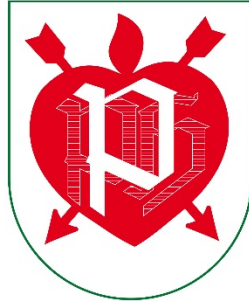
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Program Event 1: Biocidal plants (= plants used for biofumigation)



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Monday, 22 March 2021		
Day 1 - Moderator : Vincent Michel		
13.00	Welcome and information Information to the use of Hopin, presentation of Agroscope and Best4Soil	
13.30	Keynote: Crop production systems with reduced pesticide inputs: The role of biofumigation in sustainable pest, weed and disease management (01)	Back M. , Harper Adams University, Newport, UK
14.20	Functional Synergistic Biofumigation (FSB) as an innovative technology for controlling soilborne plant pathogens and root-knot nematode under intensive cropping systems in developing countries (02)	Salem M. F. , University of Sadat City, Sadat City, Egypt
14.50	Break	
15.10	Agronomical and biochemical characterization of some <i>Camelina</i> spp. accessions (03)	Matteo R. et al. , CREA-Research centre of cereal and industrial crops, Bologna, Italy
15.40	From the lab to the field: what we have learned in twenty years (04)	Gies D. , High Performance Seeds, Moses Lake, USA
16.10	<i>Brassicaceae</i> selection for Biofumigation purposes (05)	Montanari M. et al. , CREA, Bologna, Italy
16.40	Wrap-up - Summary of the day, final discussion of the day	
17.00	End of the day	
Tuesday, 23 March 2021		
Day 2 - Moderator : Matthew Back		
13.00	Welcome & information - Information to the use of	
13.10	Keynote: New tools and new application sectors for biofumigant cropping system (06)	Lazzeri L. et al. , CREA - Research centre of cereal and industrial crops, Bologna, Italy
13.50	Improving soil condition and yields using biofumigation across high value annual and perennial horticultural	Finningan J. , Australia EE Muir and Sons Pty Ltd, trading as Serve-Ag, Longford, Australia
14.20	Recent developments of the use of biofumigation for the control of soilborne diseases in Italy (08)	Infantino A. et al. , CREA, Roma, Italy
14.50	Break	
15.10	Isothiocyanates associated with <i>Brassica</i> species impedes the survival and foraging activity of the stem nematodes <i>Ditylenchus gigas</i> and <i>D. dipsa</i> (09)	Musa N. B. et al. , Harper Adams University, Newport, UK
15.40	On-Farm assessment of biofumigation and reduced tillage for soil-borne disease mitigation and soil health improvement in New York State, USA (10)	O'Dea J.K. et al. , Washington State University, Vancouver, USA
16.10	Horticultural crop health and yield and greenhouse soil conditions after 16 years of repeated treatments of biofumigation and solarization (11)	Mitidieri M. et al. , Universidad Nacional de Rosario, Instituto Nacional de Tecnología Agropecuaria, San Pedro, Argentina
16.40	Wrap-up - Summary of the day, final discussion of the day	
17.00	End of the day	
Wednesday, 24 March 2021		
Day 3 - Moderator : Matthias Lutz		
13.00	Welcome & information - Information to the use of Hopin	
13.10	Biofumigant cover crops - a promising strategy for soil and disease management in southeast Queensland (12)	Duff J. et al. , Department of Agriculture and Fisheries, Queensland, Gatton, Australia
13.40	<i>Origanum vulgare</i> vapour primes defence mechanisms in grapevine (<i>Vitis vinifera</i>) and hinders Plasmopara	Rienth M. et al. , Changins, haute école de viticulture et œnologie, Nyon, Switzerland
14.10	Biofumigation as a tool for a holistic approach to integrated wireworm population management (14)	Furlan L. et al. , Veneto Agricoltura, Settore Ricerca Agraria, Legnaro, Italy
14.40	Break	
15.00	Use of biofumigant seeds meal in liquid formulation to improve the soil fertility and to limit the symptoms of mortality in kiwifruit plants (15)	Mosso F. et al. , Agristore Srl, Nutrien SpA, CREA CI, Italy
15.30	Poster session Event 1 (P01 / P02 / P03)	Ait Kaci A.N. et al. / King L. M. et al. / Mitidieri M. et
16.15	Wrap-up - Summary of the day, final discussion, conclusion of Event 1	
16.45	End of Event 1	

Program Event-2: Non-biocidal plants (= plants used for other purposes)



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Thursday, 25 March 2021

Day 1 - Moderator : Mohamed Besri

13.00	Welcome & information Information to the use of Hopin, presentation of Agroscope and Best4Soil	
13.30	Keynote: Use of rotations, cover crops, and green manures for disease suppression in potato cropping	Larkin R. , USDA-ARS, Orono, USA
14.10	Keynote: Multi-service cover crops: towards a new paradigm for biocontrol and soil fertility enhancement	Couëdel A., Justes E. et al , CIRAD, Montpellier, France
14.50	Break	
15.10	Root exudate analysis of buckwheat and oat in the presence of redroot pigweed (18)	Gfeller A., Wirth J. , Agroscope, Nyon, Switzerland
15.40	New Rhizobiales strains isolated in North Italy from <i>Crotalaria juncea</i> Linn. nodules and production of	Kron Morelli R. , Agrifutur srl, Alfanello, Italy
16.10	Poster session Event 2 (P04 / P05)	Enouf J. et al. / Larkin R. P.
16.40	Wrap-up Summary of the day, final discussion of the day	
16.50	End of the day	

Friday, 26 March 2021

Day 2 - Moderator : Aurélie Gfeller

13.00	Welcome & information Information to the use of Hopin	
13.10	Keynote: Breeding of green manures and cover crops: Biofumigation, resistance, biocontrol and organic matter	Schlathölder M. , P. H. Petersen Saatzeit Lundsqaard GmbH, Grundhof, Germany
13.50	Use of Marigolds (<i>Tagetes</i> spp.) as cover crop for the control of tomato root knot nematodes (<i>Meloidogyne</i> spp.)	Besri M. , Institut Agronomique et Vétérinaire Hassan II, Rabat, Morocco
14.20	Eco-friendly alternatives against soil-borne diseases in strawberry cultivation in Martell Valley (South Tyrol, Italy)	Soppelsa S. et al. , Laimburg Research Centre, Laimburg, Italy
14.50	Break	
15.10	Is diversity of service plant species a way to archive higher biomass and N accumulation in winter oilseed rape - service plant intercropping? (23)	Bousselin X. et al. , Agroscope, Nyon, Switzerland
15.40	Novel microbial-based bioproducts improving soil biodiversity and the effectiveness of biocontrol and biofertilization practices in horticulture (24)	Mocali S. et al. , CREA - AA, Firenze, Italy
16.10	Plant-derived sources for anaerobic soil disinfestation in Southern California (25)	O. Daugovish et al. , University of California, Santa Cruz, USA
16.40	Wrap-up Summary of the day, final discussion, conclusion of Event 2	
17.00	End of the symposium	


List of posters

P01	Potential of Brassica cover crop and biofumigation to reduce <i>Verticillium dahliae</i> germination and Sunflower	Ait Kaci A.N. et al , Ecole d'Ingénieurs de PURPAN, Toulouse, France
P02	Examining biofumigant crops for the management of pea foot rot complex pathogens	King L.M. et al , Warwick University, Coventry, UK
P03	Biofumigation experiences in Argentina	Mitidieri M. et al , Instituto Nacional de Tecnología Agropecuaria, Argentina
P04	Sulphate catch cropping performances of rapeseed and mustard species	Enouf J. et al , INRAE Université Caen Normandie, France
P05	Soil microbiome characteristics associated with long-term potato cropping system management practices	Larkin R.P. , USDA-ARS, Orono, ME, USA

ORAL PRESENTATIONS

Oral presentations marked with this icon  are available on Youtube:


<https://www.youtube.com/playlist?list=PL5fL-uK8pWTFkBVReve1xr6hDx8pp0M4S>

01: Crop production systems with reduced pesticide inputs: The role of biofumigation in sustainable pest, weed and disease management (Keynote) 


Back M., Harper Adams University, Newport, United Kingdom

02: Functional Synergistic Biofumigation (FSB) as an innovative Technology for Controlling Soil-borne Plant Pathogens and Root-knot Nematode under Intensive Cropping Systems in Developing Countries


Salem M.F., Organic Agriculture Research Unit, Department of Environmental Biotechnology Department, Genetic Engineering and Biotechnology Research Unit, GEBRI, University of Sadat City, Egypt

03: Agronomical and biochemical characterization of some *Camelina* spp. accessions 


Matteo R. *et al.*, CREA-Research centre of cereal and industrial crops, Bologna, Italy

04: From the lab to the field: what we have learned in twenty years 


Gies D., High Performance Seeds, Moses Lake, USA

05: Brassicaceae selection for Biofumigation purposes 


Montanari M. *et al.*, CREA, Bologna, Italy

06: New tools and new application sectors for biofumigant cropping system (Keynote) 

Lazzeri L. *et al.*, CREA - Research centre of cereal and industrial crops, Bologna, Italy

07: Improving soil condition and yields using biofumigation across high value annual and perennial horticultural crops in Tasmania 


Finningan J., Australia EE Muir and Sons Pty Ltd, trading as Serve-Ag, Longford, Australia

08: Recent developments of the use of biofumigation for the control of soilborne diseases in Italy 


Infantino A. *et al.*, CREA, Roma, Italy

09: Isothiocyanates associated with Brassica species impedes the survival and foraging activity of the stem nematodes *Ditylenchus gigas* and *D. dipsa*


Musa N. B. *et al.*, Harper Adams University, Newport, United Kingdom

10: On-Farm assessment of biofumigation and reduced tillage for soil-borne disease mitigation and soil health improvement in New York State, USA 

O'Dea J.K. *et al.*, Washington State University, Vancouver, United States of America

11: Horticultural crop health and yield and greenhouse soil conditions after 16 years of repeated treatments of biofumigation and solarization 

Mitidieri M. *et al.*, Universidad Nacional de Rosario, Instituto Nacional de Tecnología Agropecuaria, San Pedro, Argentina

12: Biofumigant cover crops - a promising strategy for soil and disease management in southeast Queensland 

Duff J. *et al.*, Department of Agriculture and Fisheries, Queensland, Gatton, Australia

- 13:** *Origanum vulgare* vapour primes defence mechanisms in grapevine (*Vitis vinifera*) and hinders *Plasmopara viticola* infection ☐
Rienth M. *et al.*, Changins, haute école de viticulture et œnologie, Nyon, Switzerland
- 14:** Biofumigation as a tool for a holistic approach to integrated wireworm population management ☐
Furlan L. *et al.*, Veneto Agricoltura, Settore Ricerca Agraria, Legnaro, Italy
- 15:** Use of biofumigant seeds meal in liquid formulation to improve the soil fertility and to limit the symptoms of mortality in kiwifruit plants ☐
Mosso F. *et al.*, Agristore Srl, Nutrien SpA, CREA CI, Italy
- 16:** Use of rotations, cover crops, and green manures for disease suppression in potato cropping system (Keynote) ☐
Larkin R., USDA-ARS, Orono, United States of America
- 17:** Multi-service cover crops: towards a new paradigm for biocontrol and soil fertility enhancement (Keynote) ☐
Couëdel A., Justes E. *et al.*, CIRAD, Montpellier, France
- 18:** Root exudate analysis of buckwheat and oat in the presence of redroot pigweed
Gfeller A., Wirth J., Agroscope, Switzerland
- 19:** New Rhizobiales strains isolated in North Italy from *Crotalaria juncea* Linn. nodules and production of inoculants
Kron Morelli R., AGRIFUTUR, Italy
- 20:** Breeding of green manures and cover crops: Biofumigation, resistance, biocontrol and organic matter (Keynote) ☐
Schlathölder M., P. H. Petersen Saatzzucht Lundsgaard GmbH, Grundhof, Germany
- 21:** Use of Marigolds (*Tagetes* spp.) as cover crop for the control of tomato root knot nematodes (*Meloidogyne* spp.) in Morocco ☐
Besri M., Institut Agronomique et Vétérinaire Hassan II, Rabat, Morocco
- 22:** Eco-friendly alternatives against soil-borne diseases in strawberry cultivation in Martell Valley (South Tyrol, Italy)
Soppelsa S. *et al.*, Group for Berries and Stonefruit, Laimburg Research Centre, Ora, Italy; Research Group for Organic Farming, Laimburg Research Centre, Ora, Italy; Council for Agricultural Research and Economics (CREA), Research Center Agriculture and Environment, Bologna, Italy
- 23:** Is diversity of service plant species a way to archive higher biomass and N accumulation in winter oilseed rape - service plant intercropping?
Bousselin X. *et al.*, Agroscope, Plants and plants products, Nyon, Switzerland; USC LEVA, Ecole Supérieure d'Agricultures, INRAE, Angers, France; INRAE, UMR Agronomie INRAE, Thiverval-Grignon, France
- 24:** Novel microbial-based bioproducts improving soil biodiversity and the effectiveness of biocontrol and biofertilization practices in horticulture ☐
Mocali S. *et al.*, CREA - AA, Firenze, Italy
- 25:** Plant-derived sources for anaerobic soil disinfestation in Southern California ☐
O. Daugovish *et al.*, University of California, Santa Cruz, USA

01 Crop production systems with reduced pesticide inputs: The role of biofumigation in sustainable pest, weed and disease management

Back M.

mback@harper-adams.ac.uk

Harper Adams University, UK

Approaches to crop production are in a period of transition and the industry as a whole is placing greater emphasis on protecting soil, both physically and biologically. At the same time, agrochemical use is under greater scrutiny due to concerns over ecosystems and human health. For instance, the EU banned neonicotinoid insecticides on 27 April 2018 in connection with their impact on pollinators. Additionally, other agrochemicals could be lost due to their potential effect on endocrine disruption. In this respect, alternative approaches such as biofumigation have received far greater interest.

Biofumigation, using brassicaceous cover crops, can play an important role in crop protection due to the broad spectrum of pest, weed and disease targets that can be targeted. The technique relies on the release of volatile compounds, namely isothiocyanates, thiocyanates, nitriles and oxazolidine-thiones, from the disrupted brassica tissues following the hydrolysis of cell-bound glucosinolates by myrosinase. Biofumigation can be achieved either with freshly macerated brassica tissues from cover crops or using preserved materials that have been pelletised or formulated into a liquid or powder form.

In a recent study by Savary et al. (2019), global yield loss caused by pests and pathogens was estimated to be 10.1– 28.1% for wheat, 24.6 – 40.9% for rice, 19.5–41.1% for maize, 8.1– 21.0% for potato and 11.0–32.4% for soybean. This paper will explore the application of biofumigation for pest, weed and disease management and discuss the potential for variation between research papers. Understanding the potential causes of variation can ultimately improve the practice overall.

Keywords: pathogens, pests, weeds

Link to presentation on Youtube: <https://youtu.be/90WggFtKp2I>

Answers (A) from M. Back to questions (Q) asked after the presentation.

Q: (Frédéric Le Dily)

What about the ITC behaviour in the soil ? (adsorption on the Liquid/solid/gas phases ?

Degradation by microorganisms?)

Rearrangement with atmospheric components ?

A: You will find some good information on this topic in two publications: -

1. Gimsing and Kirkegaard (2009) – Glucosinolates and biofumigation: Fate of glucosinolates and their hydrolysis products in soil. See DOI: 10.1007/s11101-008-9105-5

2. Hanschen et al. (2015) - Degradation of Biofumigant Isothiocyanates and Allyl Glucosinolate in Soil and Their Effects on the Microbial Community Composition DOI: 10.1371/journal.pone.0132931

Q: (Susanne Vogelgsang)

Did you observe unwanted effects on beneficial soil insects and microbes?

If yes, was this quantified?

A: We did not measure or observe this in our work. However, you may be interested in the PhD work of Claire Wood. See Chapter 7 of her thesis: -

<https://theses.ncl.ac.uk/jspui/bitstream/10443/4129/1/Wood%2c%20C.%202018.pdf>

Also, take a look at the paper by Hanschen et al. (2015) DOI: 10.1371/journal.pone.0132931

Q: (Christian Kleimeier)

What would be a good biofumigation crop in an oilseed rape crop rotation?

A: Biofumigants can be included in a rotation where oilseed rape is grown. The biggest concern would be if club root (*Plasmodiophora brassicae*) was present at these sites. Most crops used in biofumigation are susceptible (good hosts) to this disease and do not cause reductions in the viability of resting spores. The exception is oilseed radish (*Rhaphanus sativus*). Other concerns might be diseases such as Verticillium wilt or pests such as cabbage root fly (*Delia radicum*).

Q: (John Duff)

Did you observe differences in GSLs between seasons as I found that a summer planting in QLD Australia resulted in more GSLs in some varieties, but not all.

A: Yes, we did. Biofumigants (*Brassica juncea* (Caliente 199), *Eruca sativa* (Nemat) and *Rhaphanus sativus* (Bento)) grown during the summer months had higher concentrations of glucosinolates than the same cultivars overwintered. For more detail, see Ngala et al. (2014) – DOI: 10.1002/ps.3849

Q: (Michaela Schlathölter)

Hi Matt, did you find differences in using fresh material or seed meal extract?

A: Unfortunately, we did not compare in the same experiment.

Q: (Lidia Sukovata)

How long does the effect of biofumigation last? Is there any way to extend the time?

A: The longer-term effects of the isothiocyanates from biofumigation will depend on the organism. We looked at cyst nematodes (*Globodera* spp) and stem nematodes (*Ditylenchus* spp.), which do not multiply without a susceptible host. Other organisms may be suppressed by biofumigation, but then multiply/reproduce and recover e.g. bacteria.

Q: If you use DSM, is it possible that too much meal (organic material) will reduce the effect of biofumigation?

A: As I understand, you are referring to the possible effect of sorption due to additional organic matter. In theory, it is possible, but I am not aware of any data to confirm or reject this effect.

Q: Is there any way to overcome the effect of low pH?

A: A low pH, leading to greater nitrile production, could be avoided by raising the pH using lime (calcium carbonate).

Q: (Soraya França)

I would like to ask a question concerning partial fumigation and beneficial microorganisms in the rhizosphere

A: Please take a look at the paper by Hanschen et al. (2015) DOI: 10.1371/journal.pone.0132931

Q: (Mariel Mitidieri)

Dr. Back, could you please tip the authors you are mentioning so we can look for the papers?

A: You should find the citations provided in the answers to the other questions.

Q: Amy Shi:

Have you test biofumigation on any perennial crops?

A: We haven't undertaken any work on perennial crops but other researchers have e.g. in vineyards (See Kruger et al. (2013) - <https://core.ac.uk/download/pdf/37420833.pdf>) and orchards (See Yim et al. (2016) – DOI: 10.1007/s11104-016-2876-3)

Q: Will the pest control effects last until crop year 2 and 3?

A: See my answer above.

Q: (Mohamed Besri)

Certified plants e.g. strawberry runners should be 100% free from any soil borne pathogen. Has biofumigation been used to produce such completely pathogens free plants? Any idea on the susceptibility of chlamydo spores of *F. graminearum* to ITC. in your presentation, you talked only on the conidia and mycelium susceptibility.

A: I doubt that biofumigants would ever provide 100% control and neither would their synthetic counterparts.

Unfortunately, we did not assess chlamydo spores but I agree that they should be considered as they may be more tolerant than conidia.

Q: (Frédéric Le Dily)

In order to optimize biofumigation process, where are the main targets ?
Physiologists and breeders (more biomass and GSL contents)?
Soil experts (where do the ITC go?) ?
Pathologists (which organisms are affected by nitriles or ITC ?)?
What is your feeling (note that I am a physiologist)?

A: There are so many factors that might affect the overall effect/success of biofumigation: -

The growing crop – exudation of GSLs from roots (relating to upregulation of exudation and volume of roots).

The concentration and type of GSLs in the biofumigant AND the biomass produced. These factors relate to biofumigant species/cultivar choice, the growth period of the crop and the inputs (N, S and water).

Crop maceration, when GSLs are at their peak (early flowering typically).

Ensuring optimal chopping and incorporation into moist soils.

See Kirkegaard and Matthiessen (2004) for an overview -

https://www.researchgate.net/publication/284099196_Developing_and_refining_the_biofumigation_concept#fullTextFileContent

Q: (Michaela Schlathölter)

What about Nitrogen leaching after biofumigation?

A: There was a nice paper (field observations) at the 5th Symposium of Biofumigation (an author called Paul Brown) that showed a reduction of nitrates in water sampled from field drains where oilseed radish was grown,

Q: As well as greenhouse gas production - is that a discussion somewhere?

A: See page 129 of our report for the AHDB for a short discussion of this topic: - https://projectblue.blob.core.windows.net/media/Default/Research%20Papers/Potatoes/R476%20Final%20Report_180419%20to%20publish.pdf)

Q: (Célia Seassau)

Could you please send us the reference talking about the effect of liquid formulation and powders of cover crop to regulate pest?

A: De Nicola et al. (2013) DOI: 10.1016/j.indcrop.2012.05.018 – Liquid formulation
Lazzeri et al. (2004) DOI: 10.1016/j.indcrop.2003.12.018 – Pellets

Q: (Nyambura Mwangi)

My question is on whether we are able to exploit most of the biofumigants in partial biofumigation?

Or is this linked to ability of different biofumigants being able to leach the GSLs through the roots, is it also related to root system of different brassica cultivars and where the GSLs are likely to be more concentrated?

A: We have observed partial biofumigation with Indian mustard (*Brassica juncea*) and oilseed radish (*Rhaphanus sativus*) but I suspect it would be possible with other species that exude suitable glucosinolates (ITC forming). Comparing root leaching between different species would be interesting. There is much more that needs to be done here.

Q: (Amy Shi)

Have you compared the cover crop, seed meal and synthetic biofumigants to see how they interact with pathogens and the host plants?

A: No, unfortunately we haven't done this yet.

02 Functional Synergistic Biofumigation (FSB) as an innovative Technology for Controlling Soil-borne Plant Pathogens and Root-knot Nematode under Intensive Cropping Systems in Developing Countries

Salem M.F.

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Organic Agriculture Research Unit, Department of Environmental Biotechnology Department, Genetic Engineering and Biotechnology Research Unit, GEBRI, University of Sadat City, Egypt

We adopted a new and innovative technology for controlling soil-borne pathogens and Root-knot nematode with a sustainable and eco-friendly appropriate for intensive agricultural systems in developing countries. This technology is suitable for use in organic or Global GAP vegetable and fruit production systems. We aimed to develop tactics that build healthy soils and promote microbial ecosystems that challenge these potentially devastating, broad spectrum pathogens. We adapted three innovative approaches of Functional Synergistic Biofumigation (FSB) to control soilborne diseases, root-knot nematode and aggressive weeds. These technologies can efficiently suppress soil-borne diseases, plant parasitic nematodes, and aggressive weeds instead of Methyl Bromide (MB) in one step and in a short period less than 25 days. This is the reason why this technology is suitable for intensive agricultural systems. We are working by these technologies here in Egypt and we developed an Ecofriendly biofumigated compost companies that produce more than 360,000 ton/year.

We adopted these new technologies i.e. Functional Synergistic Biofumigation (FSB). This paper will describe three main mechanisms as follows that can efficiently suppress soil-borne diseases, plant-parasitic nematodes, and aggressive weeds in different soil types worldwide. For example, in European countries, most of the agricultural soils are mainly acidic soils while on the other hand, most of the African and Middle East countries are mainly alkaline soils. We succeeded in solving these fatal problems in Egypt, Sudan, Tunisia, Ghana, Kingdom of Saudi Arabia and Pakistan. These modified mechanisms investigated were: 1. Functional Synergistic Physical–Biofumigation 2. Functional Synergistic Biocontrol–Biofumigation 3. Functional Synergistic Chemical–Biofumigation. We have to emphasize that each mechanism has certain advantages that suitable for each soil type and can be recommended under certain conditions. We are willing to share our experience in these novel technologies and its recommended applications to be used worldwide.

Keywords: functional synergistic biofumigation, soil-borne diseases, Root-knot nematode, aggressive weeds, biotechnology, intensive agricultural systems, developing countries

Link to presentation on Youtube: *This presentation was cancelled*

03 Agronomical and biochemical characterization of some *Camelina* spp. accessions

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Camelina (*Camelina sativa* L. Crantz) is an overlooked annual oleaginous crop with several interesting agronomical characteristics. In these last years, it has gained attention as a dedicated oilseed feedstock for biofuels, but Camelina shows also interesting features for industrial and food purposes. It can easily adapt to low input cultivation both in spring and in autumn sowing, representing a valuable minor oleaginous crop, acting also as a cover crop. In this work, a screening on a germoplasm starting from 45 *Camelina* spp. accessions from several countries was carried out in an experimental farm in Bologna, Italy. The trials were conducted using no agronomical input during cultivation. Different agronomical responses were observed and some of the tested accessions demonstrated interesting features compared to the *Camelina sativa* sel. ITALIA used as references: some accessions performed well, with interesting potential yields up to 3 ton/ha, while some others produced a low amount of seed or did not even survive. Despite differences in yields, yield components, and in total glucosinolate contents, the glucosinolate profiles resulted similar in terms of peak ratio for all the accessions. Further investigations were conducted on camelina oil and derived defatted seed meal for novel biofumigation purposes. The study highlighted an interesting potential of Camelina derived materials mainly for: i) oil to be used for oil-water emulsion in foliar biofumigant applications ii) defatted seed meal, characterized by the presence of thiofunctionalized glucosinolates, mainly 10-(Methylsulfinyl)decyl GSL (Glucocamelinin). This class of compounds is studied for the biological activity in plant pest and disease control as well as for their ability as monofunctional inducers of the phase II detoxification enzymes in mammal cancer cellular models.

Keywords: biorefinery framework, Brassicaceae, *Camelina* spp., fatty acids, glucosinolates, vegetable oil

Link to presentation on Youtube: <https://youtu.be/cOQFTctLfAM>

Answers (A) from R. Matteo to questions (Q) asked after the presentation.

Q: (Lidia Sukovata)

What about registration process for using as a plant protection product?

A: These presented products consist in defatted oilseed meals and oils obtained through mechanical defatting, so no chemical processes are involved in their production, for these reasons, they are not classified as ppp. At the moment, at least in EU, each Country has its own regulation. For example, in Italy, this material is classified as an amendment, and regulated in a law frame regarding fertilizers.

The presented studies had the aim of investigating the effect of these patented biobased products on some pests and pathogens, and only for scientific purposes.

Q: (Setu Bazie Tagele)

Can I ask about liquid formulation?

A: Liquid formulations consist in an oil-water emulsion in which the defatted seed meal is dispersed. In this condition the presence of water activate the myrosinase-glucosinolate reaction and ITCs are released. Since ITCs are hydrophobic compounds, the oil portion of the emulsion has the ability of holding them, thus creating a microfilm of oil enriched with ITC on the treated surface. The formulations are optimized to adapt both to foliar application on the epigeal portion of crops and root application through drip irrigation systems. The different approaches are explained in “The Brassicaceae Biofumigation System for Plant Cultivation and Defense. An Italian Twenty-Year Experience of Study and Application”. Acta Horticulture. 1005:375-382. http://www.actahort.org/books/1005/1005_44.htm. ISSN 0567-7572.

Q: (Michaela Schlathölter)

How do you apply the product?

Inside of the potato cultivation or before?

A: The approach consisted in three main agronomical “tool”:

- 1) Early harvest (in order to reduce tuber exposure to soil-borne pests and diseases).
- 2) Brassica derived biofumigant pellet, distributed on and incorporated to the soil in November, before the potato sowing in the next spring.
- 3) Brassica derived biofumigant liquid formulation for root application, applied through drip irrigation with standing crops, 4 treatments (on a 10/15-day basis, in the last months before harvest).

Q: (Mohamed Besri)

Has biofumigation using Camelina been commercially adopted by farmers?

If yes, how and for which crop?

A: Not yet, at least in the way we proposed it, but we are working to develop a product that

could be marketed soon. Anyway, even if we talk of plants or derived bioproducts, *Camelina sativa* is still an interesting minor oilseed crop that needs in depth studies in several applications.

Q: (Ngala Bruno)

Were the experiments with wireworms conducted in field or under controlled conditions?

A: The experiments were carried out in field conditions in big plots (at least 1000 m²), in farms where potato was already cultivated for years in the crop rotation.

Q: (Karima Bouchek-Mechiche)

What is the mechanism to reduce wireworms as well as Fungus *Rhizoctonia*, toxic?

A: From our point of view, we believe that, in addition to crop rotation, and all the agronomical solutions that could be adopted to indirectly reduce both these issues, the mechanism involves in both cases the ITC release, even if in this particular case, the early harvest had a crucial effect. In general, the aim, through different solutions that comprise for example bioactive green manure, is to improve agrobiodiversity and soil fertility.

04 From the lab to the field: what we have learned in twenty years

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Demonstrating the effects of biofumigant green manures in the lab and understanding the glucosinolate-myrosinase system on a molecular level is an important first step in moving this valuable technique to the field. Replicated plot trials confirm the statistical reductions of soilborne pests and measurable improvements to soil health. The only step left is to convince growers to incorporate this system into their complicated crop production practices, limited time schedules, high pest pressure crop rotations and tight financial conditions. In spite of the challenges, innovative growers around the world have been able to successfully adapt this green technique into their farming operations by developing cultural practices to produce large quantities of biomass, capturing the biofumigant potential of the volatile chemistry and the soil health benefits of the green manure, while producing high quality crops on schedule. We will review some of the techniques growers are currently using annually on thousands of hectares to consistently improve crop quality and soil health.

Link to presentation on Youtube: <https://youtu.be/0w7wsXJ9fUo>

Answers (A) from D. Gies to questions (Q) asked after the presentation.

Q: (Matthew Back)

Given the speed of hydrolysis. How important is water in the vacuoles versus the moisture of the soil? Open ended question!

A: Moisture in the soil is critical. We would prefer to have soil moisture at 80% Field Capacity or higher at incorporation and then irrigate or have a rain event after incorporation. In dry soils, the water in the vacuoles is not sufficient for complete hydrolysis as evidenced by crop damage in fields that were incorporated into dry soil, left dry for two to three weeks and then irrigated just prior to planting the next crop. We have seen damage in corn, wheat, small seeded vegetables and even potatoes when the soil was not hydrated properly.

Q: (Frédéric Le Dily)

Whatever the positive impacts of biofumigation, could you estimate the cost of 6-8 kg seeds/Ha and its further management (sowing, harvesting and burring)?

A: A difficult question to answer because of the huge variability of seed, fertilizer and equipment cost around the world. In the US, it would likely cost a grower USD\$300/ha for seed, fertilizer and incorporation. In some regions, the cost of irrigation could add to the total also. Fertilizer would be the most expensive input and those nutrients are returned to the soil at incorporation.

Q: (Mohamed Besri)

From the lab to the field: Excellent presentation. According to your experience, is biofumigation more used in organic production than in the conventional one?

A: It is used in both organic and conventional systems with good results. With only about 1% of the acreage in most countries being certified organic, I would say there are many more acres of conventional biofumigation than organic.

Q: Are you talking about disease suppression or disease reduction?

A: Replicated trials for various diseases in different parts of the world have shown results ranging from suppression to reduction to no effect depending on the cultivar used, biomass produced, incorporation methods and soil pH. Many growers observe equal or better results than synthetic soil fumigants are currently providing. There is always an improvement to soil health with the biofumigant green manure, not so much with the synthetic fumigants.

Q: (Annalisa Saccardo)

Which are the costs for the application of biofumigant green manures?

A: Seed, fertilizer and tillage costs vary widely around the world, as does the availability of rainfall and irrigation. You would need to calculate the costs based on local conditions.

Q: Have you registered more costs or not?

A: N/A

Q: (Amy Shi)

Have you tried using the combination of two cover crop to control a pest?

A: We have used combinations of species to reduce pest levels. Each species needs to be evaluated for what it can contribute to the process and provide overall improvement of yield and quality in the following crop. Many brassica varieties have been shown to elevate nematodes, disease or wireworms when used as a green manure, so just adding more cultivars to the mix is not always a good thing. If the object is to produce a biofumigant green manure, then only high glucosinolate varieties that have proven effective in the field should be used. We have successfully used a high glucosinolate arugula and mustard mixture to enhance wireworm and nematode control.

Q: Is certain variety targeting on certain pathogens?

A: We have found that ISCI 99 and ISCI TOP that produce high levels of volatile glucosinolates are very effective broad-spectrum biocides. Pathogens can vary in their sensitivity to these compounds, so the response will vary with the pest and the application of the biofumigation technique.

Q: (Luis Fernandez)

Did you have experience co planting indifferent seasons?

A: Hopefully, I understand the question correctly. Most growers need to produce a cash crop annually from each field. One of the main reasons we started working with brassicas 20+ years ago, was that they were quick growing and had good frost tolerance. Most growers plant them in late summer after the primary crop is harvested or in early spring before later planted summer crops.

Q: (John Duff)

Dale, what time if year do you grow your biofumigant crop?

A: Most of our green manure is planted in late summer, after grain harvest.

Q: (Mariel Mitidieri)

Can you tell how many hectares are using these biofumigants in USA or other countries?

A: That would be difficult for me to guess, but it is very common in some areas on thousands of hectares annually.

Q: (Alessandro Infantino)

Into which crop rotation do you insert biofumigation?

A: Ahead of any crop that has a disease or nematode issue. Some organic growers use it for the value of weed suppression whenever they can fit it into their rotation.

Q: (Ngala Bruno)

Very interesting PPT. I would love to know about your experiences with wireworms. Which species of wireworms have you observed susceptibility please?

A: In the Western US it has been very effective on the Dryland wireworm, *Ctenicera pruinina* and in Eastern Canada on several *Agriotes* species.

Q: Would this be due to tillage or a combination with the released ITC?

A: The potato fields receive considerable tillage to prepare for planting with minimal effect on wireworm populations here, but the AITC is very effective on this pest and has shown significant reductions in potato tuber damage.

Q: Have you or any of your associates conducted any experimental work with Biofumigation on wireworms please?

A: Dr. Lazzeri and the scientists at CREA have done some trial work. We have done thousands of hectares of commercial potatoes, where they are looking to reduce wireworms, nematodes and disease while increasing yields.

Q: (Annalisa Saccardo)

Which is the application for farmers of these plant selected?

A: Used as a biofumigant green manure for reducing pest pressure while improving soil health and crop yields.

Q: (Frédéric Le Dily)

What is the proportion of ISCI elite cv. seeds (ISCI99 for example) in commercial Caliente mixtures?

A: Over 90%.

Q: (Michaela Schlathölter)

Is Biofumigation growing in US?

A: Yes. Educating growers that green techniques applied properly can be effective and profitable in the tough economic conditions of commercial agriculture is a big part of the challenge to continued growth of this practice.

Q: (Setu Bazie Tagele)

If *Sclerotinia* stem rot affected mustard biofumigant, can we grow other crops like pepper after biofumigation?

A: We rarely see schlerotinia in the green manure fields, but it is an issue in the seed fields where the plants tend to lodge under irrigation when ripening. At or before full bloom, when the green manure crop should be incorporated, there normally is not much schlerotina present. If you have significant *Schlerotina* in the green manure crop, I would suggest to not follow immediately with another crop that is also susceptible.

Q: (Luis Fernandez)

Can we use sheep in pasture there?

A: Ruminants tend to prefer other foliage to the high glucosinolate containing leaves of these brassicas. If there is nothing else to graze on, they would likely attempt to eat them. It might take a while for their rumens to adjust.

05 Brassicaceae selection for Biofumigation purposes

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The CREA CI Brassicales seed collection, kept at the Cereal and Industrial Crops Centre of Bologna, represents the starting germoplasm material for the genetic selection carried out from early nineties for several purposes, first of all applications in soil-borne pests and diseases control through the biofumigation approach. This work led to the registration of Brown mustard (*Brassica juncea* L. Czern.) varieties ISCI 20 and ISCI 99 to the Italian Catalogue of Plant Varieties. As for these varieties, the new accessions are characterized for the glucosinolate profiles in all their tissues, and then they are reproduced in field conditions. Currently, a significant activity of Brassicaceae selection takes place at CREA CI with the objective of improving the already high performance of different crops (e.g. Brown mustard, *Eruca sativa* Mill., *Brassica carinata* A. Braun, et al.). Particularly, the aim of the research activity is i) to increase the biomass production for green manure applications, and on the other hand, ii) increase the bioactive molecules content, especially for what concern the most effective classes of GLS for industrial purposes. The last result of this activity was the USDA patent of "ISCI TOP", a high performance Brown mustard for biofumigation green manure purposes. At the same time, other Brassicaceae selections are under evaluation for agronomical/biofumigation (e.g. *Eruca sativa* Mill., *Brassica carinata* A. Braun, *Brassica nigra*), green chemistry and nutraceutical purposes (e.g. *Camelina sativa*, *Crambe abyssinica*, *Eruca sativa*).

Keywords: Allelopathic effect; glucosinolate content; plant selection, green manure

Link to presentation on Youtube: <https://youtu.be/6OWFR2AE0EE>

No questions were asked.

06 New tool and new application sectors for biofumigant cropping system

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Biofumigant plants and bioproducts are successfully applied from several years in plant management and defence all over the world both in organic and conventional farming.

In these last years the EU strongly restricted the authorization for chemicals in general and for Plant Protection Products (PPP) in particular, requesting the phase out for several fundamental pesticides widely applied in conventional farming. This decision determined for the farmers a sort of emergency for some crops historically cultivated in EU as potato, garlic, tomato, carrots fruit replant and other .

In this scenario the Biofumigation system can play a novel role for a total or partial alternative to chemicals helping the farmer to apply non chemical alternatives. In this way it will be possible a stronger contact with the aware consumers for a healthier food in a cleaner environment. This objective sounds as a prestigious tool and for this starting from previous experiences in soil fertility new and old synergies can be applied as the combination with useful microorganisms or of new allelopathic plants to offer a system alternative that will be reported and discussed in the Italian presentations.

Finally starting from the previous symposium held in South Africa it will be reported some new utilization of biofumigant system in post harvest defense carried as for bees, melliferous plants and as ingredients for healthy foods to define novel nutraceutical application that will be shortly described and discussed for a potential application in biofumigation in the short future.

Keywords: Green manure, bioproducts, cropping system

Link to presentation on Youtube: <https://youtu.be/hH7VdLUE3XE>

Answers (A) from L. Lazzeri to questions (Q) asked after the presentation.

Q: (Michaela Schlathölter)

Do you have to inoculate *Crotolaria*?

A: It is not necessary, even if some studies showed that inoculation can enhance alkaloids production, but we prefer to avoid extremely high amount of these particular kind of molecules, this mechanism should be adopted only if strongly necessary.

Q: (Neila AIT KACI AHMED)

I've got some questions about the liquid treatments (root and foliar):

Is it formulated from fresh macerated mustard?

A: No, it is not. They are formulated with defatted seed meals.

Q: How long these treatments can be conserved?

A: Since the products derive from dry materials (defatted seed meal and oil), they can be conserved as long as they can be kept in dry conditions.

Q: Are these products sterilized during the production, to avoid the development of contaminations from the mustard extracts?

A: No, they are not sterilized, even in this case because they are stocked in dry conditions, and only when they come in contact with water during the field applications, the reaction starts releasing the volatile molecules.

Q: (Julie Finnigan)

Luca, do you need to macerate the *Crotolaria* the same as the brassica's?

A: Even if the agronomical technique is similar, in terms of chopping and material incorporation, you do not need to macerate, there is no reaction such as GSLs-MYR hydrolysis, so it is not necessary to add water to activate it nor to quickly incorporate the chopped material into the soil, because there are no such volatile compounds viz. allyl ITC. It is quite a different approach.

Q: (Annalisa Saccardo)

Is it possible to apply biofumigation for extra-agricultural uses too?

A: The definition of several bioproducts permitted to apply the biofumigation system even in different contexts besides the agriculture. For example, some activities have been done for medical application, or for animal welfare, we have just published a paper on *Nosema ceranae* control introducing "biofumigant plants" in bee diet'. Another proposal regards the control of degradation of cultural heritage replacing chemical fumigants with biofumigation

and, with a "less classical" and wide ranging interpretation, several ones are surely undergoing.

Q: (Lidia Sukovata)

How do you apply the liquid product on roots?

A: Liquid products are applied in tanks for drip irrigation, in which the water-oil emulsion is produced, then the meal is added in a bag made of a tight meshed net, so the meal cannot freely float into the liquid portion in order to avoid the obstruction of the irrigation system. Once the reaction is at his higher release (at least 40-50 minutes), the irrigation system is activated releasing in the soil the biofumigant molecules.

Q: (Michaela Schlathölter)

Luca: is one of the biocidal plats your favorite or does it depend on the disease, which one to use?

A: There are several possibilities, there is not a one for all solution, it depends of course on pest or disease you would like to control, but also it depends on several aspects related to the pedo-climatic conditions, farmer equipment and crop rotation.

Q: (Brankica Tanovic)

Is there any influence of the soil pH on the effectiveness?

What is the application rate of the pellets?

A: As already mentioned, a lower pH causes lower conversion in isothiocyanate, but in our experience, at least in the southern Europe regions, this rarely concerns agricultural soils, and we have not observed a biofumigation inhibition caused by the soil pH. Pellets can be applied at a rate of 100-200 g/m², depending on the problems you need to address in the field.

Q: (Michaela Schlathölter)

Did you also did some analysis about the surrounding microbiom in the soil?

A: Yes, we did it within some experiments. For example, some indications can be found in this paper:

Mocali s., Landi, G. Curto, E. Dallavalle, A. Infantino, C. Colzi, G. d'Errico, P.F. Roversi, L. D'Avino, L. Lazzeri. 2015. Resilience of soil microbial and nematode communities after biofumigant treatment with defatted seed meals. *Industrial Crops and Products*, Volume 75, Part A, pagg 79-90. <http://dx.doi.org/10.1016/j.indcrop.2015.04.031>

Q: (Philippe LARROUDE)

Have you got some results about the effectiveness of *Crotalaria* on wireworms?

How was applied the *Crotalaria* on cover crop?

Before potatoes or others crops?

A: Yes, we have some and they are extremely encouraging, but unfortunately at the moment we have published them only partially and locally, in Italian language. The interest of *Crotalaria* is related to the control of several pathogens, N fixing and soil organic matter implementation, with the advantage of covering the soil during the summer time, thus reducing the organic matter loss when the temperature is very high. In some case studies, in northern Italy, we proposed *Crotalaria* in potato rotation, in which it was sowed in August and incorporated into the soil in October, before the next potato cultivation starting in spring.

07 Improving soil condition and yields using biofumigation across high value annual and perennial horticultural crops in Tasmania Australia

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Tasmania is the southern most Island state of Australia. It is geologically and topographically diverse, with equally diverse soils sustaining unique fauna and flora, and a growing agricultural history. Rich volcanic soils in the north have hosted horticultural cropping for many years, including pyrethrum, poppies, peas, potatoes, carrots and beans. The midlands and southern areas of the state, dominated with less fertile duplex and clay loam soils, were for many years predominantly grazed and dryland cropped. However, in the past 30 years the Tasmanian government's continued development of irrigation schemes to provide surety of water for agriculture, has seen a widespread increase in high value horticultural enterprises, both annual and perennial. With increased widespread intensive cropping, the need to manage our soils has become a major focus for practitioners, managers and consumers alike. Serve-Ag has been implementing biofumigation practices with our growers for more than 15 years, trialing many varieties, assessing glucosinolate levels, agronomic management practices and timing within our varied rotations. This investment has assisted production of many crops with increased yields, reductions in pathogen levels and minimising the impact of pests and weeds for many high value crops. Growers have seen increased soil carbon levels, can physically feel the improvements to soil resilience, and rely less on chemicals and synthetic fertilisers. The drive for organic production within perennial horticultural enterprises has also been a catalyst for biofumigation in replant situations, and the use of BioFence and BioFence FL to assist with suppression of Apple Replant Disease. Today we continue with demonstration trials, including varied establishment techniques, modification of equipment to improve the maceration and termination of crops, and targeting general cover crop selection to achieve specific outcomes. Intensive vegetable cropping provides many challenges where ground preparation, timing, crop sequences and disease risk require careful consideration to ensure the best outcomes for crop yield and soil health. The sustainability of our soils, both fertile and fragile under intensive cropping, is reliant on biocidal and non-biocidal cover crops well into the future.

Keywords: Soil condition, biofumigant crops, biofumigant products, disease suppression, weed suppression, pest suppression

Link to presentation on Youtube: <https://youtu.be/TSIX0vnaggY>

Answers (A) from J. Finnigan to questions (Q) asked after the presentation.

Q: (Mohamed Besri)

Australia continues using Methyl Bromide to produce certified strawberry runners. Could biofumigation be considered as alternative to MB to produce this propagating material in Australia?

A: Hi Mohamed. This is a good question and certainly worth exploring. Depending on the situation, Biofumigation could be used as a pre-plant treatment, either a crop or Biofence meal. The other option could be treatment with BioFence FL, the flowable option that could be applied through the irrigation system. Depending on site characteristics for growers, such as the level of pathogen or nematode pest that is trying to be suppressed, and soil type and texture etc, the rate of product application could also be played with. The thing to remember is that the process of biofumigation is not robust or as reliable as throwing on a chemical, but it does provide very worthy options for growers wishing to minimise chemical input. The best we can do is target our management options for best possible results, and follow through with monitoring to determine the level of effectiveness we can achieve. I believe Dale Gies has some strawberry data from the US that showed positive results for growers, so may be good to have a chat with him too.

Q: (Lidia Sukovata)

Julie, do you have any idea why the apple trees reacted in reduced height? Perhaps, they will get larger apples?

A: Hi Lidia. In short, I do not know why the BioFence treated trees had longer term reduced height compared to other treatments. Only 12 permanent monitoring trees were chosen per treatment, and these trees were initially selected for their leaders having equal height. This initial selection process may have introduced bias. Final measurements of all trees within each treatment showed very little difference in average height between the BioFence and the chemical fumigant, with MAP treated trees being tallest on average. Average trunk diameter was greatest in the chemical fumigant, followed by MAP, BioFence and control the smallest.

Apple harvest is now complete however, I do not have all the data at my fingertips just yet. From the first colour pick, the control and BioFence had a greatest yield, but final strip yield data is yet to be received.

08 Recent developments of the use of biofumigation for the control of soilborne diseases in Italy

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The sustainable use of soil to preserve its role as a platform for human activities is becoming a strategic issue for the development of any future action in the agricultural sector. Many agricultural practices, including the use of fertilizers, crop specialization, control of soilborne diseases, have negative impacts on soil health. In the same time, the use of biofumigation represents a sustainable strategy for soil management improving organic matter content in many cultivated agricultural soils. Recently, several developments were obtained in Italy by the use of this technique for the control of many soilborne pathogens, including nematodes. The effect of purified extracts of SIN, GER, GCH, and GST was tested against several phytopathogenic fungi and oomycetes and two *Trichoderma* spp. *in vitro*. The results obtained showed that both *Trichoderma* species were not inhibited by the concentrations of the four glucosinolates, while for the remaining species, a high variability of the response to the concentrations of the four glucosinolates in the test was registered. The potential of the use of dressing of *Brassica juncea* seeds with the biocontrol agent *Trichoderma gamsii* has been analyzed. Preliminary results showed a significative effect on plant length and the recovery of *T. gamsii* on the roots two months after sowing. To fill the gap concerning the environmental effects of biofumigation with defatted seed meals (DSM) against nematodes on non-target soil organisms, a 454-pyrosequencing approach was applied. Sinigrin DMS proved to control *M. incognita* and improved the amount of nutrients and organic compounds of soil. Significant shifts of both bacterial and fungal communities were observed after different treatments. The results showed how the overall microbial diversity did not significantly decrease after DSM treatments, suggesting a pronounced resilience of soil biological communities after biofumigation, and confirming the interesting potential of biofumigant DSM amendments as alternative to chemical fumigants for a more friendly control of some soil-borne diseases.

Keywords: Soil health, plant diseases, fungi, oomycetes, Bbiocontrol agent

Link to presentation on Youtube: <https://youtu.be/Aaiv9iYBBjs>

Answers (A) from A. Infantino to questions (Q) asked after the presentation.

Q: (Mohamed Besri)

Is biofumigation used at commercial level by farmers?

A: Yes, I have no direct data, but I know it from the Italian Company.

Q: If yes, on which crop?

A: Fruit trees, strawberries, potatoes, horticultural crops.

Q: Is it used alone or in combination with other control methods in an IPM program?

A: Our suggestion to farmers is to utilize biofumigation within IPM strategies (resistant varieties, crop rotations, use of other organic amendments, etc.).

Q: (Dale Gies)

Is *Trichoderma gamsii* compatible with *T. hazianum*?

A: Yes, I suppose. In one of the most successful product, *T. gamsii* is co-formulated with *T. asperellum*.

Q: (Brankica Tanovic)

Did you studied antagonistic properties of your *Trichoderma*?

A: Yes, I have chosen the two isolates among several other *Trichoderma* spp., based on dual culture, soluble and volatile component assays.

Q: (Mariel Mitidieri)

Is it possible to relate microbiome studies with soil borne pathogen population causing economic damages?

A: Yes, to correlate the microbiome changes with the suppression of a specific pathogens is the main goal, but it is also worth to utilize more efficient diagnostic tools (RealTime qPCR) to follow the fate of the pathogen into the soil after biofumigation.

Q: (Karima Bouchek-Mechiche)

Is their difference of efficacy of brassicacea biofumigation on different inoculum densities and soil types?

A: At present, I have not experimental data on it. Nevertheless, I am confident that the study of soil microbiome using more performant molecular tools (NGS long reads with PacBio or Nanopore sequencing) will give us more precise answers to this question.

09 Isothiocyanates associated with Brassica species impedes the survival and foraging activity of the stem nematodes *Ditylenchus gigas* and *D. dipsaci*

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Isothiocyanates (ITCs) are produced from the enzymatic degradation of sulfur-containing glucosinolates (GLS) by β -thioglucosidases (myrosinase). Brassica plants that contain glucosinolates and myrosinase are used in cropping systems as biofumigants as they produce ITCs that are toxic to a range of soil borne pests and pathogens. A concentration of 0.005% 2-propenol ITC is obtainable under field conditions through biofumigation with Indian mustard (*Brassica juncea*) (Wood et al., 2017). However, the activity of ITC's against the stem nematode (*Ditylenchus gigas* and *D. dipsaci*), important pests of field beans (*Vicia faba*) is unknown. The aim of this study therefore was to evaluate the biocidal activity of four synthetic ITCs, sulforaphane, 2-propenyl, 2-phenylethyl and benzyl against *D. gigas* and *D. dipsaci* and to determine their efficacy and LD50 at concentrations of 0.01, 0.005, 0.0025, 0.00125% (w/v) after 24, 48 and 72 h of exposure under in-vitro conditions. In addition, the host finding ability of the nematodes was examined following ITC exposure. With the exception of sulforaphane the ITCs suppressed the nematodes survival to less than 10% at 0.01 % (w/v) after 24 h ($P < 0.001$) and the survival increased with lower ITC concentrations of 0.0025, 0.00125% (w/v). Moreover, these surviving nematodes were unable to locate their host under in vitro conditions. These results suggest that Brassica plants containing 2-phenylethyl, benzyl, and 2-propenyl glucosinolates could have suppressive effects against populations of both *D. dipsaci* and *D. gigas* in soil.

Reference(s)

Wood, C., Kenyon, D.M. and Cooper, J.M., 2017. Allyl isothiocyanate shows promise as a naturally produced suppressant of the potato cyst nematode, *Globodera pallida*, in biofumigation systems. *Nematology*, 19(4), pp.389-402.

Keywords: Biofumigation, bioassay, glucosinolate

Link to presentation on Youtube: *This presentation might be published later*

Answers (A) from N. Musa to questions (Q) asked after the presentation.

Q: (Ngala Bruno)

From your experiences, what would you recommend as the best way to apply ITC's to manage *Ditylenchus* spp.?

A: Growing and incorporating brassica plants particularly *Brassica juncea* is one method that works reliably by releasing ITC which act against plant parasitic nematodes and *Ditylenchus* spp. inclusive as tested under field conditions.

Q: Have you had some field or soil based data please?

A: Yes, field studies were carried out to evaluate the performance of brassicas against *Ditylenchus* spp. We found some suppression of *D. gigas* but not *D. dipsaci*.

Q: (Alessandro Infantino)

As a general question, are present both biocidal and repellent actions for nematodes?

A: Yes, we tested and found, under in-vitro conditions, that there were biocidal effects against nematodes and at the sublethal concentrations nematodes were failing to respond to host roots. No study was conducted to determine repellent action, although this would be interesting.

Q: (Lidia Sukovata)

Do you think that sublethal dose of ITC unables the nematodes to find the ""smell"" of the host plant roots?

A: Yes, there is strong evidence that the nematodes under sub lethal doses of ITC have lost their ability to sense their host diffusates and respond to them. Nematodes rely on their amphids to sense and respond to the chemical cues in their environments. They do so by orienting themselves to their host roots. We hypothesize that these nematodes lost their sensory ability when exposed to sub-lethal doses of ITC.

Q: (Luis Fernandez)

Did you check saturated concentration about reversible action?

A: The reversible actions, in other terms recovery, we reported were mostly seen in lower concentrations/ doses of the ITC tested. Under higher concentrations/doses the nematodes recovered poorly. However, the nematodes recovering under the lower concentrations were unable to respond to their host diffusates.

Q: (Frédéric Le Dily)

From my own experience, *S. alba* seems less efficient as regard to *B. juncea*. Is there a simple

dif in ITC?

A: Yes, most analysis on the types of glucosinolates *S. alba* and *B. juncea* produce show differences. *S. alba* has sinabin while *B. juncea* sinigrin. The two glucosinolates decompose to form other chemical products. The former glucosinolate produce is unstable products which has no nematotoxic effect while the latter produce allyl isothiocyanates that has strong effect against nematodes.

Q: (Lidia Sukovata)

You showed that phenylethyl ITC caused higher mortality of nematodes, but allyl ITC cause higher arrestment?
Which effect is more important?

A: Phenylethyl ITC had stronger effect on nematodes survival than allyl ITC. This is true under high concentrations of ITCs (eg 100 mg/L). However, such high concentrations of ITCs are less obtainable under field conditions. However, allyl ITC had more effect in inhibiting the ability of nematodes to respond to their host at lower ITC concentrations (eg 25 mg/L). Under most biofumigation systems it is possible to obtain low ITC concentrations than a higher one. Hence, we recommend that focus show be on achieving high ITC concentrations as possible but also maximizing the inhibitory effects of ITC at low concentrations.

Q: (Ernesto Martin Uliarte)

Did you find some effect of biofumigation in nematophages, they are also affected like pathogen nematodes?

A: It would have been interesting to find the effect of biofumigation on any other beneficial bacteria/fungi/nematodes that act as nematophages in this study. Unfortunately, we were limited with time and resources to look at that. Some previous work has found biofumigation having negligible effect against beneficial bacteria. Ref: Taylor, 2013
<http://theses.gla.ac.uk/4854/>

10 On-farm assessment of biofumigation and reduced tillage for soil-borne disease mitigation and soil health improvement in New York State, USA

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Studies illustrating successful biofumigation with Brassicaceae cover crops by United States growers have been variable, and it is unknown if circumstantial factors and/or mismanagement may be negatively interacting with biofumigation potential in the field. Other beneficial non-biofumigation effects of these cover crops in soil health programs are also not well understood by US growers. We conducted a two-year field study in New York State, USA investigating soil-borne disease mitigation and soil health management potential of biofumigation used as a component of a 2-year soil health management program. Four Cucurbitaceae growers with infestations of the soil-borne pathogen *Phytophthora capsici* established field-scale on-farm control plots adjacent to plots where *Brassica juncea* cover crops were used for biofumigation in year 1, and reduced tillage (RT) with rye (*Secale cereale*) cover crops were used in year 2. A research station-based plot-scale study with corresponding treatments was conducted simultaneously. Cucurbitaceae crop yield response and cover crop biomass C and N was measured each year, and soil health was assessed was after year 2. On-farm evidence of treatment effects on Cucurbitaceae crop yield and soil health lacked ($p > 0.10$), with *P. capsici* blight incidence absent or very low in all treatments, farm sites, and years. Treatment differences in year 2's plot-scale trials were evident but resulted from other dynamic non-biofumigation interactions- blight was absent. Mean cover crop N and C returned to soils following biofumigation and rolled rye cover crops in RT treatments was ~3.5-5x greater than grower standard controls. These returns to soils may be noteworthy due to a positive association found between soil infiltration rates and inherent soil organic matter levels ($r^2 = 0.87$, $p < 0.01$) in the soil health data. Rye cover crop biomass may also be important in the RT component, as blight was only found in plots with $< 3000 \text{ kg ha}^{-1}$.

Keywords: On-farm, Extension, reduced tillage, short-term, biofumigation, Mustard, *Brassica juncea*, Rye, *Secale cereale*, United States, integrated, soil health

Link to presentation on Youtube: <https://youtu.be/gDGTCL33kxw>

Answers (A) from J. O’Dea to questions (Q) asked after the presentation.

Q: (Annalisa Saccardo)

How many American farmers adopt biofumigation in percentage more or less?
And how do you disseminate this sustainable practice?

A: Biofumigation practices in the US have never been quantified as far as I know, but I speculate that it is not very widely practiced (>5%?). In the US, biofumigation practices are primarily disseminated via 1) grower field days, workshops, Extension publications etc., 2) seed companies that sell biofumigation-specific varieties, and 3) farmer peer-mentors within certain farming communities where the practice has gradually become popularized.

Q: (Alessandro Infantino)

Which is the situation of chemicals reduction or banning in the USA?

A: I don’t have extensive knowledge of the situation of banning or reducing certain agricultural chemicals in the US as they relate to biofumigation, but metam sodium (the synthetic fumigant most directly related to biofumigation) is still approved for use. In general it is not uncommon for government initiatives to support efforts to find less hazardous alternatives to soil fumigants, such as the USDA’s “Methyl Bromide Transition Program”, but I have not heard of a program like this that is specific to metam sodium.

Q: (Annalisa Saccardo)

May you explain better the matter about equipment access and investment costs?

A: Both biofumigation and reduced tillage are rather equipment-intensive. Biofumigation requires 1) a flail mower (or large rotary mower), 2) a field cultivator, rotavator, or disk, 3) a roller/packer/cultipacker of some type, and 4) enough tractors to run that equipment over the field in rapid succession (if not simultaneously). Reduced tillage requires specialized equipment for seeding or performing strip tillage, and for growers not using herbicides a roller-crimper is required to roll-kill a rye cover crop into a mulch. Reduced tillage equipment was especially rare for our growers and required borrowing from neighbors or government rental or private equipment demonstration services. Borrowing equipment meant an extra step in complexity for time economy and timing the field operations. Purchasing equipment meant a relatively high commitment to invest in reduced tillage practices on the farm for the long-run.

Q: (Frédéric Le Dily)

What about NO₃ and SO₄ releasing after intercropping?

It may depend of the N and S (organic/mineral?) contents of the biofumigant crop ?

Do you have a point of view?

A: We did not measure actual N or S release in soils in our study. Anecdotally, there appears to be strong nutrient returns to subsequent crops though. This is unsurprising because N

levels in residues from biofumigation cover crops typically high (averaging 147 kg/ha in our study), the average C:N ratio of biofumigation cover crops at termination in our study was approximately 13:1, and cover crops are additionally flail-chopped before incorporation into soils. Each of these factors would theoretically help facilitate strong net nutrient mineralization from residues in soils after incorporation, and at levels that would substantially contribute to what is available to the succeeding crop.

Q: (Minori Arai)

What do you think about no-till?

A: No-till has many advantages, but it is also a rather advanced cropping strategy and requires appropriate expectations from growers regarding 1) the investment required to practice it effectively and 2) possible downsides (such as increases in perennial weed pressure), and 3) likely delays in return on the investment into the practice (such as lower nutrient availability during the transition period).

Q: (Setu Bazie Tagele)

Do we need fertilization for growing biofumigants?

A: Yes, typically we do. Brassica cover crops are non-mycorrhizal, and therefore rely heavily on the availability of soluble nutrients. They are exceptional at recovering soluble nutrients from soils when it is available, and notably susceptible to failure if soluble nutrient levels in soils are low. Assuming all other insoluble nutrients are adequate, S fertility is typically added along with N to assure that the plant is not limited in producing adequate biomass and the sulfur component of isothiocyanates that are essential for effective biofumigation. Soluble nutrients are typically low in spring in particular, and unfertilized biofumigation cover crops are highly likely to fail as biofumigation cover crops without supplemental fertility. Fall-planted biofumigation cover crops may be more likely to succeed without supplemental fertility, but there is still a risk of failure without assuring adequate fertility. One exceptional observation we have made though is that on farms with robust soil-building programs (esp. including a history of considerable manure/compost additions), existing/inherent soluble nutrient levels may be exceptionally high and potentially sufficient to produce an effective biofumigation cover crop.

Q: (Neila AIT KACI AHMED)

On average how much initial soluble nutrient of S is needed for a sufficient synthesis of ITCs?

A: Typically, N and S are added to biofumigation cover crops in a 5:1 ratio, respectively, to assure adequate biomass production and the respective ITC concentrations needed for effective biofumigation. In the US (depending on inherent fertility levels), N is typically added at an average rate of ~113 kg/ha, along with ~23 kg/ha of S.

11 Horticultural crop health and yield and greenhouse soil conditions after 17 years of repeated treatments of biofumigation and solarization

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The combination of biofumigation and solarization is commonly known as biosolarization. A experience was performed from 2003 to 2019 in a greenhouse at INTA San Pedro, Buenos Aires province, Argentina (33°44'12.7"S 59°47'58.2"W). Treatments (TRAT) were applied every two years. TRAT evaluated were: 1=Control; 2= Solarization, 3= BIOROT, was a succession of organic amendments (chicken manure, broccoli, chicken manure, broccoli, tomato and pepper crop debris, mustard, tomato crop debris, broccoli), 4=BIOBRAS was based only on the use of brassicas (rapeseed, broccoli, broccoli, mustard, mustard, mustard, *Brassica campestris*, broccoli). The treatments were carried out in spring and during short periods in summer, so a late season tomato crop could be grown after them. The tomato hybrid used was Superman (Petoseed), except for the last season where the hybrid used was Rodeo (BHN). Fungal pathogens controlled were *Pyrenochaeta lycopersici*, *Fusarium solani*, *Sclerotium rolfsii* and *Sclerotinia sclerotiorum*, as well as nematodes like *Nacobbus aberrans*, *Helycotylenchus* and *Criconemella*. Fungus of *Aspergillus* genera were observed growing on death sclerotia of *Sclerotinia sclerotiorum* and *Sclerotium rolfsii* in BIOBRAS and BIOROT. Tomato plants in control showed higher percentage of death plants, root rots and lower root dry matter at the end of each crop. Solarization alone without adding organic matter reduced this parameter in the soil, and showed more death plants and less yield than BIOBRAS and BIOROT. Tomato and pepper crop debris used as biofumigants produced high yield values and adequate pathogen control. Beneficial organisms as *Trichoderma* and actinomycetes were isolated from rhizosphere of tomato growing on BIOBRAS and BIOROT soils. Good results were also obtained with other crops as lettuce, spinach, grafted tomato plants, broccoli, beets, chard, sweet potato for seedlings production, etc. Biofumigation in combination with solarization is an effective technique for managing soilborne pathogens in greenhouses and is being adopted by horticultural growers in Argentina.

Keywords: nematodes, soil borne pathogens, long term assay

Link to presentation on Youtube: https://youtu.be/sM436c_5JCM

Answers (A) from M. Mitidieri to questions (Q) asked after the presentation.

Q: (John Duff)

How long do you leave the plastic sheeting down for in the solarization process?

A: We have assayed 15 days in summer up to 30-42 in spring or summer. In order to control pathogens and weeds is better to apply long periods, but regarding that farmers need to use the soil it is more convenient 15 days in January or 30 days in November which allows to plant a late season tomato crop in Buenos Aires province, a region with mild winter weather.

Q: (Mohamed Besri)

According to your presentation, Biosolarisation is adopted by horticultural growers in Argentina to control soilborne pathogens in Tomato greenhouses including *Nacobbus aberrans*. Could you please explain why MB is still used as critical use exemption, in Mar del Plata and La Plata to control tomato soil borne pathogens and particularly the false root nematode?

A: We have demonstrated that Biosolarisation can be a solution for soil borne pathogens and false root nematode in tomato under protected cultivation. Some growers have adopted the technique, but others are more confident on chemical fumigants. As MB has become very expensive in our country, this situation enhances changes to alternative practices.

Puerta, A., Adlercreutz, E.; Delmazzo, P. Cuellas, M.; Amoia, P.; Borquez, A.M. y Mollineda, V. 2020. Alternativas sustentables para la desinfección del suelo y los sustratos y la eliminación del bromuro de metilo en los cultivos de frutilla, hortalizas bajo cubierta y ornamentales en la Argentina. Proyecto "TIERRA SANA" MP/ARG/00/033 INTA-ONUDI

Q: (Setu Bazie Tagele)

How could biofumigation would be possible without plastic cover?

A: If the infestation with pathogens is a limiting factor, it would be necessary a first treatment of biosolarisation to reduce nematode and pathogen populations. Then it would be possible to apply integrated pest management based on rotations with non host crops, grafted plants and biofumigation without plastic cover. IPM should be applied prior to reach high populations of pathogens so biofumigation could be effective.

Q: (Annalisa Saccardo)

Don't you use biodegradable film in agriculture?

A: We have not assayed this kind of film yet.

Q: (Lidia Sukovata)

Mariel, you use different plants to control different pathogens. Have you screened them

first?

A: We have not screened them. In the poster we have published “in vitro” assays of some colleagues regarding Fusarium control.

Q: (Setu Bazie Tagele)

For which crop-pathogen-system is biofumigation effective in Argentina?

A: *Nacobbus aberrans*, *Fusarium solani* and *Pyrenochaeta lycopersici* in tomato. *Sclerotinia sclerotiorum*, *Sclerotium rolfsii* that affect several horticultural crops in open field and in the greenhouse; also weeds like *Urtica dioica* that make difficult harvest of leafy vegetables.

Q: (Mohamed Besri)

Can Biofumigation control *Nacobbus*?

A: I have not assayed biofumigation without solarization.

Q: (Brankica Tanovic)

How to solve a problem with effectiveness in soil with low pH? Is it related only to Brassicas or with sudan grass as well?

A: In our region we have high pH values because of the water composition that has high sodium contents. I think pH values is a problem only with Brassicas because of the myrosinase reaction.

12 Biofumigant cover crops—a promising strategy for soil and disease management in southeast Queensland

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The Queensland Department of Agriculture and Fisheries evaluated fourteen (14) commercially available brassica biofumigant cover crops over a 12 month period, for performance characteristics and efficacy against known soilborne pathogens. These varieties were across 4 genera and 6 species types including *Brassica juncea*, *B. carinata*, *B. napus*, *B. nigra*, *Eruca sativa*, *Raphanus sativus*, *Sinapis alba*. The performance characteristics assessed included days to incorporation (25% flowering), biomass, glucosinolate concentrations and efficacy against known soilborne pathogens. The three soilborne pathogens used in efficacy assessments were *Sclerotium rolfsii*, *Sclerotinia sclerotiorum* and *Macrophomina phaseolina*.

The key findings from this work were:

- Brassica biofumigant cover crops can be grown during summer months in south-east Queensland outside of their typical winter growth window.
- Biofumigants have much faster time (from 36 days) to incorporation during summer months.
- Glucosinolate production varied considerably between varieties and with different growing seasons. Biofumigants generally produced higher glucosinolate levels and exhibited increased efficacy against known soilborne pathogens when grown during summer months. For example, BQ mulch (*B. carinata*/*B. nigra*) produced 20mole/ha in winter and 133mole/ha in summer, while Tillage Radish (*R. sativus*) produced 19.2mole/ha in winter and 114mole/ha in spring. *Brassica juncea* types did however produce more glucosinolates in winter with 320mole/ha compared to 260mole/ha in summer for Caliente. Each type appears to be different in their response to growing conditions.
- Field efficacy against known soilborne pathogens was also variable (7-100% mortality) with variety and growing season with higher levels of pathogen mortality typically observed following summer growing seasons.
- Fallow treatments also exhibited high levels of mortality with *S. sclerotiorum* was not managed to any great degree by any of the biofumigants tests compared to a fallow treatment (87% mortality).

While cover cropping is primarily a means to manage soil erosion during high risk summer storm periods and build organic matter, the potential of biofumigants to contribute to soilborne disease management makes them an attractive summer cover crop option for vegetable producers in southeast Queensland.

Keywords: biofumigant, cover crop, glucosinolate, *Sclerotium*, *Sclerotinia*

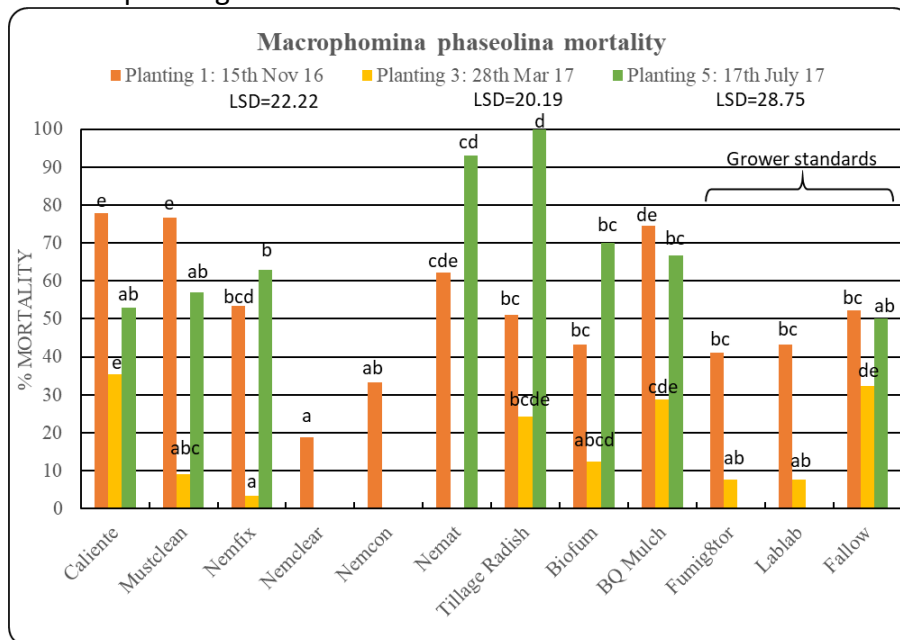
Link to presentation on Youtube: <https://youtu.be/hlk19PnRjSw>

Answers (A) from J. Duff to questions (Q) asked after the presentation.

Q: (Mohamed Besri)

As you know, *Macrophomina phaseolina* and *Fusarium oxysporum* f.sp. *fragariae* are significant soil-borne pathogens of strawberry crops . Could biofumigation control these 2 soil borne pathogens?

A: I don't know much about the Fof but from what I have read, Fusariums can be a little harder to control with biofumigation. The *Macrophomina* we have been able to achieve very high levels of control using *Eruca sativa* (Nemat) and an oil seed radish *Raphanus sativus* (Tillage radish, see below graph. There was very little difference between biofumigants with a summer planting, although Caliente and Mustclean (both *Brassica juncea*) and BQ Mulch (*B. nigra* and *B. napus*) were the better performers during the summer plantings window.



Q: If yes, what will be the cultural practices calendar: biofumigant choice, biofumigant sowing and incorporation times , strawberry planting time?

A: Check out the varieties above for the better performers and the times of year that they were planted as some varieties worked better in winter while others were better during a summer planting window. So long as there is water to get the crop established and there is plenty of subsoil moisture, then these crops can be grown anytime of the year. Summer is harder on the crops due to our high temperatures and so subsequent irrigations could be required if no rainfall occurs. Summer plantings did perform better with some biofumigants compared to others. Again check out the graph above. Summer plantings mature a lot faster than the cooler months. See table below. So depending on when you plant your strawberries you can pick a biofumigant that could be grown and incorporated well before you need to plant your strawberry runners.

	NOV	DEC	JAN	FEB	MAR	APR	MAY	JUN	JUL	AUG	SEP	OCT	NOV	DEC
BQ Mulch	36 DAYS			44 DAYS		44 DAYS		58 DAYS		59 DAYS		44 DAYS		
Nemfix	36 DAYS			44 DAYS		63 DAYS		90 DAYS		81 DAYS		61 DAYS		
Caliente	44 DAYS			50 DAYS		97 DAYS		98 DAYS		81 DAYS		72 DAYS		
Mustclean	44 DAYS			44 DAYS		63 DAYS		90 DAYS		81 DAYS		61 DAYS		
Biofum	70 DAYS			87 DAYS		97 DAYS		98 DAYS		89 DAYS		81 DAYS		
Tillage Radish	70 DAYS			94 DAYS		97 DAYS		98 DAYS		67 DAYS		81 DAYS		
Nemat	70 DAYS			87 DAYS		69 DAYS		79 DAYS		67 DAYS		78 DAYS		
Lablab	85 DAYS			102 DAYS		83 DAYS						88 DAYS		
Fumig8tor/ Scavanger	101 DAYS (3 CUTS)			102 DAYS (3 CUTS)		83 DAYS (1 CUT)				96 DAYS (1 CUT)		88 DAYS (2 CUTS)		
Nemclear	101 DAYS			102 DAYS		115 DAYS								
Nemcon	101 DAYS			102 DAYS		115 DAYS								

(Be aware that these data are from the Southern Hemisphere!)

Q: Could a matrix of biofumigation to control soil borne pathogens be developed for specific crops e.g.strawberries?

A: I can't see why not so long as the work is done in a strawberry growing region so that the effect on the biofumigants would be the same as the strawberries grown there. There is more and more biofumigants becoming available so developing a matrix would certainly keep you working hard to expand on the matrix.

13 ***Origanum vulgare* vapour primes defence mechanisms in grapevine (*Vitis vinifera*) and hinders *Plasmopara viticola* infection**

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The amount of synthetic pesticides applied in viticulture is relatively high compared to other agricultural crops, due to the high sensitivity of the grapevine (*Vitis vinifera* L.) to fungal diseases such as downy mildew (*Plasmopara viticola*). Alternatives to reduce fungicides are utterly needed to ensure a sustainable vineyard-ecosystems and consumer acceptance.

Essential oils (EOs) are amongst the most promising natural plant protection products due to their antibacterial, antiviral and antifungal properties. However, the efficiency of EOs depends highly on timing and method of application and the molecular interactions of host, pathogen and EO, which underlie the efficiency of EOs, are not well understood. To circumvent the drawbacks of a direct application, the presented study aimed a) to evaluate whether a continuous fumigation of EO can control downy mildew and b) to decipher molecular mechanisms that are triggered in host and pathogen by EO application.

Therefore, we customized a climatic chamber, which permitted a continuous fumigation of potted vines with different EOs. Several experiments with vines, infected with *Plasmopara viticola* and subsequently exposed to continuous fumigation of different EOs with different concentrations and application times were conducted. Experiments were stopped when signs of infections were clearly visible on the control after sporulation was induced. Strikingly oregano oil vapor treatment reduced downy mildew development to 95%. RNA. Analysis of differentially expressed genes yielded in a total of 4800 EO modulated transcripts in vines. Strikingly many genes linked to the plant immune system were triggered by EO vapour (ethylene synthesis, phenylpropanoids and flavonoid synthesis), which indicates for the first time, that the antifungal efficiency of EO is mainly due to the priming of resistance pathways inside the host plants. These results are of major importance for the production and research on biopesticides, plant stimulation products as well as for resistance breeding strategies.

Keywords: Plant defense; essential oil; *Plasmopara viticola*; Grapevine

Link to presentation on Youtube: <https://youtu.be/GCl2ShCcJBY>

Answers (A) from M. Rienth to questions (Q) asked after the presentation.

Q: (Mohamed Besri)

Plasmopara viticola is an airborne pathogen. Your experiments have been conducted in a climatic chamber, which permitted a continuous fumigation of potted vines with different EOs. Is this control method technically and economically feasible in the field?

If yes, could you please explain how the fumigation of vines could be conducted in farmers' conditions?

A: As discussed during the conference, it is still difficult, we are trying different protocols but haven't found the best yet.. One of the best solutions seems to be an encapsulation, this is what we are testing at the moment.

Q: (Lidia Sukovata)

Have you tried to encapsulate ITCs?

A: We are trying different methods currently.

Q: (Luca Lazzeri)

Which is the price for a biological encapsulates of 1 kg?

A: Sorry I have no idea, yet. Cause we do not have a finished product yet.

14 Biofumigation as a tool for the holistic integrated management of wireworm populations

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Many factors interact continuously in agricultural ecosystems, including a number that are not under a farmer's control. Therefore, Integrated Pest Management (IPM) may be reliably and sustainably implemented only by using specific holistic models that predict the outputs of the complex interaction between numerous abiotic and biotic factors. These models include timing the formation of harmful pest stages and its overlapping with periods of crop susceptibility. When holistic models are being considered for use, the factors that interfere with the development of *Agriotes* populations are: 1) the implementation of agronomic tactics, e.g. altering rotation, choosing tolerant varieties, timing tillage, and irrigation in accordance with the life cycle of each *Agriotes* species; and 2) the application of biological tools (e.g. biocidal plants, entomopathogens). Long-term research carried out in Italy has shown that "cultivation packages" that include wireworm risk assessment, suitable cover crops, defatted seed meals, plus choice of varieties and harvesting as early as possible, may produce high-quality potatoes with low or negligible soil pest damage without using any chemical insecticide. The results of applying specific packages that include biofumigant materials to fields with mainly *Agriotes sordidus* Illiger wireworm population will be presented.

Keywords: *Agriotes* spp., Integrated Pest Management, holistic models

Link to presentation on Youtube: <https://youtu.be/NXUfDQpi3qo>

Answers (A) from L. Furlan to questions (Q) asked after the presentation.

Q: (Lidia Sukovata)

How high are the costs of application of pellets?

A: It depends on: i) pellet price, ii) pellet quantity and quality in terms of glucosinolate content, iii) shipment costs; based on our trials an effective pellet application against wireworms requires 1000 to 2000 kg/ha (quantity mainly depends on the glucosinolate content of defatted seed meals and soil incorporation depth); please see Furlan et al. 2010 for conditions needed to make pellets work. Based on this, pellet treatment costs 1,500.00 to 3,000.00 €/ha at current pellet prices. This means that biofumigation carried out with a good pellet has a cost comparable to that of chemical fumigation (1,3-Dichloropropene). You should also consider the reduction in the cost of slow-release nitrogen due to the 6% of organic N content in the pellets.

Q: Can potato growers afford using them?

A: It depends on the product price (e.g. organic high-quality potatoes might compensate cost better than other production types). The incorporation of biocidal plants is much more affordable (seed + cultural interventions are worth about € 200-300). See Furlan et al. 2009.

Q: Did you water the field after using pellets?

A: Not when soil moisture was near to water capacity; in most trials, there was no irrigation; please see Furlan et al. 2010).

Q: (Federica Mosso)

How much meal is recommended per square meter?

A: It depends on glucosinolates/(and myrosinases) concentrations; as stated above, application of an effective meal/meal pellet against wireworms requires 1000 to 2000 kg/ha; (quantity mainly depends on the glucosinolate content of defatted seed meals and soil incorporation depth); please see Furlan et al. 2010 for conditions needed to make pellets work. Based on this, recommended dosage ranges from 0.1 to 0.2 kg per square meter.

Q: (Matthew Back)

Have you or anyone investigated a mixture of biofumigant and trap crops for wireworm?

A: No, sorry

Q: (Ngala Bruno)

I've read some of your articles on the lifecycle of wireworms. Have you had some experiences rearing larvae under controlled conditions?

A: Our rearing activity is mainly based on rearing cages as described in Furlan 2004. Part of the larvae are produced under controlled conditions following a method very similar to that described by Kölliker et al. 2009.

Q: (Philippe LARROUDE)

Have you got some results about the reduction of populations of wireworms with *Crotalaria* compared to *Brassica carinata* or *juncea*?

A: We are running research on this issue in controlled conditions and open fields. Results have not been published yet. We are observing significant potato protection using *Crotalaria juncea* (variety Madras) although wireworm mortality caused by the soil-incorporated *Crotalaria* plants is negligible, at least in the short term. We are trying to understand the mechanisms behind this.

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15 Use of biofumigant seeds meal in liquid formulation to improve the soil fertility and to limit the symptoms of mortality in kiwifruit plants

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Italy has the largest area of specialized kiwifruit in Europe. 10% of the national area is located in Veneto. However in the last years, this crop, traditionally with good profitability, recorded a decrease of 8%. From a survey conducted in 15 large farms in Veneto some issues have been identified that can make less attractive the crop.

An over exploitation of the soil is affecting the yields of plants. The symptoms are visible in the upper part of the plant that shows a reduced foliage development, reduced vigour and a consequent lack of production up to a complete summer defoliation and the early mortality of the plant.

Assuming a correlation between the level of soil fertility and the dead of plants, studies have been conducted in three representative farms of Verona area. In two years, materials based on biofumigant formulated DSM were applied to the soil on adult plants of kiwifruit. After a preliminary evaluation,

The applications were carried out in summer with a patent solid/liquid formulation. Different timing and dosage has been evaluated. For each thesis general evaluations were made on the phytotoxicity of the treatments and on the health of the plant as well as on the quantity and quality of the productions. Always the comparison has been made with an untreated test. Given the positive results obtained, further studies were carried out to identify simple tools to mechanize the treatments. . At the end of the test an assessment of benefits was also conducted, involving the producers.

Keywords: kiwifruit, early plants mortality, soil fertility

Link to presentation on Youtube: <https://youtu.be/aGJsEwL-23g>

Answers (A) from F. Mosso to questions (Q) asked after the presentation.

Q: (Lidia Sukovata)

If I understood well, you applied the formulation to the soil/on roots. Were there any signs of phytotoxicity?

A: Phytotoxicity effects can only occur if the flour is not well buried and wetted before sowing or transplanting. The important thing is to wait at least 7 days before sowing or transplanting the new crop.

Q: (Frédéric Le Dily)

Nice demonstration ! Question : Is the use of biofumigant seeds meal in liquid formulations subjected to accreditation?

A: I didn't quite understand the question, if by accreditation you mean recognition by institutions with state subsidies, this depends on how the different products are considered, are fair and green manure preferable.

Q: (Niels Agerbirk)

You started out saying that the cause of the kiwi-disease is not completely understood. Now biofumigation had an effect, can you come closer to a cause of the kiwi-disease?

A: Unfortunately not, there is a lot of research going on, not least because analysing the roots reveals a variety of pathogens, but what they trigger with is still unclear.

Q: (Luca Riccioni)

Did you also improve the drainage of the soil, other than make the treatments?

A: I didn't do anything about drainage but I worked on gravelly soils where water stagnation is really impossible and the problem was there anyway.

Q: (Lidia Sukovata)

Have you estimated when (at what concentration level) a phytotoxicity can appear?

A: Yes, up to 4 times the recommended dosage, nothing happens, only an increase in costs!

Q: (Julie Finnigan)

Is this management approach cost effective for your growers?
Return on Investment?

A: Yes, the treatment allows you to bring the crop into production with excellent fruit quality at a very balanced cost, obviously the crop must not be compromised even before treatment otherwise it will not recover.

16 Use of rotations, cover crops, and green manures for disease suppression in potato cropping systems

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Crop rotations and inclusion of cover crops and green manures are primary tools in the sustainable management of soilborne diseases in crop production systems. Crop rotations can reduce soilborne disease through three general mechanisms: (1) serving as a break in host-pathogen cycle; (2) by altering the soil physical, chemical, or biological characteristics to stimulate microbial activity and diversity; or (3) directly inhibiting pathogens through the release of suppressive or toxic compounds or the enhancement of specific antagonists. Brassicas, sudangrass, and related plant types are disease-suppressive crops well-known for their biofumigation potential, but also have other effects on soil microbiology that are important in disease suppression. Efficacy of rotations for reducing soilborne diseases is dependent on several factors, including crop type, rotation length, rotation sequence, and use of the crop (as full-season rotation, cover crop, or green manure). Years of field research with Brassica and non-Brassica rotation crops in potato cropping systems in Maine have documented the efficacy of Brassica green manures for the reduction of multiple soilborne diseases, but have also indicated that these crops can provide disease control even when not incorporated as green manures, and that other non-biofumigant crops (such as barley, ryegrass, and buckwheat) can be just as effective as Brassicas under the right conditions. In general, all crops provided better disease control when used as a green manure vs. as a cover crop, but the addition of a cover crop can improve control provided by most rotation crops. In long-term cropping system trials, rotations incorporating multiple soil health management practices, such as longer rotations, disease-suppressive rotation crops, cover crops and green manures, and/or organic amendments, have resulted in greater yield and microbial activity, and less disease problems than standard rotations. These results indicate that improved cropping systems may enhance productivity, sustainability, and economic viability.

Keywords: Crop rotation, cover crops, green manures, soilborne disease, potato cropping system

Link to presentation on Youtube: <https://youtu.be/orhS27pPNg>

Answers (A) from R. Larkin to questions (Q) asked after the presentation.

Q: (Annalisa Saccardo)

How much is widespread the practice of rotations among US farmers and for which crops do they apply it more often? Rotations are used in conventional agriculture such as in organic agriculture?

A: Crop rotation, in general, is very widespread in the U.S. I think it is well-established and recognized by growers regarding the importance of at least some form of crop rotation and is used consistently by both conventional and organic growers for most crop production systems. There may be some grain and forage crop producers that grow multiple years of the same crop (such as wheat or alfalfa, etc.), but still generally employ some form of crop rotation every few years. Overall, conventional growers tend to have shorter rotations than organic growers, due to greater focus on a specific high-value cash crop, whereas organic growers tend to be more diversified with longer multi-year rotations and greater concern for building soil quality. However, consistent use of cover crops has not yet been widely adopted in most conventional crop production systems.

Q: (Michaela Schlathölter)

Which type of buckwheat do you use: *Fagopyrum esculentum* or *tataricum*?

A: The buckwheat I am familiar with are all in the species *F. esculentum*. I am not aware of the other species being used as a cover crop to any degree in the U.S.

Q: (Mohamed Besri)

Your results have shown that Brassica and non-Brassica plants can provide disease control even when not incorporated as green manures and that other non-biofumigant crops (such as barley, ryegrass, and buckwheat) can be just as effective as Brassicas under the right conditions. What are these right conditions?

A: Generally, the conditions where the non-biofumigant crops are as effective as biofumigants are when conditions are not that favorable or optimal for biofumigation. That is, when there are issues such as insufficient moisture or inadequate biomass production, or crops not incorporated quickly, or other conditions that do not allow optimal biofumigation effects, other types of disease-suppressive crops may function as well or better. And in actual practice, this happens quite often. Thus, sometimes other non-biofumigant crops can produce comparable results.

Q: If non-biofumigant crops are as efficient as non biofumigant ones, how we can choose between these plants to control soilborne pathogens of a specific cash crop?

A: First, the non-biofumigant crops generally are not 'as efficient' as biofumigant crops. The thing that makes the biofumigant crop more effective is that, in general, it has multiple mechanisms of action. It functions not only through the chemical process of biofumigation, but also through alterations to the soil microbiology and the soil improving properties of

organic matter additions. Non-biofumigant crops have some other disease-suppressive properties, but don't have the added benefit of biofumigation. These other non-biofumigant-type disease-suppressive crops should be used in situations where the biofumigant crops don't fit into the rotation or situation very well, or in places where a particular type of crop is needed. For example, if you already have a Brassica crop as part of your cropping system, may not want to use a Brassica green manure, as it could increase disease in your other Brassica crop. There may be situations where a green manure is not feasible, or a grain or grass crop is needed for other reasons. These are situations where a non-biofumigant crop that has disease-suppressive potential would be most useful. The specific rotation crop used, whether disease-suppressive or not, should always be based on the best fit with other crops in the rotation and what the primary purpose the rotation crop needs to achieve within that rotation.

Q: (Bruno Parisi)

Do you have experience about black dot (*Colletotrichum coccodes*) and stem rot (*Sclerotium rolfsii*) suppression with crop rotation/green manure strategies in US?

A: No, I have not had direct experience with potential effects on either black dot or stem rot disease, simply because we have not observed those diseases in our trial fields. So I do not have evidence or examples of control for those diseases, but we have observed disease reductions for multiple pathogens and all soilborne diseases that were observed in a given field. So, we would expect at least some degree of control. However, it has been reported that *C. coccodes* can colonize the roots of yellow mustard and canola, so it is possible that some Brassicas could potentially increase black dot by serving as a host plant, but I do not have any data that shows whether that is actually the case or not.

Q: (Michaela Schlathölter)

In Germany we mostly use radish as precrops before potatoes. Did you tested also radish?

A: Yes, we have looked at radish, including oilseed radish, forage radish, and tillage radish as potential disease-suppressive rotation or cover crops, and we have seen overall good results. Growers particularly like to use tillage radish as a fall cover crop for its beneficial effects on reducing compaction and aerating the soil.

Q: (Matthew Back)

What is your hypothesis regarding the effect seen when 'mustard' grown as a cover crop?

A: Even when biofumigant crops are grown as a 'regular' crop or cover crop and not incorporated as a green manure, they still produce the same glucosinolate compounds in the roots and root secretions while growing, and these are still capable of producing a partial biofumigation effect in the soil. In this case the biofumigation effect is much smaller, because there is not the large quantities of biomass incorporated that is needed for the full effects of biofumigation, but there is still an effect. However, I think the most important effects are changes to the soil microbiology that result in disease-suppression. Brassica plants appear to affect the soil microbiome differently than other plant groups, and I think

these changes are important in the development of disease-suppression. These types of changes, based more on soil microbiology, are also the most likely mechanism involved with other non-biofumigant disease-suppressive crops.

Q: (Mariel Mitidieri)

Do you have any experience for sweet potato?

A: No, I do not have any direct experience or knowledge with disease-suppressive rotation crops for sweet potato. But the soilborne pathogens that impact sweet potato are similar to those affecting other crops (*Fusarium*, *Streptomyces*, etc) and the same strategies should provide at least some control.

17 Multi-service cover crops: towards a new paradigm for biocontrol and soil fertility enhancement

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Usually grown between two cash crops in an annual rotation, cover crops can increase species diversity and provide a number of ecosystem-services as an efficient tool to support sustainable agricultural production based on the principles of agroecology. Brassicaceae or crucifers can generate glucosinolate-related biocidal effects both during the growing period when hydrolysis products such as isothiocyanates are released in the rhizosphere around the growing roots or when tissue disruption occurs following cover crop termination and incorporation. The new paradigm of multi-service cover crop has been proposed by Couedel et al. (2019) to capture these multiple benefits and emphasise the need to refocus cover crop biocontrol research from a largely “pesticide” paradigm, targeting maximum production of bioactive compounds by sole crucifer cover crops, to a multi-service paradigm in which selected crucifer-legume mixtures may offer promise in the quest for the sustainable intensification of agriculture. By taking a wider systems approach, beyond pest management, and then by using a multi-service paradigm, cover crops may enhance the physical, chemical and biological fertility of the soil. Understanding the trade-offs between abiotic and biotic services within a system approach is now key to assess services and disservices provided by cover crop mixtures. Now, the potential provision of multi-services depends on species or mixture of species, the management of the cover crop, the pedo-climatic conditions and the cash crop sequence involved. We showed that the biocontrol services of crucifer sole crops could be largely maintained in crucifer-legume mixtures while improving nutrient services and C sequestration, and reducing potential disservices, such as water and nitrogen pre-emptive negative effects for the following cash crop. However, multi-service cover crop mixtures cannot be seen as a stand-alone solution to manage pests and pathogens, but it is likely that adequate levels of control will require combinations with other pest management tactics.

Keywords: multi service cover crops, biofumigation, biocontrol, intercropping, *Brassicaceae*, *Fabaceae*, glucosinolates, disservices, trade-off management, system approach

Link to presentation on Youtube: <https://youtu.be/FLf59R0IBu0>

Answers (A) from A. Couëdel to questions (Q) asked after the presentation.

Q: (Luca Lazzeri)

Which is the benefit to mix biofumigant products with legumes?

A: Legumes enable to bring exogenous nitrogen from the atmosphere through symbiotic fixation. Once terminated legumes will then bring a green manure effect as more nitrogen will be added to the system for the next crop compared to pure stands of crucifers.

See 10.1016/j.eja.2016.05.010 and <https://doi.org/10.1016/j.agee.2017.11.017> for more information.

Q: (John Duff)

What proportions of biofumigants and legumes did you use?

A: We use a 50%-50% crucifer legume mixtures. See <https://doi.org/10.1016/j.agee.2017.11.017> for more details.

Q: (Mohamed Besri)

I agree that the multi-service cover crop mixtures cannot be seen as a stand-alone solution to manage soilborne pathogens, but it should be a component of an IPM program. Have you developed such a program for a specific cash crop?

A: No we haven't but colleagues from Australia (John Kirkegaard, James Hunt) are working on it to mix some solution (planting date, GM canola, biofumigants, soil preparation)?

Q: (Xavier Boussetin)

If I understood well you used a model to estimate the S and N mineralization. I would like to know which model it was?

A: It was linear equations:

$$N_{\text{mineralised}} = N_{\text{acquired}} \times (0,72 - 2,657 \times C:N)$$

$$S_{\text{mineralised}} = S_{\text{acquired}} \times (70 - 0.16 \times C:S)$$

Please see <https://doi.org/10.1016/j.agee.2017.11.017> and <https://doi.org/10.1007/s11104-018-3615-8> for more details.

Q: (Arnoud Maaswinkel)

How to present and convince (to) the farmers of the various advantages of cover crop mixtures?

A: Compared to pure stands cover crop mixtures enable to increase biomass from 20 to 30%, then it adds more carbon into the soil. Using legumes add exogenous nitrogen to the system thanks to symbiotic fixation, reducing the reliance on N fertilizers for the next crop. Mixtures

reduce risk of cover crop failure as an unfavorable year for one specie may be compensated by the other specie and vice versa.

Q: (Michaela Schlathölter)

How to convince the farmer, as legumes are more expensive?

A: Legumes enable to save nitrogen fertilizers and help to store more carbon into the soil as it enable a 20 to 30% biomass increase in a mixture compared to pure crucifers. Then legumes are key for soil fertility and would help save money on the long term.

Q: (Mohamed Besri)

You are proposing a new paradigm of multi-service cover crop to capture the multiple benefits of bio fumigation. This paradigm takes a wider systems approach, beyond pest management, by enhancing the physical, chemical and biological fertility of the soil. Has this multiservice been adopted by farmers?

A: Yes farmers are already adopting mixtures of cover crops sold by seed companies for multipurpose. They target soil decompaction, nitrogen fixation, bee populations enhancement, biocontrol ...

Q: (Dale Gies)

Are there considerable nematode hosting concerns with the legumes?

A: We didn't hear about any hosting nematode problem with a legume but for more information please check: <https://linkinghub.elsevier.com/retrieve/pii/S0065211319300562>

Q: (Luis Fernandez)

Do you plan to combine cattle raising with agriculture?

A: It can be done but you will recycle less nitrogen and carbon to your soil as the aboveground part will be removed. It depends on the services you are targeting.

Q: (Lidia Sukovata)

How did you mix the plants? One by one or in rows?

I wonder if the plant density could have any effect on the outcome? Particularly when you take into account that legumes are slowly growing plants.

A: We mixed the seeds into the tractor seeder when planting.

Yes the plant density will affect the outcome. We saw that adding more legume density enable to have more N from the atmosphere while it will not be detrimental to the crucifer as legumes are not very competitive. On the other hand, increasing the crucifer density is not recommended as competition will be too high for both legumes.

18 Root exudate analysis of buckwheat and oat in the presence of redroot pigweed

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Cover crops as an ecological weed management technique are of great interest for farmers. Several studies have been performed to evaluate the effect of cover crop biomass on weed suppression. However, little is known about the impact of allelopathy on weed growth in a cover crop stand.

Our experiments combine field trials and a metabolic approach with the aim to identify chemical compounds produced as the result of the interaction of a cover crop and a weed. For this, we studied buckwheat (*Fagopyrum esculentum*) and black oat (*Avena strigosa*) root exudation in the presence and absence of the weed redroot pigweed (*Amaranthus retroflexus*) with a splitroot system allowing to collect specifically the buckwheat and black oat root exudates. We could show that the two cover crop species modify their root exudation in the presence of redroot pigweed. Buckwheat root exudates lead to pigweed root growth reduction.

Our results suggest that allelopathy is a factor to be considered in weed management by cover crops. Inducible allelopathic root exudates could provide a growth advantage to allelopathic crops.

Keywords: cover crop, weed control, allelopathy

Link to presentation on Youtube: *This presentation is not available as video*

Answers (A) from A. Gfeller to questions (Q) asked after the presentation.

Q: (Mohamed Besri)

Could you please explain more what do you mean by induced allelopathy and how can we induce it?

A: The ability to induce allelopathic compound production in response to competition might be adaptive, as plants can save costs of metabolite production in the absence of competitors.

The presence of a neighbored plant and the increase of competition for resources due to the presence of the neighbored plant might induce allelochemicals release.

Q: Why did you choose *Fagopyrum esculentum* and *Avena strigosa* as cover crops to control *Amaranthus retroflexus*?

A: Because we observed in field assays and pot assays growth reduction of *Amaranthus retroflexus* in presence of this two cover crops and in the agronomical context of Switzerland this two cover crops have potential.

Q: (Pieter Van soest)

Can exudates from buckwheat used as herbicide against pigweed?

A: At that stage, we are interested in understanding the mechanism of growth suppression of *Amaranthus retroflexus* by *Fagopyrum esculentum* and *Avena strigosa*. We do not have any cue that buckwheat root exudates could be used as herbicides.

Q: How does it disintegrate?

A: Some soil microorganisms uses root exudates as nutrient sources. Considering this, we hypothesize that they can be rapidly degraded or transformed by soil microorganisms.

Q: (Setu Bazie Tagele)

Would it be selective or have abroad spectrum effect?

A: We cannot give explanations at that stage of the research.

19 New Rhizobiales strains isolated in North Italy from *Crotalaria juncea* Linn. nodules and production of inoculants

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This communication reports on the results and achievements related to isolation of Rhizobiales strains from Sun Hemp (*Crotalaria juncea* Linn.) nodules. Sun Hemp, a tropical leguminous plant, was found nodulating by symbiotic bacteria in a biogeographic area where the plant is not autochthonous; and in that soil it was never cultivated before.

The isolated strains have been checked to be able to nodulate Sun Hemp in controlled condition. Those strains are used to produce single and multi-strains inoculants suitable for seed treatment.

The research has been started and carried out in the last 3 years (2017-2019).

Keywords: Rhizobiales, rhizobium, nitrogen fixing bacteria, sun hemp, *Crotalaria juncea*, inoculant, seed treatment

Link to presentation on Youtube: *This presentation was cancelled*

20 Breeding of green manures and cover crops: Biofumigation, resistance, biocontrol and organic matter

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Green manure and cover crops become an increasing importance in solving challenges of today agricultural: increasing pressure of plant pathogens while chemical solution are limited and crop rotations and crops are subject to rapid change and high economic pressure. The complexity and rapid adaptation of plant parasitism require breeders for the constant search of new resistance sources and strategies in order to ensure high and stable yield for the farmers. Since crop resistance sources are limited, usage of cover crops proved to be a successful method for disease control. In addition, cover crops also contribute for improving soil fertility by adding organic matter, efficient nitrogen fixation, erosion control, enrichment of the biodiversity and beneficial soil microorganisms and thus help for sustainable farming and environmentally friendly crop production.

Different breeding approaches at P.H. Petersen Saatzucht Lundsgaard GmbH improve the use of cover crops for biofumigation, resistance, biocontrol and soil fertility.

Link to presentation on Youtube: <https://youtu.be/fmmqfopiBGE>

Answers (A) from M. Schlathölter to questions (Q) asked after the presentation.

Q: (Annalisa Saccardo)

If I have well understood with this sustainable practices you have studied you have more environmental benefits but the yields decrease. So how the reduction in income is compensated for farmers?

A: There is no real reduction of income, because the cover cropping is outside of the normal cash crop growing time. After harvesting the main crop cover cropping takes the time in the field till planting the next cash crop. In our area it is harvesting winter cereals in July and the next crop is sugar beet or corn or potatoes or faba beans..... The cover crop in between covers the soil with all the benefits in weeds suppressing, erosion control and pushing the soil fertility. If you choose the right cover crop you will have more yield of the main crop (or better quality). Mostly the better main crop afterwards compensates the costs for cover cropping (depending on prices for the main crop harvest the profit varies)

Q: (Mohamed Besri)

Are the sowing dates for Germany ?

A: Traditional the sowing times were after harvesting winter cereals (early begin of July with barley and later after winter wheat harvest. There cover cropping was mainly against nematodes, more organic matter on sandy soil or forage production (gras/clover mixtures). Actually we have a lot of programs and motivations for the farmer to have cover crops against erosion (late sowing after corn harvest in October), winterhard cover crops in water protection areas, no chemical used areas to maintain wildlife, ...So it becomes more and more manyfold.

Q: The multiresistant radish varieties are resistant to which pathogens?

A: The term 'Multiresistant' was introduced to distinguish varieties with resistance against Beet Cyst Nematodes from those which also reduce *Meloidogyne chitwoodi*. But there is also resistance against *Meloidogyne incognita*, *Meloidogyne hapla*, Corky Ringspot (transmitted by *Trichodorus*), *Heterodera cruciferae*, *Pratylenchus crenatus*, *Rhizoctonia solani*, *Plasmodiophora brassicae*... That means it is the umbrella term for varieties with several resistances and reduction possibilities.

Q: (Martin Koller)

You demonstrated very well, that blue lupin is promising in helping against compacted soils. How long lupins has do been grown for this effect or which sawing time one has to aim, for this effect (under German conditions)?

A: Normally the root development is the within 8 weeks. As pionier plant in soils which have to restricted lupins are used in spring growing and destruction in autumn. If you use lupin for

several years you have to take care for diseases because you can not grow every type of lupin for several years.

Q: (Roberto Matteo)

Do your company work only in field conditions for breeding? Or you also use lab techniques to improve your varieties?

A: We work very traditional in cover crops breeding by selecting plants by their phenotypical behaviour – in the field and in greenhouse experiments. There is a lot of handwork to be done for selecting and we really have experienced stuff for testing 40 to 80000 single plants against nematodes each year. But we are looking for possibilities which are offered for the big crops: invitro techniques, PCR, maybe.... So we set up a invitro nematode laboratory to widen up nematode testing and we are in close contact to Universities and Institutes for further educating and techniques.

Q: (Lidia Sukovata)

What is your procedure for advising farmers?

Do they need to do some chemical analyses or analyses of soil structure prior to asking for your help?

A: The general questions are: what is your crop rotation, what is soil type and which area is your agricultural business. I would say 85 % of the decisions could be done with this answers. If not, we ask: do you notice problems in your crop production (less yield, lodging, spots on potatoes....) and if not we ask for soil analysis by soil analysing institutes or companies, do have a look inside the soil. It is more often a disease than a soil problem itself.

Q: (Alessandro Infantino)

Do you suggest the use of cover crops as prevention or as a cure for diseases? Or both?

A: I am a real fan of good crop rotation. For me it means I do not give risk disease multiplication a chance. So if I take care as a farmer not to overheat with the same crop or crop family it is the basic for solid and good yield.

As you are not able to make the best decision for crop rotation and sometime you have to grow a crop because of the profit more often than it is good for pest prevention, we also look for solution within cover crop to cure diseases by non host (nematodes and other insects) or having a special ingredient to kill or repel an attacker. I would say 85 % of the decisions could be done with these answers.

Q: (Mariel Mitidieri)

What are the more efficient cover crops for wire worm control?

A: I would go one step backward: Choose a cover crop which is not multiplying and adapt your soil preparation and crop rotation to get wire worms controlled or on a low level

without profit losses.

Q: (Manuela Bagatta)

What about flax as a cover crop? Do you practice autumn sowing?

A: Yes, we practice autumn growing with flax. Flax is relatively healthy, small seeded and a strong partner also together with strong growing brassica cover crops.



Picture (linseed in blend with *Raphanus sativus*)

For the GAP regulations in Germany a farmer has to grow cover crops as a blend to get the subsidies. There are specific regulations and linseed fits perfectly in most of the cases.

Q: (Martin Koller)

If you compare the resistance of oil radish to marigold, which one work better against RKN?

A: I did not know that there is resistance in marigold against RKN. The problem of RKN is: they can use a lot of plants for multiplication and/or to survive. So if you cannot kill the directly you have to reduce the possibilities for multiplication and/or surviving. That is possible if you destroy every potential multiplying plant (bare soil for 8 weeks). But then your soil has no structure any longer.

Therefore we looked for plants which are covering the soil as good as possible to reduce potential host plant weeds, are rooting – but do not multiply the nematode. Actually the PKN resistant plant are as good in reduction as bare soil for more than 8 weeks, but with additional booster for soil life and without soil compaction.

21 Use of Marigolds (*Tagetes* spp) as cover crop for the control of tomato root knot nematodes (*Meloidogyne* spp) in Morocco

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Morocco is the fourth tomato exporting country in the world. The region of Souss Massa (Agadir area), produces 90% of the country exported vegetables including 60% tomato . This crop is attacked by many soil-borne pathogens which are used to be controlled by Methyl Bromide. The loss of this fumigant has encouraged several countries to develop, demonstrate and adopt alternatives to this powerful ozone layer depleting chemical. In the region, many environmental conditions are favorable to the development of root-knot nematodes (RKN), *Meloidogyne javanica*, *M.incognita*, *M.hapla* and *M.arenaria*, the first two species are the most common (96 % of the nematodes population). A widely sustainable control method of RKN reported in many countries is the use of Marigold (*Tagetes* spp.) as green manure. Experiments with this cover crop have been conducted during two years in the region. *T.erecta*, *T.patula* and *T.minuta* were chosen because of their efficacy in controlling the four nematodes species. A seeds mixture of the three *Tagetes* spp. were sown, then, after two months, the plants were plowed under and incorporated into the soil shortly after flowering. Seedlings of a tomato hybrid (Daniella) susceptible to RKN were planted in the plots amended and non amended with *Tagetes* spp. *Meloidogyne* larvae (L2) population in the plots before and after soil amendment and at the end of tomato crop, root knot indexes (end of the tomato crop), total tomato yields and exported productions were determined. L2 populations in the plots amended with *Tagetes* spp were significantly lower than in the non amended ones. Total tomato yields and exported tomato were significantly increased by *Tagetes* and the nematodes severity significantly reduced. However, the results obtained in the Souss Massa region should not be generalized to other tomato producing regions because the efficacy of *Tagetes* is specific to a nematode or to some nematodes. Therefore, it is important to know the importance and the distribution of the *Meloidogyne* species present in the tomato producing region in order to choose the most effective *Tagetes* species. *Tagetes* spp. do not completely eradicate RKN. Therefore, it should be a component of an IPM program. In organic tomato production, currently, Marigold is never used alone, but always in combination with other non chemical control methods. In conventional tomato production, this technology is also not considered by the vegetable growers by itself as a control method, but it is combined with others chemical and non chemical control methods.

Keywords: Cover crop, *Tagetes* spp, *Meloidogyne* spp, tomato, Morocco

Link to presentation on Youtube: <https://youtu.be/JehyammmWzA>

Answers (A) from M. Besri to questions (Q) asked after the presentation.

Q: (Aureli Gfeller)

Do you know how RKN are controlled by incorporated marigolds? Based on literature, which compounds from marigold affects?

A: Marigold may reduce PPN populations by several means, including (1) acting as a non-host or a poor host, (2) producing allelopathic compounds that are toxic or inhibit PPN development, (3) creating an environment that favors nematode antagonistic flora or fauna or (4) behaving as a trap. These mechanisms may occur separately or in combination resulting in lower PPN numbers. Marigold roots release the chemical alpha-terthienyl, one of the most toxic naturally occurring compounds found to date. This compound is nematicidal, insecticidal, antiviral, and cytotoxic. The presence of alpha-terthienyl inhibits the hatching of nematode eggs. However, it is unclear if marigolds producing alpha-terthienyl inhibit development because of the alpha-terthienyl itself or because marigolds are a non-host for certain nematodes. Nematodes may not feed or develop on non-host plants even when they do not contain allelopathic compounds (Krueger et al, 2019, Karakas M and Bolukbasi 2019, Bhattacharyya, M. 2017)

Q: (Martin Koller, Michaela Schlathölter)

If you compare the resistance of oil radish to marigold, which one works better against RKN?

A: In Morocco, no experiment in Morocco was conducted to compare the effect of Marigolds and of other plants including oil radish. In several studies, the efficacy of nematode suppression by marigold was compared to other cover crops. The results obtained are extremely variable. They depend on several factors such as the population of nematodes, the inoculum density, the soil texture and the soil temperature (Hooks et al 2010).

Q: (Mariel Mitidieri)

Mohamed, what is the cost of marigold seed?

A: *Tagetes erecta*, *T. patula* and *T. minuta* are widely distributed in Morocco: In our experiments, the seeds were collected from *Tagetes* wild plants. As reported in my presentation, *Tagetes* seeds are also available in the market. The costs vary with the species, the varieties and also with the seed companies. The seeds prices are available on the web sites of the seed suppliers e.g. <https://seedmegastore.co.uk/herb-marigold-pot/>, <https://seedmegastore.co.uk/herb-marigold-pot/>

Q: (Cliven Njekete)

Are there practices in Morocco where marigold is used as an intercrop or flower strip and still useful in reducing PPN in the crop?

A: In Morocco, *Tagetes* is used only as cover crop. The soil is first cleaned from plant residues, and then *Tagetes* spp. is sown (0, 5 g of seeds /m²). The cover crop is frequently

ferti-irrigated . After 2 months, the plants are incorporated into the soil and the cash crop planted (Tomato, . .). Many research have shown that intercropping with marigold did not reduce plant-parasitic nematodes. Typically root-knot nematodes will find and reproduce on roots of a susceptible crop or weed. So interplanting marigold and susceptible crops is very risky and may result in damage to the susceptible crops (Krueger et al.2019, Cerruti 2010, Hooks 2010).

Q: (Matthew Back)

Thank-you Mohamed Besri. Other than root knot and root lesion nematodes, are you aware of any work showing suppression of free living PPNs using marigold?

A: In addition to the control of RKN, *Tagetes* spp. also suppress reniform nematode (*Rotylenchulus reniformis*), lesions nematodes (*Pratylenchus penetrans* and *P. pratensis*), burrowing nematode, (*Radopholus similis*), spiral nematode (*Helicotylenchus multicinctus*), and lance nematode (*Hoplolaimus indicus*) of various crops (Karakas and Bolukbasi 2019, Bhattacharyya,2017)

Q: (Caroline Caporalino)

Do you know why *Tagetes patula* controls *M. arenaria* and *M. javanica* and not *M. incognita*? Specificity can come from R-genes but compounds produced by *T. patula* may act on all species. Do you have references of this specificity?

A: The bioactive compounds of different marigold species and cultivars may differ in composition, quality and quantity. Thus, certain species may be highly effective against one nematode species but have limited to no impact on or possibly increase populations of other plant parasitic nematodes. Suppression of PPNs by marigold is variable. Therefore, it is critically important to know which nematode species is responsible for crop damage before selecting a marigold plant (Karakas and Bolukbasi 2019, Bhattacharyya., 2017, Ploeg, 2002)

Q: (Manuela Bagatta)

Marigold attracts pollinators. Did you notice any higher presence of insects on the flowers?

A: We did not study the attractiveness of bees by *Tagetes*. Therefore, I could not answer this question. I should greatly appreciate receiving some references.

Q: (Lidia Sukovata)

Mohamed Besri ,what are requirements of *Tagetes* to soil, including pH?"

A: Moroccan soils are basic. We do not know the impact of the pH on the effectiveness of *Tagetes*. Such studies should be carried out in the laboratory, under controlled conditions. However, it has been reported that soil temperature is an important factor for nematode suppression by marigold (Karakas and Bolukbasi 2019)

Some selected references

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22 Eco-friendly alternatives against soil-borne diseases in strawberry cultivation in Martell Valley (South Tyrol, Italy)

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Strawberry production in South Tyrol (Italy) is an economically important item of the local horticulture because it supplies the national market with late strawberries. Besides a high level of soil organic matter, soil health is currently maintained with periodical intervals with other crops, especially *Brassicaceae*, or with periodic intervals with permanent vegetal cover. However, the interest in eco-friendly alternatives for controlling soil borne pathogens is currently growing. Within this framework, an experiment was carried out in a strawberry specialized valley of South Tyrol, the Martell Valley (1,312 m a.s.l.). This experiment was conducted in a field where strawberry has been repeated for 15 years. That field showed typical crop decline symptoms such as reduced growth in post-transplant and collapse at harvesting. One month before transplanting, three random rows across the field were assigned to each of the following treatments: anaerobic digestate of liquid manure, compost from anaerobic digestate of organic fraction of municipal solid waste (OFMSW), a commercial product (Herbie72[®]), an experimental product, Dazomet (Basamid[®]) as positive control and untreated control. Strawberry plants were cultivated on raised beds with plastic films and drip irrigation. Vegetative biomass, yield and fruit quality were evaluated at the end of two production cycles (2019 and 2020). Our preliminary results showed that Herbie72[®] and Dazomet increased the above-ground biomass (+30% as compared to untreated plants) and both gave over 50% increase in yield per plant. Promising results came from our experimental product which doubled the yield and plant biomass in comparison to untreated plants during the second year. The compost from OFMSW gave phytotoxic effects in the first year, probably due to a too short interval between application time and transplant. On the contrary, it gave a significant yield improvement compared to untreated during the second year. The digestate of liquid manure performed similarly to the control. Fruit quality traits were not affected by treatments. Preliminary data suggested that strawberry cultivation in Martell Valley needs strategies for maintaining and improving soil health. At the end of this study soil microbial populations will be investigated to relate microbial changes with crop response to the treatments and further orient the actions for controlling soil borne pathogens.

Keywords: *Fragaria × ananassa*, soil-borne diseases, nonchemical alternatives, organic amendment, plant growth, soil biology

Link to presentation on Youtube: *This presentation is not available as video*

Answers (A) from S. Soppelsa to questions (Q) asked after the presentation.

Q: (Soraya Franca)

Have you checked N levels in soil after Herbie application?

A: NA.

Q: (Hagen Thoss)

What was the origin of the compost?

A: Compost was obtained from anaerobic digestion of organic fraction of municipal solid waste (OFMSW).

Q: (Stefano Mocali)

Have you checked the effects of Herbie on soil biota?

A: The analyses are ongoing.

Q: (Lidia Sukovata)

Did you make those treatments in both years or only in the first year?

A: Treatments were applied before the transplant (only in the first year).

Q: (Natalia Meneguzzi)

Which are the main soil pathogens you found?

A: Some pathogens are: *Pestalotia longisetula*, *Cylindrocarpon*-like fungi, *Pythium* sp.

Soppelsa, S.; Manici, L.M.; Caputo, F.; Zago, M.; Kelderer, M. Locally Available Organic Waste for Counteracting Strawberry Decline in a Mountain Specialized Cropping Area. Sustainability 2021, 13, 3964. <https://doi.org/10.3390/su13073964>

Q: (Setu Bazie Tagele)

How many times is the experiment replicated?

A: A field trial over three growing seasons (2019, 2020, 2021) was conducted. Treatments were applied before the transplant (only in the first year). The experiment setup was organized as a completely randomized block design with three replicates per treatment.

23 Is diversity of service plant species a way to archive higher biomass and N accumulation in winter oilseed rape - service plant intercropping?

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Winter oilseed rape (WOSR) intercropping with frost sensitive service plant (SP) is a popular practice in Switzerland. SP are known to contribute to weed control, and thanks to N accumulation in fall, could provide nitrogen to the soil after their destruction by frost and improve nitrogen availability for WOSR in spring. Most farmers use SP mixtures of four to ten species. This diversity was pointed out as a potential way to increase both the number and the stability of ecosystem services provided by SP. However, biomass production and nitrogen accumulation of complex SP mixtures has not been widely studied. In a two-year field experiment, we assessed the biomass of SP mixtures intercropped with WOSR. The mixtures were made of one, two, three (faba bean, Berseem clover and niger) or seven species. In a greenhouse experiment, we studied different combinations of faba bean, grass pea and niger intercropped with WOSR. Both experiments focused on the early stage of WOSR and SP development. The biomass and N content of SP and WOSR, as well as N₂ fixation of legumes SP were measured. In the field, the seven species mixture produced the highest biomass both years, mostly because of buckwheat. High biomasses were also found in the mixtures including niger, but only in the second year. In the greenhouse experiment, the faba bean produced more biomass and fixed more N₂ than grass pea, resulting in a general higher nitrogen accumulation in all modalities with faba bean. In these experiments, biomass production and N accumulation were mostly driven by a single component of the mixture. However, total biomass varied between the 2 years in the field and different species could take the advantage depending on the year conditions. Additionally, different species could contribute to total biomass, N fixation or total N accumulation.

Keywords: Winter oilseed rape, specific diversity, service plant, nitrogen symbiotic fixation

Link to presentation on Youtube: *This presentation is not available as video*

Answers (A) from X. Boussein to questions (Q) asked after the presentation.

Q: (Frédéric Le Dily)

Is the low SIR measured in WOSR just a consequence of winter conditions?

A: The soil Substrate Induced Respiration (SIR) was measured in WOSR early stage (BBCH 19) in a greenhouse experiment where the mean temperature was $15\pm 1.8^{\circ}\text{C}$. Which is close to WOSR fall growing conditions. All the other species was grown in the same growing conditions.

24 Novel microbial-based bioproducts improving soil biodiversity and the effectiveness of biocontrol and biofertilization practices in horticulture

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The H2020 project EXCALIBUR - “Exploiting the multifunctional potential of belowground biodiversity in horticultural farming” will be briefly presented: the project aims to develop a comprehensive strategy of soil management improving the effectiveness of biocontrol and biofertilization practices in horticulture. Specifically, the presentation will highlight the needs for a new approach on soil management in horticulture, taking into account the potential role of the native soil biodiversity for enhancing the efficacy of biocontrol and biofertilization practices in horticultural farming. To do that, new multifunctional soil microbial inoculants and bio-effectors (compounds or by-products which directly or indirectly enhance plant performance) will be tested on three model crops (tomato, apple, strawberry) under conventional and organic management across Europe. The overall approach and the first results will be discussed.

Keywords: H2020, microbial inocula, soil biodiversity, bioproduct, biocontrol, biofertilization

Link to presentation on Youtube: <https://youtu.be/BKZ2CU3ehno>

Answers (A) from S. Mocali to questions (Q) asked after the presentation.

Q: (Michaela Schlathölter)

Do you have an idea, how many years of vegetation it takes to see a change in soil organism?

A: It depends on soil quality, soil organisms and vegetation. In case of microbial communities, it is well known that soil microbiome often changes in response to different plant species, which can in turn modify the composition and productivity of plant communities. It was also shown that the same vegetation might differently affect diverse soil organisms. For instance, some studies showed that grassland vegetation influences fungal communities (particularly mycorrhizae), whereas bacterial community structure is influenced more by the quality or composition of soil organic matter, thereby reflecting carbon inputs to the soil over decades (<https://doi.org/10.1007/s10705-009-9314-3>)

Q: (Setu Bazie Tagele)

What kind of additive carriers do you use for the microbial consortia? Microbial proportion?

A: As additive carriers you can add everything...clay minerals, skim milk, rock phosphate, starch, etc. The proportion should be previously studied but 1 to 3% is a good one. The additives play different roles: from improving the carrier structure to provide nutrients. Briefly, 1 to 3% of gel carrier + microbial cells or spores + 1 to 3% additives. This scheme is valid for 1 single microorganism and consortium as well.

25 Plant-derived sources for anaerobic soil disinfestation in Southern California

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Anaerobic soil disinfestation (ASD) has been optimized and adopted as organic alternative to chemical fumigation by strawberry growers in California as a method of treating soil that utilizes a carbon source and moisture to produce anaerobic conditions in soil for 2-4 weeks. As a result of chemical, physical and microbiological changes in soil, ASD suppressed important strawberry pathogens *Macrophomina phaseolina*, *Verticillium dahliae* and controlled some weeds, while providing yields similar to those in chemically fumigated soil. ASD was also efficacious against *Fusarium oxysporum* f. sp. *fragariae* in soils with temperatures above 30 C. Southern California warm soils in July-September are suitable for ASD conducted in preparation for strawberry planting in October. Continued ASD adoption is hindered by increasing cost of rice bran, currently widely used carbon source. We evaluated alternative locally produced or sourced plant-based products that can be more affordable than rice bran. ASD with drip-applied glycerin did not have adequate distribution in beds and performed similar to untreated check. ASD with incorporated coffee grounds from one supplier provided 83% increase in fruit yields compared to untreated soil but grounds from a different supplier decreased strawberry yields. Carbon, nitrogen and their ratios had important impacts on efficacy of ASD with coffee grounds and grape pomace. ASD with wheat midds at 20 t/ha provided strawberry yields similar to chloropicrin-fumigated soil, while substituting 30% of rice bran carbon rate with on-site grown cereal cover crop biomass resulted in yields similar to full rate of rice bran but at reduced cost. As we continue exploring cost-effective methods of soil disinfestation, we utilize ASD integrated with other pest management tools, such as use of resistant cultivars and crop rotation for sustainable production

Keywords: Carbon to nitrogen ratio; soil-borne pathogens; soil chemistry and microbiology; organic strawberry production.

Link to presentation on Youtube: <https://youtu.be/XNJBGUw7xKg>

Answers (A) from O. Daugovich to questions (Q) asked after the presentation.

Q: (Vincent Michel)

Are there commercial products available, like Herbie in Europe?

A: In US there are companies marketing sources for ASD under different names on limited basis, and the products are approved by Organic Martials Research Institute. But active ingredient is essential, as for example, rice bran for California systems.

Q: (Alessandro Infantino)

Is ASD utilized for other soilborne diseases?

A: Yes, ASD has been reported (in multiple countries) efficacious for controlling *Fusarium* spp., *Verticillium dahliae*, *Rhizoctonia solani*, *Sclerotium rolfsii*, *Pyrenochaeta lycopersici*, *Colletotrichum coccodes* as well as *Phytophthora* and *Pythium* spp. and nematodes such as Root knot nematodes (*Meloidogyne* spp.) and lesion nematodes (*Pratylenchus penetrans*).

Q: (Mariel Mitidieri)

Oleg, do you think that anaerobic treatment is a kind of biofumigation using C₂O?

A: We do not consider it as fumigation, rather a dramatic shift in microbiological, chemical and physical properties in soil during and after ASD that create soil environment that benefits plant growth and pest and pathogen suppression.

Q: (Frédéric Le Dily)

Do you have an idea upon the sustainability of this practice over the years (hypoxia?, reduction of microbial diversity?)

A: The continuous ASD (season after season) has been evaluated for no more than 3 years and, since post ASD (3-4 weeks) soil returns to aerobic state for the rest of the season we did not see reduction in microbiological diversity. There is evidence of increases in non-pathogenic and decreases in pathogenic fungi over several seasons in similar work in Spain in strawberry.

Q: (Natalia Meneguzzi)

Fusarium is a very common soil fungi, as well as entophytic, I wonder if it is a real problem for strawberries?

A: Most of *Fusarium* we recover from strawberries are non-pathogenic but in the last 13 years *F. oxysporum* f. sp. *fragariae* has been lethal to many strawberry cultivars as it enters vascular system, blocks passage and the plants rapidly desiccate and wilt. This pathogenic *Fusarium* has been characterized and evacuated widely in California, Japan, Australia and

Southern Europe (please check on references closest to you. For example for California:
<https://www.tandfonline.com/doi/full/10.1080/15538362.2016.1219294>

POSTERS

The posters are available on the Agroscope website :

<https://www.agroscope.admin.ch/agroscope/en/home/news/events/biofumigation-7/posters.html>

P01: Potential of Brassicacea cover crop and biofumigation to reduce *Verticillium dahliae* germination and Sunflower *Verticillium* Wilt

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P02: Examining biofumigant crops for the management of Pea foot rot complex pathogens
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P03: Biofumigation experiences in Argentina

Mitidieri M.S. *et al.*, Instituto Nacional de Tecnología Agropecuaria, Argentina

P04: Sulphate catch cropping performances of rapeseed and mustard species

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P05: Soil microbiome characteristics associated with long-term potato cropping system management practices

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P01 Potential of Brassicacea cover crop and biofumigation to reduce *Verticillium dahliae* germination and Sunflower Verticillium Wilt

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Sunflower Verticillium Wilt (SVW), caused by *Verticillium dahliae*, has been a widespread soilborne disease in France since 2011, causing up to 50% yield loss. Since the prohibition of chemical fumigants, SVW management has been challenging. The fungus can persist in the soil as microsclerotia for nearly 13 years and survive without suitable host. In this context, Brassicaceae or crucifer cover crops are interesting for pathogen suppression due to the release of biocidal hydrolysis products such as isothiocyanates (ITCs) from glucosinolates (GSLs) present in their tissues. Crucifer can generate GSL-related biocidal effects both during the growing period around roots (allelopathy) or when tissue disruption occurs following cover crop incorporation (biofumigation).

Because GSL types and concentrations vary among crucifer species, and between root and shoot tissues of the same species, this study first aimed at characterizing the GSL profile of 21 crucifers. Thus, to evaluate the potential of grinded crucifer on the fungus and biofumigation on SVW, (i) the toxicity of 5 selected crucifers was assessed on *V. dahliae* microsclerotia germination and development in a laboratory assay and (ii) two fields trials aimed at evaluating the biofumigant potential of the 3 most efficient crucifers, grown during the fallow period, by assessing weakly disease symptoms on sunflowers.

For the laboratory essay, brown and white mustard, fodder radish, turnip rape and rapeseed were selected for their higher concentration and diversity on GSLs. All crucifers reduced *V. dahliae* microsclerotia germination on growing media (PDA) by 90% (brown mustard) to 63% (fodder radish), and fungus development by 90% (turnip rape) to 69% (white mustard) compared to control treatments without tissues. In the field, biofumigation after brown mustard, fodder radish or turnip rape significantly reduced the incidence and severity of SVW compared with bare soil both years. These results indicate that Brassicacea cover crops could have a potential to reduce SVW.

Keywords: *Helianthus annuus*, soilborne fungus, crucifer, glucosinolates, agroecological service crop

No questions were asked.

P02 Examining biofumigant crops for the management of Pea foot rot complex pathogens

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The field pea (*Pisum sativum*) is an important legume grown for human consumption in over 110 countries worldwide. In the UK, growth is limited to areas within close proximity of processing facilities, which has led to intensive production and yield declines of up to 40% in recent years. This is mostly attributed to a build-up of fungal and oomycete pathogens involved in the pea foot rot complex (PFRC), in particular; *Fusarium solani* f. sp. *lisi* (FSP), *Fusarium oxysporum* (FO), *Aphanomyces euteiches* (AE) and *Didymella pinodella* (DP). Current strategies for disease management are limited, especially for the complex as a whole. Therefore, this project aims to identify green manure and biofumigant crops that can suppress PFRC pathogens.

In-vitro experiments have begun to screen several biofumigants, including varieties of *Brassica juncea*, *B. carinata*, *Raphanus sativus*, *Eruca sativa* and *Sinapis alba* for their effects on mycelial growth and inhibition of PFRC pathogens. Plant material was grown under glasshouse conditions, before being oven dried and milled to a fine powder. Primary glucosinolate concentrations were analysed with high-performance liquid chromatography. Experiments consisted of using a two Petri dish base system, one containing potato dextrose agar and a mycelial plug of a PFRC isolate, placed on top of a second base containing hydrated biofumigant powder (water only for the control), sealed together with Parafilm. Colony growth rate was assessed daily and the final percentage inhibition calculated after one week's growth (method adapted from Sexton *et al.*, 1999).

For AE, all varieties screened significantly reduced mycelial growth compared to the control. For both FSP and DP, only the *R. sativus* variety did not significantly reduce growth. For FO, only the two *B. juncea* varieties significantly reduced mycelial growth. The screens will inform biofumigant variety choice for future glasshouse experiments.

No questions were asked.

P03 Biofumigation experiences in Argentina

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Biofumigation experiences in Argentina have been held along a wide territory, and have proved to be much more effective when combined with solarization.

These practices have been successfully implemented, allowing the disinfection of soils in a sustainable manner and the improvement of their physical, chemical and biological properties.

In Corrientes a subtropical province specialized in off season production, incorporation of chicken and cattle manure into the greenhouse soil prior to solarization was effective against *Ralstonia solanacearum*, *Pythium aphanidermatum*, *Rhizoctonia solani* and *Sclerotium rolfsii*, other biofumigants essayed were pine tree fallen leaves, grass, cabbage and sorghum.

In the centre of Argentina, horticultural and ornamental crops are grown under mild winter climate. Biosolarization (biofumigation + solarization) was effective controlling *Pyrenochaeta lycopersici*, *Fusarium solani*, *Sclerotium rolfsii* and *Sclerotinia sclerotiorum*, weeds and damping off pathogens, as well as nematodes like *Nacobbus aberrans*, *Helycotylenchus* and *Criconebella*. The amendments used were chicken manure, broccoli, sorghum, tomato and pepper crop debris, mustard, rapeseed and *Brassica campestris*.

At the west of the country, in Mendoza a province with arid and continental weather, summer is hot, and good control of strawberry diseases as *Phytophthora*, *Rhizoctonia*, *Phytium*, *Verticilium*, *Macrophomina*, and nematodes as *Meloidogyne*, *Ditylenchus* has been achieved using rapeseed as fumigant in the greenhouse.

In Bahía Blanca, a city at the south of Buenos Aires province with a colder weather *Meloidogyne hapla* was controlled using cattle manure and cauliflower in spring and summer in the greenhouse, nematodes of the same genus were controlled in winter using *Melia azedarach* seeds as fumigant.

At the North of Patagonia, a semiarid region with hot summers but very cold winters, weeds in onion open field nurseries were controlled in summer using chicken manure and cabbage. Similar results were obtained at the northwest of Rio Negro province, were weeds were controlled using cabbage in spring for open field tomato crops. In the same province *Fusarium oxysporum* in onion was controlled using cabbage in autumn and summer.

Keywords: horticulture, nematodes, soil borne pathogens, soil disinfection

No questions were asked.

P04 Sulphate catch cropping performances of rapeseed and mustard species

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For more than 50 years, intensive breeding of rapeseed (*B. napus*) to reach high grain yield with low sulphur (S) content has led to low efficiency in saving nitrogen (N) fertilization. Around 1960, researchers started developing new rapeseed cultivars free of erucic acid (simple 0) and glucosinolates (double 0). Following the apparition of these new genotypes, rapeseed became less used than brown mustards (*B. juncea*) for biofumigation and green manuring. The relationships between S storage properties, N economy and biofumigant efficiency are clearly set and the objectives of this study are to demonstrate the ability of mustard (i) to store reduced S compounds and therefore (ii) to provide soil S-catch crop and S green-manure services as compared to rapeseed.

Seven week old plants (*B. napus* cv Liform ; *B. juncea* cv ISCI-99) were subjected to a Hoagland nutrient solution sufficiently supplied (S+) or limited (S-) with sulphate. Leaves, roots, stems and flowers were harvested regularly. Total S, proteic-S and SO₄²⁻ contents were determined with an elemental analyser (XRF), Bradford measurements and ion-exchange chromatography. Differences between S and SO₄²⁻ are referred as reduced sulphur (-SH).

Under S+ conditions, total S content is 50% higher in mustards than in rapeseed. Rapeseed preferentially accumulates S as sulphate, whereas mustards store S under -SH forms, whatever SO₄²⁻ availability. As compared to mustards that have been bred specifically for biofumigation and green manuring, rapeseed has partly lost the ability to store S, especially under reduced -SH forms.

Leaves of improved rapeseed cultivars are subjected to an early/regular senescing process. Concomitantly, the ability of rapeseed vegetative organs to store S is diminished, as well as the assimilation of SO₄²⁻ into reduced -SH compounds. In coming experiments we intend to rely these observations to the soil-release capabilities of organic and mineral S forms stored by infill plants.

Keywords: Brassicaceae ; cover crop ; mineral availability ; S cycle

Answers (A) from J. Enouf / F. Le Dily to questions (Q) asked after the presentation.

Q: (Mohamed Besri)

You have shown that rapeseed became less used than brown mustards for biofumigation and green manuring. I do not understand very well the correlation between sulphur and Nitrogen. Could you please explain?

A: Nitrite reductases and bacterial sulphite reductases catalyse the reduction of nitrite to ammonia, as well as sulfite to sulfide. Assimilatory nitrite reductases contain a prosthetic group termed sirohaem and an iron-sulphur cluster. S is a determinant key for N reduction and assimilation ([https://doi.org/10.1016/0005-2728\(73\)90153-9](https://doi.org/10.1016/0005-2728(73)90153-9)).

Otherwise, S is an essential nutrient in plants as a constituent element of some amino acids, metal cofactors, coenzymes, and secondary metabolites. Not surprisingly, sulphur deficiency decreases plant growth, photosynthesis, and seed yield in both legumes and non-legumes. In nodulated legumes, sulphur supply is positively linked to symbiotic nitrogen fixation (<https://doi.org/10.3389/fpls.2018.01434>).

Q: Soil health management practices (longer rotations, disease-suppressive crops, cover crops and green manures etc.) can alter soil microbial communities in specific ways. Could we develop a model to forecast these changes?

A: Yes, probably. Longer rotations and the introduction of organic fertilizers will promote the stability of soil microbial communities. Unfortunately, I am not aware of any models to predict it.

Q: (Antoine Couëdel)

Did you see links between sulphur uptake and other molecules than N ? (P, K, Mg, Ca, ...)?

A: Yes, recent studies about the changes of ionome homeostasis in response to various S fertilization reveals the complexity of nutrients networks in plants. A first interaction is linked to the large accumulation of sulfate in the leaf vacuoles, with its reduced osmotic contribution under S deficiency being compensated for by an increase in both Cl⁻ and PO₄²⁻ uptake and accumulation (<https://doi.org/10.1093/jxb/erz214>, <https://doi.org/10.3390/plants7020037>). The broad specificity of root sulfate transporters that are up-regulated during S deficiency can also increase the uptake of Mo and Se (<https://doi.org/10.3389/fpls.2021.641648>). Other interactions between S and micronutrients concern metabolic networks where several nutrients are essential, such as the synthesis of the Mo co-factor needed by some essential enzymes, which requires S, Fe, Zn and Cu for its synthesis, and the synthesis and regulation of Fe-S clusters (<https://doi.org/10.1093/jxb/erz214>).

P05 Soil microbiome characteristics associated with long-term potato cropping system management practices

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Potato cropping system practices substantially affect soil microbial communities and the development of soilborne diseases. Cropping systems incorporating soil health management practices, such as longer rotations, disease-suppressive crops, cover crops and green manures, reduced tillage, and/or organic amendments can potentially alter the soil microbiome, reduce soilborne potato diseases, and increase productivity. In field trials originally established in 2004, different 3-yr potato cropping systems focused on specific management goals of soil conservation (SC, including cover crops and limited tillage), soil improvement (SI, SC system plus compost amendment), and disease suppression (DS, inclusion of disease-suppressive green manures) were evaluated and compared to a 2-yr standard rotation (SQ, potato-barley/clover) and a non-rotation control (PP, potato each year) for their effects on soil microbial communities and soilborne disease development over time. After 12-14 years, the SI system produced higher potato yield and greater microbial activity than all other systems, as well as low disease levels. DS maintained lower disease (black scurf and common scab) severity than all other systems. Characteristics and composition of the soil microbiome, as determined by taxon identification from 16S rDNA (bacterial) and ITS2 (fungal) amplicon sequencing, were uniquely different for each cropping system. Preliminary analyses indicated that SI generally resulted in the greatest abundance and diversity of bacterial and fungal taxons among systems, with PP the lowest. For fungi, SI resulted in greater abundance of Zygomycetes, DS greater abundance of Basidiomycetes, and PP and SQ greater abundance of Ascomycetes, Chytridiomycetes, and numerous potato pathogens. For bacteria, SI resulted in greater abundance of Bacteroidetes, Actinobacteria, and Nitrospirae; DS more Firmicutes, Actinobacteria, and Thermobacteria; SC more Acidobacteria and Verrucomicrobia;, and PP more Gemmatimonadetes and Proteobacteria. These results indicate that soil health management practices incorporated into potato cropping systems can alter soil microbial communities in specific ways and sustainably reduce soilborne diseases.

Keywords: soil microbiome, cover crops, green manures, cropping systems, soilborne disease, micro-organism interactions

Answers (A) from R. Larkin to questions (Q) asked after the presentation.

Q: (Matthew Back)

What are the 'others' listed for Figure 1?

A: For the fungi, the large group of 'others' represent OTUs or sequences that could not be positively identified as part of any of the known fungal phyla. This largely indicates gaps in the fungal databases. These are fungi that could not be identified because they did not match anything in the databases at the time of analyses. The databases for environmental fungi are much less complete than for bacteria and are still being compiled. These data are from a few years ago, and we are currently re-analyzing these to see if more can now be better identified.

Q: Interesting to see the reduction of ascomycetes under the Soil Improving treatment (with compost)

A: Yes, there were very distinct differences among the different cropping systems, particularly for fungi, with a general reduction in ascomycetes and increase in zygomycetes in SI, increase in basidiomycetes in DS, and increases in ascomycetes and chytrids in PP and SQ. The differences were even more interesting at the genus level, where the nonrotation PP showed the highest levels of pathogen groups such as *Verticillium*, *Colletotrichum*, and *Alternaria*, which were much reduced in SI, and beneficial genus *Trichoderma* increased most in the disease-suppressive DS system.

Q: (Setu Bazie Tagele)

Did you find microbiome shift after biofumigation helpful?

A: First, these are results from different cropping systems that involve multiple different crops and practices over multiple years, and it is hard to definitively associate any specific changes with a specific practice such as biofumigation. Second, it is also hard to identify specific changes that could be called helpful or beneficial, at least at this point, because we don't yet know enough about the microbiome to be able to identify what 'helpful' changes look like. We do know that the microbiomes associated with the Soil Improving, Disease-Suppressive, and Standard Control systems are all very different from each other, and that that the composition of the microbiomes may be at least partly responsible for the positive and negative effects associated with these systems, but we don't yet know what specific taxa are most important for those traits. That's what we hope to determine with more microbiome analyses.

Q: (Lidia Sukovata)

Wouldn't a compost bring much more different microorganisms degrading organic matter?

A: Some of the microorganisms present in compost are quite different than those that readily occur in soil, but there are also many in soil that do not readily occur in compost (thus diversity may not be that different). The type and quantity of organic matter present is very important in shaping the microbiome, in both the compost and soil, as organic matter is the primary food source for the microbiome. One aspect that has been debated, however, is just how much or how well the microorganisms in the compost adapt or survive when incorporated into the soil, since the soil is a very different environment than a compost pile, with some saying only a small proportion remain active or important in the soil environment. However, some recent studies from Canada found that many of the prominent microorganisms in compost were still able to be detected and monitored in soil long after incorporation, indicating that they successfully established in soil, changing the microbiome characteristics.

Q: Does OTU relate to live organisms or both live and dead?

A: OTUs refer to a DNA sequence that is associated with a specific taxonomic group, which is based on DNA sequences extracted from soil. So, no it does not distinguish between DNA from living and dead organisms. However, DNA has a very short lifespan in soil after an organism dies (once the organism dies, DNA is quickly broken down in soil and thus is generally not preserved or available to be extracted). Thus, the vast majority of DNA extracted from soil comes from organisms that were living at the time of sampling, and samples are immediately frozen when taken in the field, thus preserving all DNA present at sampling time. Thus, this is generally not an issue.