

1. Cross-Protection (vaccination) against *Pepino mosaic virus severe strains* in tomato

Tomato (*Lycopersicon esculentum*) X *Pepino mosaic virus* (PepMV)

PepMV is a mechanically-transmitted plant pathogen present throughout Europe that is chiefly controlled by the implementation of strict hygiene conditions. Infection of tomatoes with the most aggressive strain of the virus causes severe fruit marbling (Fig. 1.1) but pre-infection with mild isolates in areas where the disease is endemic can provide protection and prevent symptom development (cross protection).



Fig. 1.1

Mechanism of Action & Use

Cross protection was first described in 1929 by H. H. McKinney (Fig. 1.2), who demonstrated that inoculation with a mild strain of a particular virus can induce protection against a subsequent challenge with a severe strain of the same virus. During the 1990s, a natural phenomenon of immunity in plants was discovered: a sequence-specific RNA-based mechanism that protects plants from invading pathogens called **RNA interference or silencing**. Cross protection is the result of RNA silencing and in the case of PepMV, the symptoms in “vaccinated” tomato plants following infection with severe strains in the field can be greatly reduced (Aguero et al., 2018). Furthermore, V10, a natural crop protection product developed by Valto and distributed by Koppert Biological Systems, is used to prevent the emergence of PepMV.

JOURNAL OF AGRICULTURAL RESEARCH
Vol. 37 WASHINGTON, D. C., AUGUST 1, 1928 No. 3
HOSTS AND SYMPTOMS OF RING SPOT, A VIRUS DISEASE
OF PLANTS
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INTRODUCTION



Fig. 1.2

2A. Mutagenesis of a host virus susceptibility gene using the CRISPR/ Cas9 technology

Tomato (*Lycopersicon esculentum*) X Tomato brown rugose fruit virus (ToBRFV). ToBRFV is an rapidly-spreading virus that affects tomato plantations, where losses can reach 100% (Zhang et al., 2022; Fig. 2.1). ToBRFV is transmitted mainly *via* contaminated seeds, or mechanically through standard horticultural practices. First reported in the Middle East in 2015, multiple ToBRFV outbreaks have been reported throughout Europe in recent years (Fig. 2.2). ToBRFV can break down genetic resistance to tobamoviruses conferred by the R genes Tm-1, Tm-2, and Tm-22 in tomato and L1 and L2 alleles in pepper. Currently, no commercial ToBRFV-resistant tomato cultivars are available .

Mechanism of Action & Use

Ishikawa and co-workers (2022; Fig. 2.3) used the CRISPR/Cas9 technology to mutate four tomato homologues of TOBAMOVIRUS MULTIPLICATION1 (TOM1), an *Arabidopsis* gene, which is essential for tobamovirus multiplication, conferring resistance to ToBRFV in tomato plants.

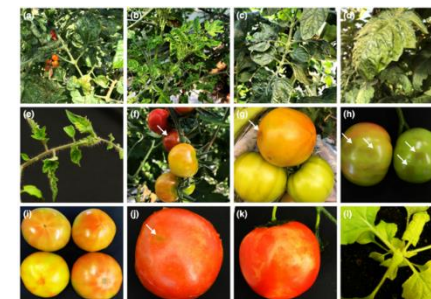


Fig. 2.1

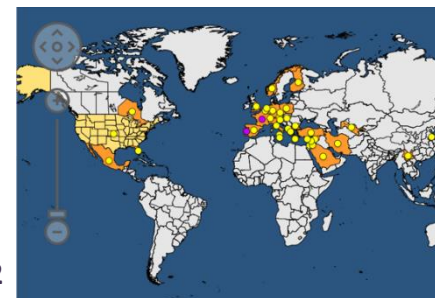


Fig. 2.2



Tomato brown rugose fruit virus resistance generated by quadruple knockout of homologs of TOBAMOVIRUS MULTIPLICATION1 in tomato

Masayuki Ishikawa ¹, Tetsuya Yoshida ¹, Momoko Matsuyama ¹, Yusuke Kouzai,² Akihito Kano ³ and Kazuhiro Ishibashi ^{1,4*}

Fig. 2.3

ch Article

2B. Mutagenesis of a host virus susceptibility gene using the CRISPR/ Cas9 technology

TABLE 3 Summary of studies that have employed CRISPR/Cas9 strategies for the targeting of host susceptibility genes

Plant species	Name of the susceptibility (S) gene targeted	Virus name	Reference
<i>Arabidopsis thaliana</i>	<i>AtelF(iso)4E</i>	Turnip mosaic virus (TMV)	Pyott et al. (2016)
	<i>eIF4E1</i>	Clover yellow vein virus (CYVV)	Bastet et al. (2019)
<i>Hordeum vulgare</i> (barley)	<i>eIF4E1</i>	Barley mild mosaic virus (BaMMV)	Hoffie et al. (2021)
<i>Manihot esculenta</i> (cassava)	<i>nCBP-1/2</i>	Cassava brown streak virus (CBSV)	Gomez et al. (2019)
<i>Cucumis sativus</i> (cucumber)	<i>CseIF4E</i>	Zucchini yellow mosaic virus (ZYMV) Cucumber vein yellowing virus (CVYV) Papaya ring spot mosaic virus-W (PRSV-W)	Chandrasekaran et al. (2016)
<i>Nicotiana benthamiana</i>	<i>CLC-Nb1a/b</i>	Potato virus Y (PVY)	Sun et al. (2018)
<i>Oryza sativa</i> (rice)	<i>OseIF4G</i>	Rice tungro spherical virus (RTSV)	Macovei et al. (2018)
<i>Solanum tuberosum</i> (potato)	<i>Coilin</i>	Potato virus Y (PVY)	Makhotenko et al. (2019)
<i>Glycine max</i> (soybean)	<i>GmF3H1/2, FNSII-1</i>	Soybean mosaic virus (SMV)	Zhang et al. (2020)
<i>Solanum lycopersicum</i> (tomato)	<i>TOM1</i>	Tomato brown rugose fruit virus (ToBRFV)	Ishikawa et al. (2022)
	<i>eIF4E1</i>	Pepper mottle virus (PepMoV)	Yoon et al. (2020)
	<i>eIF4E1</i>	Cucumber mosaic virus (CMV) Potato virus Y (PVY)	Atarashi et al. (2020)
	<i>eIF4E1</i>	Pepper veinal mottle virus (PVMV)	Kuroiwa et al. (2022)
	<i>SlEIF4E1, SlEIF4E2</i>	Potato virus Y (PVY)	Kumar et al. (2022)
<i>Triticum aestivum</i> (wheat)	<i>TaPDIL5-1</i>	Wheat yellow mosaic virus (WYMV)	Kan et al. (2022)

3. Insect pest monitoring - Camera-equipped traps

Coffee x Berry borer [*Hypothenemus hampei*]

Female coffee berry borers (CBB) lay eggs in coffee fruit, which their larvae then destroy (Fig. 3.1). Electronic traps can eliminate pests without the use of pesticides, and smart traps with IoT (Internet of Things) capabilities and computer vision can selectively target specific pests.



Fig. 3.1

Mechanism of Action & Use

The trap consists of three components: a) an embedded system with a camera, GPS sensor, and motor actuators, b) a database service provider, and c) a web application that displays data through a configurable heat map. When a beetle enters the trap, successive images are processed, and compared with standard CBB body characteristics. If identification is positive, the capture fan directs the CBB into a cage where it is imprisoned and destroyed (Fig.3.2-3; Figuredo et al., 2020).

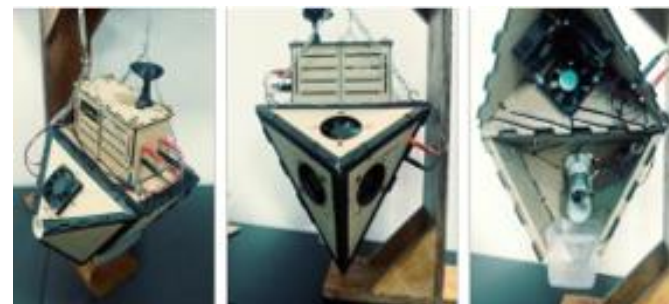


Fig. 3.2

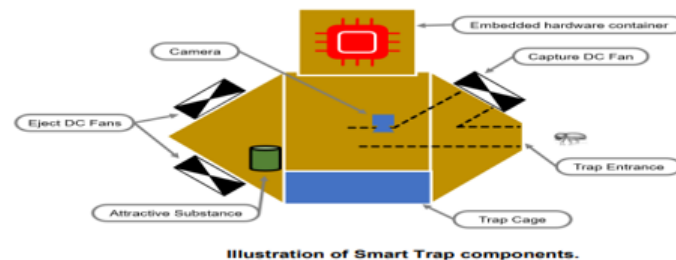


Illustration of Smart Trap components.

Fig. 3.3

4. *Beauveria bassiana* (white muscardine fungus) entomopathogenic fungus

Multiple crops x various insect pests: [*Cephus pygmeus*, *Helicoverpa armigera*, *Lobesia botrana*, *Popillia japonica*, *Spodoptera frugiperda*, thrips, aphids, whiteflies]

Beauveria bassiana is a fungus that grows naturally in soils throughout the world that parasitises various arthropod species causing white muscardine disease (Fig 4). It is used as a biological insecticide to control a number of pests, including termites, thrips, whiteflies, aphids and various beetles. In culture, *B. bassiana* grows as a white mould and produces many dry, powdery conidia. The spores are sprayed on affected crops as an emulsified suspension or wettable powder (*Wikipedia*).



Fig. 4

Mechanism of Action & Use:

Entomopathogenic fungi are a group of fungi living in soil that infect insects by penetrating their cuticle, eventually killing and feeding on them (Dara, 2017). After invading insect hosts, *B. bassiana* produces a variety of toxins (secondary metabolites) including beauvericin, bassianin, bassianolide, beauverolides, tenellin, oosporein, and oxalic acid, that facilitate the parasitization and death of the hosts (Wang et al., 2021).

5. Clay nanosheets for topical delivery of RNAi against plant viruses - Nanophytovirology

Tobacco, tomato X *Cucumber mosaic virus* (CMV)

Phytoviruses are highly destructive plant pathogens, causing significant agricultural losses due to their genomic diversity, rapid and dynamic evolution, and the general inadequacy of management options such as chemical means.

Mechanism of Action & Use

dsRNA is the triggering molecule of RNA silencing. Non-toxic, biodegradable, layered double hydroxide (LDH) clay nanosheets can be loaded with dsRNA. After spraying onto plants the LDH breaks down, and plant cells take up the dsRNA causing topical silencing of homologous RNA viruses (Mitter et al., 2017). A single spray with dsRNA-loaded LDH (BioClay) was shown to provide viral resistance for at least 20 days. The method seems promising means to protect against plant viruses (Fig. 5.1-2) and bacterial diseases (Fig. 5.3; Ren et al., 2022).

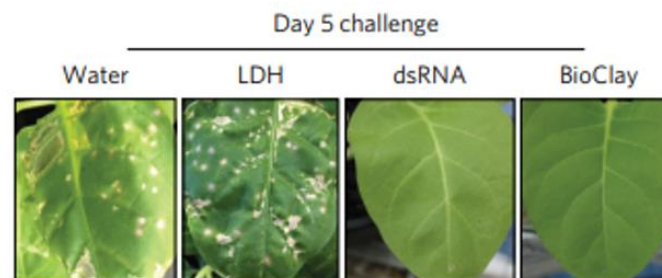


Fig. 5.1



Clay nanosheets for topical delivery of RNAi for sustained protection against plant viruses

Neena Mitter^{1*}, Elizabeth A. Worrall¹, Karl E. Robinson¹, Peng Li², Ritesh G. Jain¹, Christelle Taochy^{1,3}, Stephen J. Fletcher^{1,3}, Bernard J. Carroll³, G. Q. (Max) Lu^{2,4} and Zhi Ping Xu^{2*}

Fig. 5.2



Communication

Evaluation of the Abilities of Three Kinds of Copper-Based Nanoparticles to Control Kiwifruit Bacterial Canker

Ganggang Ren^{1,2}, Zhenghao Ding¹, Xin Pan², Guohai Wei², Peiyi Wang^{1,*} and Liwei Liu^{1,*}

Fig. 5.3

6. Plant growth-promoting *Rhizobacteria*-mediated induction of systemic resistance

Tobacco / various plant viruses [CMV, TYLCV, TSWV]

Plant growth-promoting rhizobacteria (PGPR) are diverse groups of plant-associated microorganisms, which can reduce the severity or incidence of disease by antagonism with bacteria and soil-borne pathogens, as well as by eliciting a systemic resistance defensive response in host plants (Meena et al., 2020).

Mechanism of Action & Use

Soil inoculation with *Paenibacillus lentimorbus* (B-30488) isolated from cow's milk increased plant vigour, while significantly decreasing (91%) *Cucumber mosaic virus* (CMV) RNA accumulation in systemically-infected tobacco leaves (Kumar et al., 2016; Fig. 6.1-2). In this study, defence-related enzyme production induced by CMV-infection was ameliorated in B-30488-treated plants, suggesting that systemic induced resistance mediated the against CMV.



Paenibacillus lentimorbus
strain (B-30488)

Fig. 6.1



Fig. 6.2

7A. Stacking of ecosystem services: mechanisms and interactions for optimal crop protection, pollination enhancement and productivity – EcoStack [EU-funded project] (Slide1/2)

Pest(s) X Host Range: mainly insect pests

Project aims & Method mechanism: EcoStack project will develop and support ecologically-, economically- and socially-sustainable crop production *via* stacking & protection of functional biodiversity (Fig. 7A.1-2).

More specifically:

- It will assess sustainable crop production needs based on functional biodiversity, using an interactive forum of stakeholders,
- It will evaluate and optimise the role of main off-crop habitats supplying ecosystem services for crop production,
- It will design and test in-crop interventions, which support the generation of ecosystem services (Hokkanen et al., 2017) within the crop, and which may carry over to the next crop in the rotation,
- It will develop, design & implement integrated systems for optimised provision of ecosystem services and use of plant protection tools, with focus on ecological, economic and social sustainability of integrated systems.



Fig. 7A.1



Fig. 7A.2

Photos by Rothamsted Research Limited (UK).

7B. Agroecosystem services: A review of concepts, indicators, assessment methods & research perspectives (slide 2/2)

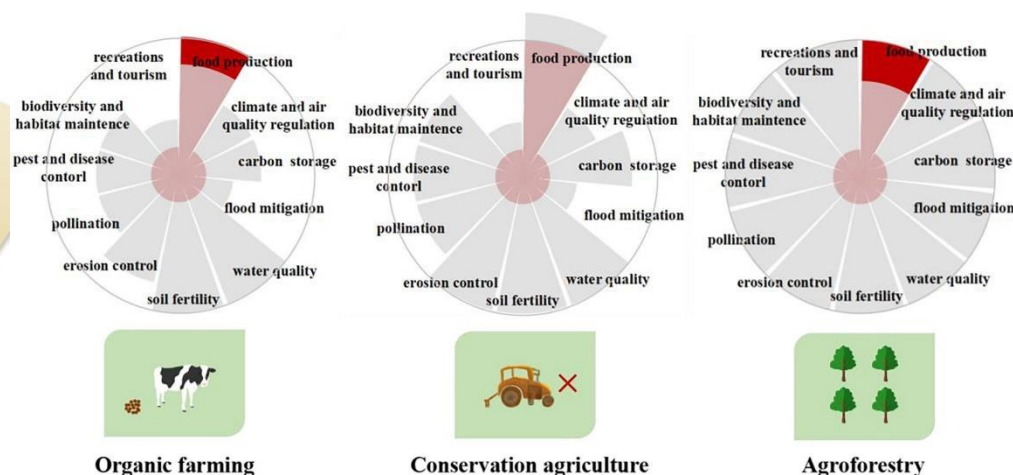
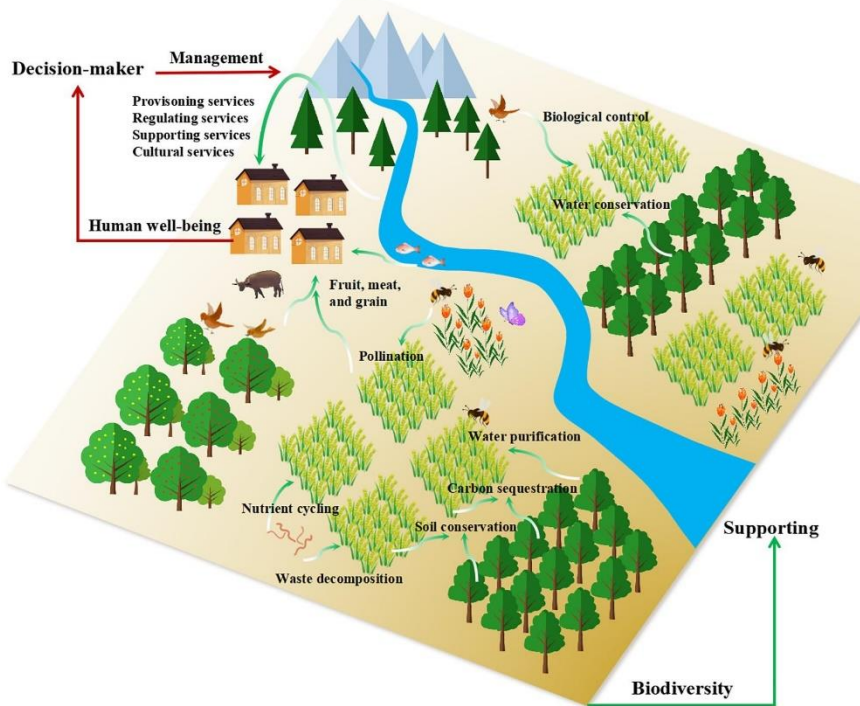


Fig. 7B.1. Ecosystem services within the agroecosystem. Biodiversity is the basis of agroecosystem, and provide many ecosystem services which are usually affected by social management (Liu et al., 2022).

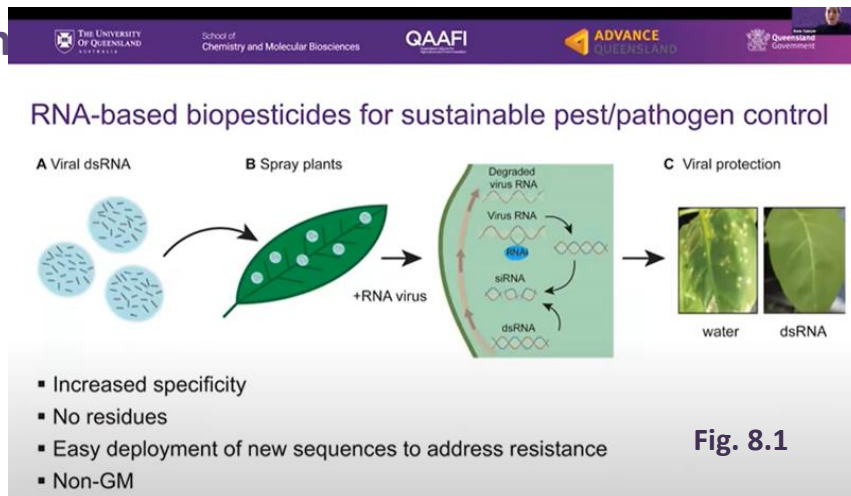
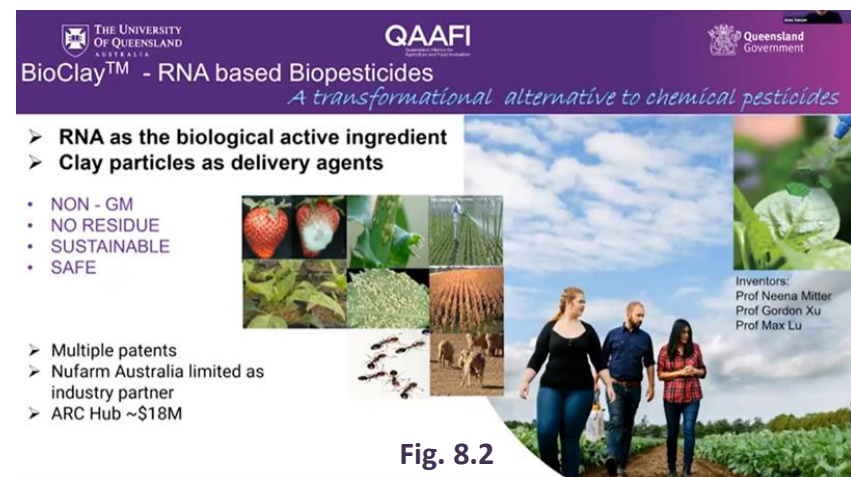
Fig. 7B.2. Comparison of conventional intensification (shown in red) and alternative farming approaches (shown in grey) for AES trade-offs (Liu et al., 2022).

8A. Exogenous RNAi for sustainable crop protection

Pest(s) X Host Range: potentially ALL

Introduction: Pioneering early work on the overexpression of the chalcone synthase gene (van der Krol et al., 1990) in petunia and *Tobacco etch virus* (Lindbo & Dougherty, 1993), led to the identification of the phenomenon present in many eukaryotic organisms, RNA-activated sequence-specific RNA degradation. The **Nobel Prize in Physiology/ Medicine 2006** was shared by A. Fire and C. Mello for the discovery that double-stranded (ds) RNA triggers suppression of gene activity in a homology-dependent manner, a process named RNA interference (RNAi) (Fire et al., 1998).

Mode of action: the accumulation of dsRNA in plant cells triggers RNAi through its recognition and cleavage into 21-24 nt small interfering (siRNAs) by an RNaseIII-like enzyme called DICER. siRNAs guide a nuclease complex referred to as the RNA-induced silencing complex (RISC) to homologous single-stranded (ss) mRNAs that are degraded. Researchers have learned how to trigger RNAi for specific genes, which can result in better disease and pest resistance (Fig. 8.1-2).

BioClay™ - RNA based Biopesticides
A transformational alternative to chemical pesticides

- RNA as the biological active ingredient
- Clay particles as delivery agents

- NON - GM
- NO RESIDUE
- SUSTAINABLE
- SAFE

- Multiple patents
- Nufarm Australia limited as industry partner
- ARC Hub ~\$18M

Inventors:
Prof Neena Mitter
Prof Gordon Xu
Prof Max Lu

Fig. 8.2

8B. Exogenous RNAi for sustainable crop protection (slide 2/2)

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nature plants | ARTICLES | <https://doi.org/10.1038/s41477-022-01152-8> | Check for updates

Foliar application of clay-delivered RNA interference for whitefly control

Ritesh G. Jain¹, Stephen J. Fletcher¹, Narelle Manzie¹, Karl E. Robinson¹, Peng Li¹, Elvin Lu¹, Christopher A. Brosnan¹, Zhi Ping Xu¹ and Neena Mitter¹

Detached leaf-mediated uptake of dsRNA

Fig. 8.3

The University of Queensland Australia | School of Chemistry and Molecular Biosciences | QAAFI | ADVANCE QUEENSLAND | Queensland Government

Can dsRNA be applied curatively and preventatively?

Treatment timepoints:

- 24 hours post-infection
- 6 days post-infection (first symptoms)
- 8 days post-infection (first pustules)
- 14-19 days post-infection (established infection)

Fig. 8.4

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Can we control fungal/oomycete diseases with exogenous RNAi?

<i>Botrytis cinerea</i> Grey mould	<i>Colletotrichum fructicola</i> anthracnose	<i>Austropuccinia psidii</i> myrtle rust	<i>Phytophthora cinnamomi</i> Phytophthora root rot	<i>Verticillium dahliae</i> Verticillium wilt

Fig. 8.5

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Testing different RNA application methods

Crown dips	Foliar sprays	Petiole soaking	Trunk injections

Fig. 8.6

9A. Gene Editing [CRISPR-Cas9 technology] in Crop

Protection

Pest(s) X Host Range: potentially ALL

Mode of action: CRISPR is a family of DNA sequences found in the genomes of prokaryotic organisms derived from DNA fragments of bacteriophages that had previously infected the prokaryote. Cas9 (or "CRISPR-associated protein 9") is an enzyme that uses CRISPR sequences as a guide to recognize and cleave specific strands of DNA that are complementary to the CRISPR sequence. CRISPR-Cas9 is a technology that can be used to edit genes within organisms (Jinek et al., 2012). This editing process has a wide variety of applications including basic biological research, development of biotechnological products, and treatment of diseases (Fig. 9.1-2; Karavolias et al., 2012).

The development of the CRISPR-Cas9 genome editing technique was recognized by the **Nobel Prize in Chemistry in 2020**, which was awarded to E. Charpentier & J. Doudna.

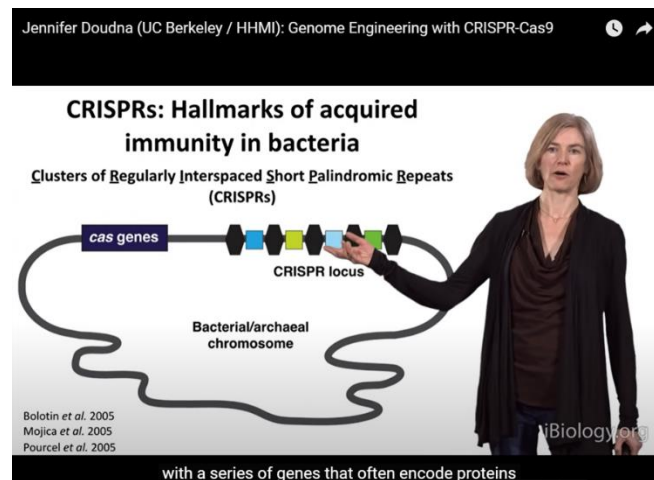


Fig. 9.1



Fig. 9.2

9B. Gene Editing [CRISPR-Cas9 technology] in Crop Protection (Slide 2/2)

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SCIENTIFIC REPORTS

OPEN

Rapid generation of a transgene-free powdery mildew resistant tomato by genome deletion

Vladimir Nekrasov^{1,4}, Congmao Wang², Joe Win¹, Christa Lanz³, Detlef Weigel¹ & Sophien Kamoun¹

Received: 16 February 2017
Accepted: 22 February 2017
Published online: 28 March 2017

Fig. 9.3

ARTICLES

<https://doi.org/10.1038/s41587-019-0267-z>

nature
biotechnology

OPEN

Broad-spectrum resistance to bacterial blight in rice using genome editing

Ricardo Oliva^{1,12*}, Chonghui Ji^{2,12}, Genelou Atienza-Grande^{1,10,12}, José C. Huguet-Tapia^{3,12}, Alvaro Perez-Quintero^{4,11,12}, Ting Li⁵, Joon-Seob Eom⁶, Chenhao Li², Hanna Nguyen¹, Bo Liu², Florence Auguy⁴, Coline Sciallano⁴, Van T. Luu⁶, Gerbert S. Dossa⁷, Sébastien Cunnac⁴, Sarah M. Schmidt⁶, Inez H. Slamet-Loedin¹, Casiana Vera Cruz¹, Boris Szurek⁴, Wolf B. Frommer^{6,8*}, Frank F. White³ and Bing Yang^{2,9*}

Fig. 9.4

Plant Biotechnology Journal

doi: 10.1111/tpb.12881

Establishing RNA virus resistance in plants by harnessing CRISPR immune system

Tong Zhang¹, Qiufeng Zheng¹, Xin Yi², Hong An³, Yaling Zhao¹, Siqi Ma¹ and Guohui Zhou^{1,*}

Fig. 9.5

nature plants

BRIEF COMMUNICATION

PUBLISHED: 28 SEPTEMBER 2015 | ARTICLE NUMBER: 15144 | DOI: 10.1038/NPLANTS.2015.144

Establishing a CRISPR-Cas-like immune system conferring DNA virus resistance in plants

Xiang Ji^{1,2†}, Huawei Zhang^{3†}, Yi Zhang^{1,2}, Yanpeng Wang^{1,2} and Caixia Gao^{1*}

Fig. 9.6

10. Cover crops are more effective than insecticides for managing pests

Insect pests X corn (*Zea mays*) – potentially ALL insect pests

Cover crops, planted to cover the soil rather than for harvest can regulate soil erosion, soil fertility, soil quality, water, weeds, pests, diseases, biodiversity and wildlife in an agroecosystem. Cover crops may be an off-season crop planted after harvesting of the cash crop, be planted between crop plants, or grow over winter.

Mode of action: Increasing scientific evidence indicates that no-till & cover crops support populations of resident arthropod predators and protect annual crops from insect pests. On the other hand, the use of neonicotinoid seed coatings, represents a common practice against early-season insect pests. The interaction between preventive pest management (PPM), integrated pest management (IPM) and the conservation practice of cover cropping was investigated by Rowen and co-workers (2022) in a 3-year

corn–soy (*Zea mays*-*Glycine max* L.) rotation, the response of invertebrate pests and predators to PPM and IPM, with and without a cover crop.

Results: PPM in year 1 decreased predation compared to a no-pest management control. Contrary to expectations, the IPM strategy requiring a single insecticide application, was more disruptive to the predator community than PPM, likely because the applied pyrethroid was more acutely toxic to a wider range of arthropods than neonicotinoids. Enhanced early-season cover was more effective at reducing pest density and damage than either intervention-based strategy. As part of a conservation-based approach to farming, cover crops can promote natural-enemy populations that can support effective biological control of insect pest populations.

11. Effect of kaolin clay on migrant green peach aphid in orchards

Peach [*Prunus persica* L.] X *Myzus persicae*

The green peach aphid, *Myzus persicae* (Hemiptera: *Aphididae*), is a serious pest in peach and nectarine orchards (Fig. 11.1). Direct feeding damage caused by heavy infestations early in the spring leads to leaf curling and severe disturbances in shoot growth. *Myzus persicae* is an efficient vector of the *Plum pox virus* that causes the notorious “sharka” disease.

Mechanism of Action & Use:

Surround® WP Kaolin Clay forms fine film coatings of microscopic mineral particles are sprayed onto plant surfaces (Fig. 11.2), acting as a protective barrier that either controls or suppresses pests, while at the same time many beneficial pests that do not feed on the plant surfaces generally remain unharmed. It protects fruit against direct sunburn and heat stress damage, promotes plant health, which leads to more efficient photosynthesis and higher yields under extreme light and heat growing conditions. Autumn applications might be an alternative to the insecticides commonly used in spring to control aphids in orchards. The use of kaolin clay to impede aphid egg laying in autumn reduces winter egg-laying by about 50%. It is not sufficient to control aphid colonies in the spring, but it could be used as part of a supervised control strategy, combined with the application of mineral oils in late winter.



Fig. 11.1



Fig. 11.2

Innovative & Environmentally-Friendly Methods in Crop Protection

12. Bio-Insecticide for the efficient control of *Spodoptera* species and other noctuid moths

Wide range of crops X *Spodoptera* spp., *Helicoverpa armigera*

Noctuid moths (Lepidoptera: *Noctuidae*) (Fig. 12.1) are polyphagous pests with a cosmopolitan distribution, damaging many economically important crops. They are widespread in distribution throughout Asia, Africa, Australia and the Mediterranean Europe. Noctuid moths have a high reproductive rate and cause heavy losses to crops. Larvae (Fig. 12.2) feed gregariously on plant leaves and later eat almost every plant part.



Fig. 12.1



Fig. 12.2

Mechanism of Action & Use:

Nomu-Protec is based on the insect-pathogenic fungus *Metarhizium rileyi* (previously known as *Nomuraea rileyi*) that infects and controls lepidopteran insect pests, especially those of the family *Noctuidae*. *Metarhizium rileyi* spores can either penetrate through the cuticle or enter the larvae through ingestion during feeding. Once inside the larvae, the fungus grows and multiplies, killing the larvae by internal tissue destruction. Between 2-4 days after the initial infection the larva stops feeding and dies 5-7 days later. Once the larva has died the fungus sporulates (Fig. 12.3), therefore having the ability to remain in the environment and re-infect the next generation of pests. Nomu-Protec also shows effective reduction in feeding damage shortly after infection. Recommended are 4 weekly applications of 300 g/ha and 600 g/ha, starting at first appearance of insect pest with good spray coverage and at higher



Fig. 12.3

13. Effect of silicon on two major insect pests of tomato

Tomato X *Tuta absoluta*, *Bemisia tabaci*

Tomato is attacked by several insect pest species, among which, the whitefly *Bemisia tabaci* Gennadius (Hemiptera: *Aleyrodidae*) (Fig. 13.1) and the tomato leaf miner *Tuta absoluta* Meyrick (Lepidoptera: *Gelechiidae*) (Fig. 13.2) are of greatest importance. Vast application of pesticides is harmful to the environment, human health and may increase the risk of pest resistance on insect populations. One of the promising strategies which are compatible with organic farming is application of silicon for enhancing plant vigor and resistance to pest damage on various agricultural crops.

Mechanism of Action & Use:

Silicon is known to enhance the crop resistance to biotic and abiotic stresses through physical and allelochemical mechanisms. AB Yellow[®] silicic acid formulation could be applied in two ways soil drench treatment or foliar spraying, with 2% Si concentration. The Silicon applications significantly decreased the population of immature of both whiteflies and tomato leaf miner on tomato crop in the greenhouse. Si-foliar spraying is more effective in reducing the population density of these key pests compared to Si-soil drench application.



Fig. 13.1



Fig. 13.2

14. Biological control of western flower thrips using the entomopathogenic fungus *Beauveria bassiana*

Vegetables, fruits, ornamentals X *Frankliniella occidentalis*

Western flower thrips, *Frankliniella occidentalis* (Fig. 14.1), is one of the most destructive pests of vegetables, fruits and ornamental crops worldwide, causing extensive damage by direct feeding of the crop and transmitting economically important viruses (Fig. 14.2).

Mechanism of Action & Use:

BotaniGard ES is a highly effective biological insecticide containing *Beauveria bassiana*, an entomopathogenic fungus that attacks a long-list of troublesome crop pests (not plants) like aphids, thrips, whitefly, spider mites, mealybugs, root aphids and more.

This naturally occurring mycoinsecticide works on contact and thorough coverage is required to achieve control. The applied spores attach to the insect, germinate and penetrate through the insect cuticle (skin). The fungus then grows rapidly within the insect, causing mortality in 7-10 days (Fig. 14.3). The product could be used in greenhouse, nursery, vegetables, etc. Effectiveness is NOT dependent on high relative humidity. The fungus controls ALL stages of the most troublesome crop pests.



Fig. 14.1



Fig. 14.2

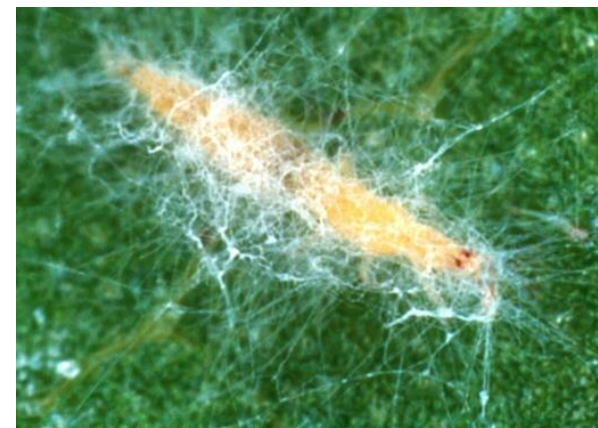


Fig. 14.3

15. Control of the tomato leaf miner with baculoviruses

Tomato (*Lycopersicon esculentum*) X *Tuta absoluta*

The tomato leafminer *Tuta absoluta* (Meyrick 1917) (Lepidoptera: *Gelechiidae*) is a devastating pest, causing losses of up to 100% (Fig. 15.1). Originating in South America, it has been spreading throughout the Mediterranean region but also continental Europe, the Middle East and Africa. The larvae of the pest (Fig. 15.2) mine into leaves and fruits, which can fast lead to complete crop loss. Many tomato leafminer populations are resistant to a wide array of pesticides, both chemical and biological. An innovative tool for its control is the use of the insect specific viruses of family *Baculoviridae*.

Mechanism of Action & Use:

The viral bio-insecticide Tutavir contains a *Phthorimaea operculella* granulovirus (PhopGV) for highly effective and selective control of the tomato leafminer. Once this natural pathogen is ingested by a suitable insect host, it reproduces within gut cells of the insect causing it to become ill and die. Due to its high compatibility with pollinators, beneficials and other inputs, Tutavir is the best candidate for integrated pest management programs. Because of its new and unique mode of action, Tutavir is an important tool for resistance management in conventional and biological production systems. Tutavir is applied at 100 ml/ha, 5 weekly applications. Assessment of the pest severity is made on 50 leaves per plot.



Fig. 15.1



Fig. 15.2

16. Mechanical control of peat fly larvae by sharp sand application

Greenhouse grown potted plants x Peat fly (*Bradysia* spp.)

The peat fly (Fig. 16.1) is a common pest whose larvae eat the hair roots of potted plants and seedlings grown in greenhouses (Fig. 16.2). Peat-based soil is a major source of the larvae. Quartz sharp sand (Fig. 16.3) placed on the surface of the growing medium results kills the majority of the larvae.

Mechanism of Action & Use:

Quartz sharp sand causes mechanical damage to the larvae as they move. The peat fly cannot reproduce in the presence of the quartz crystal material and cannot harm the seedlings, and thus is of benefits to many seedlings. (Fig. 16.4-5).

Mix 1 cubic metre of potting soil mixture with 10% quartz sand. The mixture is recommended for use for nursery seedlings in pots, but is not recommended for use in open fields.



Fig. 16.1



Fig. 16.2



Fig. 16.3



Fig. 16.4



Fig. 16.5

Innovative & Environmentally-Friendly Methods in Crop Protection

17. Use of nudibranch ash extract to deter Spanish naked snails

Multiple field cultivations x The Spanish nudibranch (*Arion vulgaris*)

Also known as the killer snail this nudibranch (Fig. 17.1) has recently appeared in vegetable and fruit farms in Romania. In addition to vegetation, it also eats other snails. The pest particularly affects vegetable and fruit farms (Fig. 17.2-3), but also eats other snails. An aqueous suspension of the ash made by burning the snails has been observed shown to is used to reduce *A. vulgaris* infestations.

Mechanism of Action & Use:

Some farmers have been using the locust control method of the early last century with success against slugs. According to Theresa von Beiersdorf, during locust epidemics they would collect the locust larvae, burn them alive and spread the ash on the crop by mixing it with water. The operation needs to be performed 1-2/year, on a regular basis. It does not completely eliminate the snails, but there is no invasion. Burn the snails on a beech wood fire, put about 200 g of ash (two handfuls) in 200 l of water and mix it (tradition says you have to mix it for an hour, until the solution becomes uniformly opalescent). This should be dispensed in the evening. Not harmful to plants. It is recommended to treat the whole area initially, then later only the edges of the area, where the snails may enter. (Fig. 17.4-5).



Fig. 17.1



Fig. 17.2



Fig. 17.3

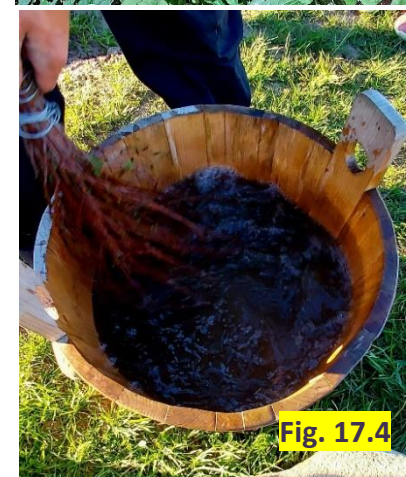


Fig. 17.4



Fig. 17.5

18. Control of aphids and thrips using a decoction of spindle berries & tansy

Thrips (order *Thysanoptera*), and aphids (*Aphidoidea spp*) (Fig. 18.1-2) are among the most common and destructive pests found in greenhouses, open fields and gardens. They weaken plants by sucking sap, and causing deformation (Fig. 18.3-5) and act as vectors for plant viruses. Aphids also leave deposits of honeydew which favour sooty mould growth. Aphids also leave deposits of honeydew which favour sooty mould growth. An aqueous decoction of spindle (*Euonymus europaeus*) berries (Fig. 18.6) and tansy (*Tanacetum vulgare*, Fig. 18.7) contains alkaloids and other active substances. When used to spray plants it can effectively reduce infestation levels.

Mechanism of Action & Use:

Preparation: add 50-60 gr of spindle berries and 100 gr of tansy in 5 litres of water, boil for 20 minutes, let stand for 12 hours, strain and dilute to 10 litres. You can put in 50 gr sugar as a glue and bait. It can be used for cabbage, peppers, tomatoes, potatoes where lice appear. Spray early in the morning or during the evening. This is poisonous for aphid pests. Does not scorch and can be used on very young plants. It is a universal aphid repellent, but very effective against thrips in peppers.



Fig. 18.1



Fig. 18.2



Fig. 18.3



Fig. 18.4



Fig. 18.5



Fig. 18.6



Fig. 18.7

19. Protection against mites using tobacco extracts

Multiple plants x *Acariforme* and *Parasitiforme* arachnids

Mites, small eight-legged arthropods of the orders *Acariforme* and *Parasitiforme*, (Fig. 19.1), identifiable with the use of a magnifying glass, are destructive plant pests that feed on the cellular fluids of plants, weaken them, reduce vigour and can ultimately cause plant death (Fig. 19.2). Aqueous tobacco extracts can effectively kill arachnid mites.

Mechanism of Action & Use:

Tobacco decoction, or tobacco steeped in water (Fig. 19.3) can be used against mites. Home-grown tobacco may be less harmful than commercially-available tobaccos that may cause damage (scorching) to the plants.

50 gr dried tobacco in 10 l of water (Fig. 19.4). After fermentation (approx. 5 days), the liquid is sprayed on plants, preferably at dawn or in the evening, avoiding direct sun exposure. Use on cucumbers, peppers, eggplant, celery. In summer, a tobacco ferment tobacco can be made as for nettle tea.



Fig. 19.2



Fig. 19.3



Fig. 19.4

Fig. 19.1

20. Crop rotation for crop protection in organic agriculture

The majority of open field crops, some greenhouse crops x multiple pests.

Crop rotation is the practice of planting/sowing different crops in succession on the same plot of land to improve soil health, optimize soil nutrients and control pests and weeds. A simple crop rotation may contain two or three plants, while a complex rotation may contain a dozen or more (Fig. 20.1). According to EU Regulation No 2092/91: "Pests, diseases and weeds shall be controlled by a combination of the following measures: - the application of an appropriate crop rotation system" Annual rotation limits the spread of pathogens and pests (root and stem diseases, nematodes, etc.), as well as the proliferation of weeds that are dominant in certain crops. It has long been observed that even two to three years of non-rotational cultivation increase disease susceptibility and insect damage. For this reason, monoculture production is practically unfeasible without a significant level of chemical plant protection (Fig. 20.2).

	Plant 1	Plant 2	Plant 3	Plant 4
First year	Beet	Spring barley or red clover	Red clover 2 years old	Winter wheat
Second year	Spring barley or red clover	Red clover 2 years old	Winter wheat	Beet
Third year	Red clover 2 years old	Winter wheat	Beet	Spring barley or red clover
Fourth year	Winter wheat	Beet	Spring barley or red clover	Red clover 2 years old
Fifth year = first year	Beet	Spring barley or red clover	Red clover 2 years old	Winter wheat

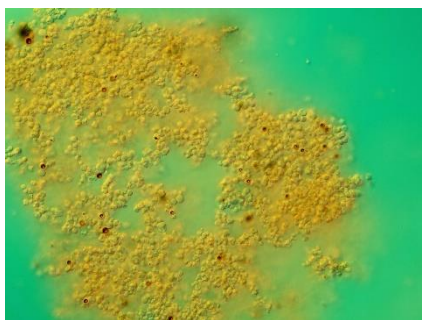
Fig. 20.1. An example of a multi-year open-field crop rotation scheme.



Fig. 20.2. The spread of sorghum (*Sorghum halepense*) can be effectively controlled by giving preference to crops within the rotation that can be harvested before the weed sets seed (e.g., kales, alfalfa, etc.).

21. Fungicidal plant protection product based on Thyme (*Thymus vulgaris*) essential oil

Field crops x *Fusarium culmorum*, *Blumeria graminis* & *Pyrenophora teres* on cereals



Mechanism of Action & Use:

The innovative aspect of this product is that the fungistatic agent *Thymus vulgaris* essential oil is incorporated into biopolymer microdroplets to increase its persistence in the crop. The product has been shown to significantly reduce fungal pathogen infestation and mycotoxin content in grain when applied at a rate of 200-400 l/ha using conventional sprayers. The product has been patented but is yet not on the market.

22. EDN - Ethandinitrile

Norway spruce trees (*Picea abies*) x *Ips typographus*, *Ips duplicatus*



Mechanism of Action & Use:

Ethandinitrile (EDN) is an insecticide highly effective against all developmental stages of bark beetles and other wood-boring insects. The product is prepared as a gas and supplied in 50 kg cylinders. The treatment of harvested timber takes place in forest landfills, where logs are sealed in polyethylene sheeting and fumigated for 10 hours. This product leaves no harmful residues. The product can be used under the exception of the so-called "Emergency conditions in plant protection".

23. Bluefume HCN

Garlic (*Allium sativum*) x *Ditylenchus dipsaci*, *Aceria tulipae*, *Fusarium sp.*



Mechanism of Action & Use:

Fumigation using the active ingredient cyanide (HCN) active against all developmental stages of the phytoparasitic nematodes *Ditylenchus dipsaci*, *Aceria tulipae* mites and *Fusarium spp.* fungi, which cause extensive damage to garlic seedlings. The treatment is carried out in specially-adapted shipping containers, equipped with a gas supply and measurement sensors. This product has been approved for the treatment of wood-boring insects in historical furniture, and is used worldwide to destroy insect pests in harvested bananas, but in the Czech Republic it is in the process of registration for use in garlic.

24. `Rubelit` apple trees resistant to apple scab caused by *Venturia inaequalis*

Apple trees x apple scab (*Venturia inaequalis*)

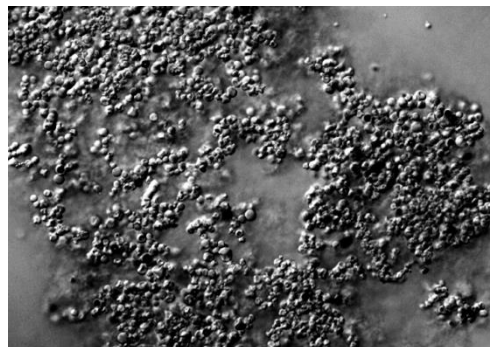
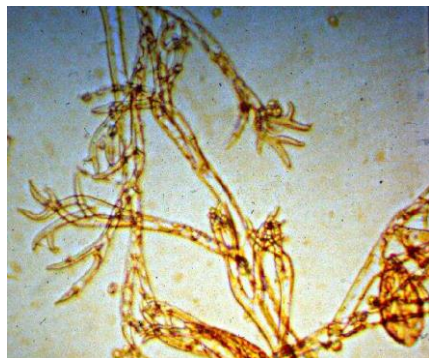


Mechanism of Action & Use:

This cultivar of the winter apple 'Rubelit', prefers drier, non-waterlogged soils that are light, fertile and neutral or slightly acidic. A sunny, warm site is recommended, ideally south-facing. Resistance is based on the apple Vf gene. The variety is certified by the Schweizerische Eidgenossenschaft and is sold freely on the market.

25. Protection against *Peronospora destructor* (onion blight) using the essential oil of *Pelargonium graveolens*

Onion X *Peronospora destructor*



Mechanism of Action & Use:

Onion blight is highly resistant to many plant protection products, but is susceptible to *Pelargonium graveolens* essential oil. The essential oil is diluted with rapeseed oil and formulated into bio-polymer microcapsules to reduce phytotoxicity. The active ingredient is EO from *Pelargonium graveolens* with the most dominant ingredient being citronellol. The fungicide is applied by spraying (200-400 l/ha). The product is protected by a utility model, and has not yet been approved.

26A. Neem oil against insects (Slide 1/2)

Multiple crops X small soft-bodied insects like aphids, mealybugs, mites, thrips & whiteflies



Mechanism of Action & Use:

One of the main components of neem tree (*Azadirachta indica*) seed oil (neem oil) is the chemical azadirachtin, but it also contains other active compounds. As an insecticide, neem oil works in two main ways: It serves as an anti-feedant when insects come in contact with or ingest it. Using a surfactant (spreader/sticker) when applying neem oil will increase spray coverage. Neem oil functions as a hormone disruptor and growth regulator of affected insects, preventing their normal development by blocking release of the hormones that trigger growth and maturation.

26B. Neem oil against insects (slide 2/2)

- It is not harmful to beneficial living organisms.
- It is a deep-acting preparation that is absorbed into the leaf blade and is thus able to act against pests with a hidden lifestyle and difficult to control, such as leaf-mining moths.
- Neem Azal can also be used in controlled organic farming!
- Complex mechanism of action
- It has excellent resistance breaking properties
- Residue-free protection



27A. PREV-GOLD, orange oil for multiple pest control (slide 1/2)

Multiple crops X powdery mildew, gray mould, spider mite species, moths and other insects with stinging and sucking mouthparts.



27B. PREV-GOLD, orange oil (Slide 2/2)

Mechanism of Action & Use (cont.):

PREV-GOLD® is a universal insecticide, fungicide and acaricide - all in one, based on a mixture of natural cold pressed orange oil 60g / l, which acts on many types of pests and diseases that usually require different control products.

PREV-GOLD® is a contact product with a physical mode of action that dries the cuticles of insects such as whiteflies, thrips, lice and mites, as well as cell walls or the phospholipid layer of fungal diseases. This is due to the lipophilic properties of orange oil, which has the ability to penetrate and destroy the protective layers of insects and external mycelium and sporangia of fungi, causing high mortality in pests and significantly reducing the development of pathogens.

The product does not cause resistance and is not phytotoxic. PREV-GOLD® is ideal for application in integrated production and integrated pest management (IPM) programs focused on reducing chemical residues on edible crops. It has little effect on beneficial organisms.

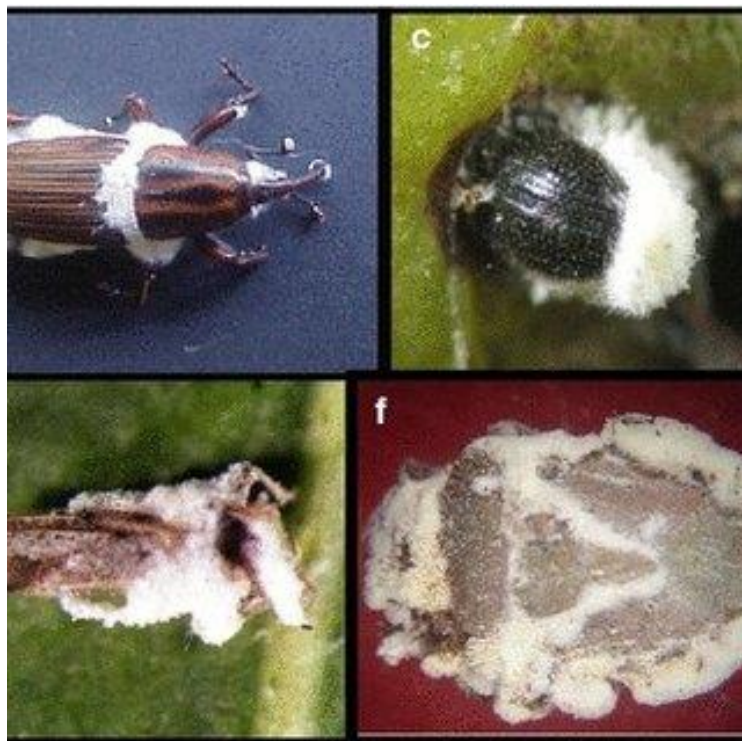
There are no residues in the production, which makes it an ideal choice for treatments just before harvest. It also does not require special storage conditions, is easy to use and has an immediate knock-down effect.



28A. Naturalis-L *Beauveria bassiana* insect parasite fungus

Multiple crops x Multiple invertebrate pathogens

Beauveria bassiana has assumed a key role in the management of numerous arthropod agricultural, veterinary and forestry pests.



Metarhizium anisopliae (57) v1 on mealworm: O. Coleoptera



Beauveria bassiana (35) v1 on mealworm: O. Coleoptera



Metarhizium anisopliae on cat flea: O. Siphonaptera



Metarhizium cf flavoviride (59) v1 on forest cockroach: O. Blattodea



Beauveria bassiana on termite: O. Isoptera



Beauveria bassiana on fruit fly: O. Diptera

28B. Naturalis-L *Beauveria bassiana* insect parasite fungus

Mechanism of Action & Use (cont.):

Naturalis-L is a bioinsecticide based on the entomopathogenic fungus *Beauveria bassiana* (strain ATCC 74040). Compared to many other strains of *Beauveria*, Naturalis-L infects a very wide range of economically damaging pests such as whitefly, mites, thrips and some groups of flies. In addition, the vegetable oil dispersion (OD) formulation means that Naturalis-L has a long shelf-life, is easy to use and gives excellent efficacy in real-world conditions.

The mode of action of Naturalis-L makes it a perfect tool for the control of pests and mites on vegetables, fruit crops and ornamentals. Naturalis-L can be successfully used in both organic production and integrated pest management programmes, especially if a reduction in residue levels and a number of traditional chemical sprays is desirable.

Naturalis-L does not leave any chemical residues and there is no harvest interval, so it can be applied through the entire life of the crop. As both an insecticide and miticide, Naturalis-L fits perfectly into pest management programmes aimed at minimizing the risk of resistance to conventional insecticides. Furthermore, Naturalis-L is compatible with beneficial insects and it is non-toxic to bees and pollinators.

29A. *Bacillus thuringiensis* ssp *kurstaki* against *Lepidoptera* (Slide 1/2)

Multiple crops X *Lepidoptera* caterpillars

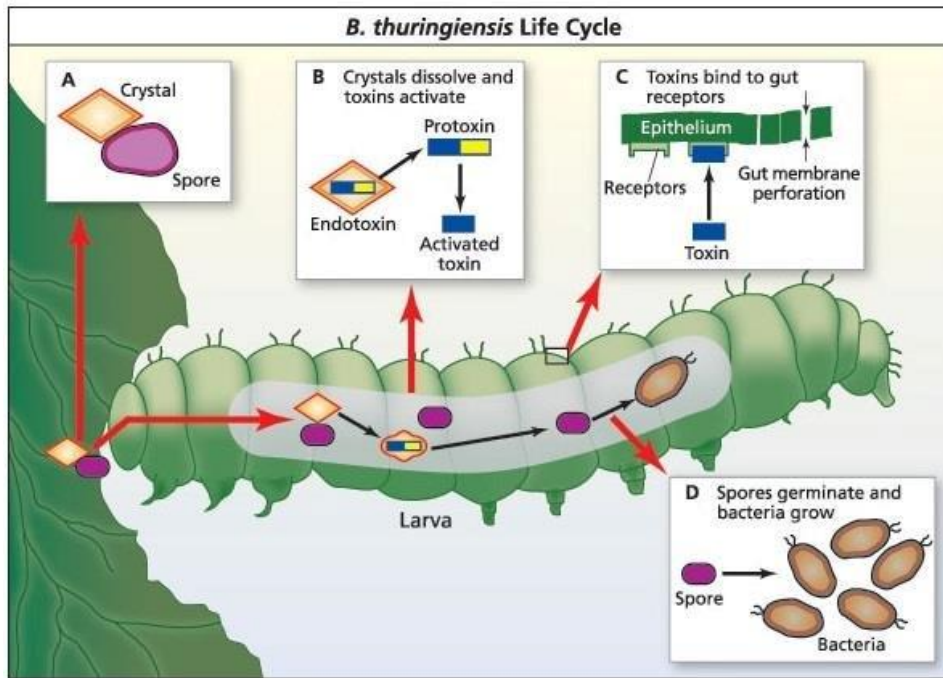


Fig. 29.1. uploaded by [Bruno Vinicius Daquila](#)
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Fig. 29.2. [Nigel Cattlin](#) / Alamy Stock Photo

29B. *Bacillus thuringiensis* ssp *kurstaki* against *Lepidoptera* (Slide 2/2)

Mechanism of Action & Use (cont.):

Bacillus thuringiensis kurstaki (Btk) is a gram-positive, rod-shaped bacterium native to the soil in a wide range of regions globally. A subspecies of *Bacillus thuringiensis*, Btk controls Lepidoptera. This order includes gypsy moths, cabbage loopers, tomato hornworms and grape leaf skeletonizers.

One of the many advantages to using Btk is that it does not pose a threat to other animals or insects outside of the order *Lepidoptera* in the environment once it has been sprayed or ingested by the target pest. Similar to *Bacillus thuringiensis israelensis*, birds and other predators can feed on the infected pests without ingesting toxic chemicals. As with most biological control measures, Btk applications will be most effective when made early in the pest's life cycle, particularly during the 1st and 2nd larval instars. Once ingested, the alkaline environment of the caterpillar's digestive system triggers the Btk bacterium to release a crystalline protein, a type of endotoxin, which paralyzes the caterpillar's digestive tract. The caterpillars will stop feeding and die shortly after this occurs.



30. Kaolinite to manage whitefly, *Ceratitis capitata*, oriental fruit fly

Multiple crops X whitefly, *Ceratitis capitata*, oriental fruit fly

Kaolinite is a clay mineral based on the aluminium silicate compound $AlSi_2O_5(OH)_4$. The active ingredient of Surround® WP is calcined kaolin, a biological insect repellent registered by the EPA, in a powder formulation. In order to be effective against insects, Surround® WP should be applied as a prevention and sprayed before the appearance of insects. Surround® WP reduces pest pressure and can delay or eliminate the need for conventional insecticide spraying. Adult individuals of the pest become heavily coated with kaolin particles within 24 hours of spraying. The insects are then occupied by trying to remove the particles from their bodies, and are unable to feed or lay eggs.

It forms a grayish coating on the surface of the leaf, so attention should be paid to the last application before harvesting.



Nigel Cattlin / Alamy Stock Photo

31. Use of Nemastar (*Steinernema carpocapsae*)

Multiple crops X Multiple insect pests

Infective juveniles of the threadworm *Steinernema carpocapsae* parasitize the juvenile forms of a large variety of insects, especially beetles, fleas, cutworms (*Agrotis* spp.), and moths. It is 100% safe to humans and pets, and primarily used for the control of beetles, fleas, cutworms and moths in soil. The juveniles of nemastar® are ambush predators, and are most effective against mobile prey. Once caught, they crawl inside the prey through breathing spiracles or other orifices, release a beneficial bacteria to break down the pests internal organs, and feeds on the bacterial slurry. The nematodes then breed in the cadaver, which eventually breaks apart releasing new generations of nematodes into the soil.



32. Mating disruption techniques against moths

Field crops X *Grapholita funebrana*, *Tortricidae* spp.

For the effective operation of air circulation, the size of the field cannot be smaller than 4 hectares and the field must be located in the same direction as the wind direction. Pheromones are species-specific, and individuals of different sexes do not find each other for mating. Moth monitoring requires weather data and pheromone traps. The dispensers are placed before the end of the biofix date determined on the basis of the heat amount. The pest control is successful, but the appearance of new *Tortricidea* species is expected.



33. Effect of Sudan grass (*Sorghum sudanese*) as a secondary crop.

Field crops X *Agrotis segetum*, *Elateridae* spp., *Delia brassicae*, *Phyllotreta atra*, thrips, *Tetranychus* spp.

According to decades of practice, the insect repellent Sudan grass is used in Hungarian agriculture as a pre-crop for vegetable cultivation. Sudanese grass has a high cyanide content when less than 60 cm in height. In this state, the root residues and foliage, when turned under and incorporated as green manure, ensure a high cyanide saturation in the root zone of vegetables for the next 2 years. Following the withdrawal of many conventional soil disinfectants, this method is a valuable cultivation technique to reduce the numbers and activity of soil-dwelling pests.



34. Secondary effect of fenugreek / *Trigonella foenum-graecum*

Field crops X *Agrotis segetum*, *Elateridae* spp., *Delia brassicae*, *Phyllotreta atra*, thrips, *Tetranychus* spp.

The soil disinfectant effect of the remains of the plant species fenugreek (family *Fabaceae*) farming can be reused in vegetable cultivation. The plant itself provides a good soil structure and air permeability, as a result of which the root system of the subsequent crop can be well infested by mycorrhizae, boosting root system development and increasing the vigour of the vegetables. The root and green parts of fenugreek have an insect repellent effect, which provides protection against pests in the soil and close to the soil for one or two years.



35. *Heterorhabditis bacteriophora* nematode against the vine weevil *Otiorhynchus ligustici*

Multiple crops X *Otiorhynchus ligustici*

The larvae (Fig. 35.1) of the vine weevil *Otiorhynchus ligustici* (Fig. 35.2) feed on the roots of legumes, hop, ornamental shrubs, pines and vine. As females lay several hundred eggs, the damage done may be devastating, on young plants in particular. Adults are nocturnal and feed on leaves, buds and flowers. The species has a 2-year lifecycle, overwintering in the larval and pupal stages in the first and the second winter, respectively.

Mechanism of Action & Use:

Heterorhabditis bacteriophora (Fig. 35.3) is a microscopic nematode. It has a bacterial symbiont *Photobacterium luminescens* which is spread by the nematode and converts the inside of soil dwelling insects into nutrients that both partners can utilise, killing the insects in the process in a matter of hours. The nematodes actively seek out their prey and breed after feeding. The cadaver of the prey eventually breaks apart and releases the new generation of nematodes (Fig. 35.4). The commercial products may be stored for several weeks between 4 and 10 °C. The recommended time of application is spring and autumn. The product should be mixed with water and applied as a spray on the soil surface. The soil should be kept wet for 3 weeks after the application.



Fig. 35.1



Fig. 35.2



Fig. 35.3



Fig. 35.4

36. Use of *Erwiphage* bacteriophage products against fireblight (*Erwinia amylovora*)

Apple, pear, quince X *Erwinia amylovora*

Fireblight, caused by the bacterium *Erwinia amylovora* causes severe damage in *Rosaceae* fruits such as apples, pears and quince. It first appeared in Europe in the 1950s. Infected blooms first appear water soaked, then shrivel and turn black (Fig. 36.1). The disease spreads to the spurs and eventually to the trunk, often evolving into canker. Infected fruits appear greyish, then dark brown; later they become mummified (Fig. 36.2).



Fig. 36.1



Fig. 36.2

Mechanism of Action & Use:

Erwiphage Forte (Fig. 36.3) was the first Hungarian pest control product using bacteriophages as a highly efficient preventive treatment against fireblight. A temporary permit for use is issued each year, is valid for 120 days in the blooming season. The solution includes a substance to protect the active ingredient against UV radiation and to promote adhesion of the bacteriophage. The product is stored at 3-8 °C. 3 treatments are recommended in the blossoming season. Erwiphage cannot be applied together with copper products.



Fig. 36.3

37. Use of the beetle *Delphastus catalinae* against the tobacco whitefly (*Bemisia tabaci*) in greenhouse grown vegetables

Greenhouse vegetable X Tobacco whitefly

The tobacco whitefly (Fig. 37.1) is an insect pest of a wide range of vegetable crops. Both adults and larvae suck on the green parts, weakening the plants. They also produce honeydew (Fig. 37.2). The saliva of the insect is toxic. It is a vector of more than a hundred plant viruses including the *Tomato Leaf Curl New Delhi* virus. It reproduces rapidly and is highly resistant to many insecticides.

Mechanism of Action & Use:

Delphastus catalinae is a predatory beetle, with both the adults and the larvae (Fig. 37.3-4) feeding on whiteflies. Not having a diapause, it may be used all year round and may be combined with parasitic wasps as it will avoid parasitised whitefly eggs. The product contains adult beetles; it cannot be stored for more than 1-2 days and must not be chilled. The beetles should be applied when the first whitefly colonies are discovered, ideally in the morning or evening, and should be repeated at least three times, once a week or until the pest is successfully managed. The minimum temperature for the beetle to be effective is 20°C.



Fig.37. 1



Fig. 37.2



Fig. 37.3



Fig. 37.4

38. Combined use of *Beauveria bassiana* and *Arthrobotrys oligospora* against cockchafer grubs

Field crops, vineyards, orchards X Cockchafers (*Melolonthinae*)

In Hungary, the cockchafer species with economic significance are the May beetle (*Melolontha melolontha*, Fig. 38.1A) and the Northern cockchafer (*Melolontha hippocastani*, Fig. 38.1B). Although swarms of adult beetles can be spectacular (Fig. 38.2), the real damage is done by the soil-dwelling grubs (Fig. 38.3). Depending on the species, the larvae spend 2-4 years in the soil, feeding on roots and causing severe damage in orchards, vineyards, field grown vegetables and lawns. As robust pesticides have been phased out, grubs have become once again an important issue.

Mechanism of Action & Use:

The entomopathogenic fungus *Beauveria bassiana*, responsible for white muscardine disease (Fig. 38.4), is a well-known biocontrol agent, that may be applied successfully against grubs, but in some cases it works too slowly. *Arthrobotrys oligospora* is a nematode-capturing fungus (Fig. 38.5), widely used for crop protection purposes. While it does not readily attack healthy grubs, it will penetrate them when they are infected by *Beauveria bassiana* and improve the efficiency of the latter. Both fungi may be used in spray format all year round; however, over 35°C they will die. They must not be combined with fungicides or herbicides. The treated area should be kept moist.

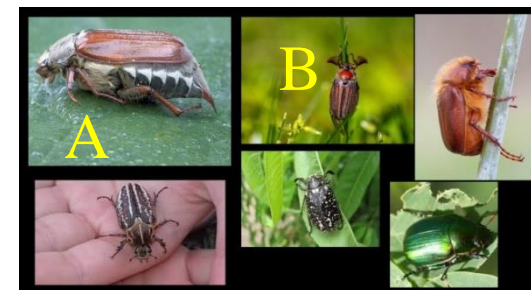


Fig. 38.1



Fig. 38.2



Fig. 38.3

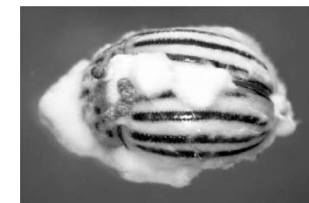


Fig. 38.4

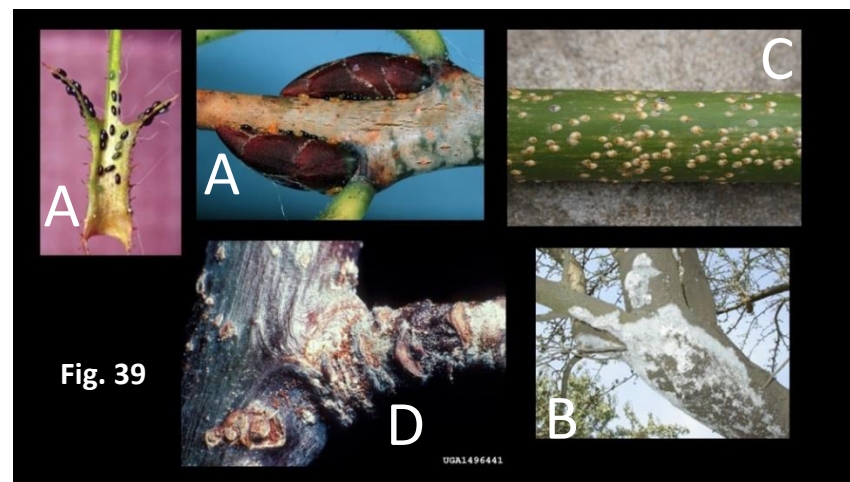


Fig. 38.5

39. Orange oil as dormant oil against the overwintering forms of insect pests

Tree crops X aphids, mealybugs, thrips, whiteflies etc.

Dormant oil treatment is a classic preventive management technique in orchards. Its application before the vegetative season reduces the populations of a wide range of insect pests such as aphids (Fig. 39A), mealybugs (B), thrips, whiteflies, leafhoppers, scales (C) and mites (D). Ecological farming requires the substitution of highly refined petroleum oils with less harmful alternatives.



Mechanism of Action & Use:

Orange oil is extracted from the rind of the sweet orange. When used as a crop protection agent, it melts the exoskeleton of small insects causing their dehydration. It also suffocates insect eggs and is effective against powdery mildews. As a humectant, it increases the efficiency of other crop protection products, being often combined with alcohol ethoxylate to increase its effect. As a dormant oil, orange oil may be used in higher than usual concentrations (e.g. 50 ml/10 l water) during dormant periods. At lower concentrations, combined with copper and/or sulphur products permitted in ecological farming, it may also be used at the beginning of leafing out. In this case, large volumes are needed to provide for a through “washing down” effect.

40. Diatomaceous earth to eliminate various bugs and pests

Multiple crops X Potentially ALL insects (especially those with an exoskeleton (e.g. ants or cockroaches))

What is:

Diatomaceous earth is fossilized algae dust that helps eliminate bugs by dehydrating them.

Mode of action:

Diatomaceous earth works as an insecticide in two ways: i) removes moisture from the habitat, making it inhospitable ii) when the diatomaceous earth makes direct contact with an insect's exoskeleton, it causes dehydration and may be fatal.

How to use:

- Apply to insect tunnels and pathways.
- Apply near the base of your houseplants for pest control.
- Keep diatomaceous earth away from high-traffic areas.

For garden use (Fig. 40) re-apply after rain. Can be sprinkled directly over plants.

Caution:

No discrimination between pollinators and unwanted insects.



Fig. 40

41. *Metarhizium*: jack of all trades, master of many

Potentially all crops X all insects

What it is:

Metarhizium is a genus of highly abundant fungi that grow naturally in the soil with several identities. They are best known for their ability to infect and kill many different arthropods, but most are also saprophytes, rhizosphere colonizers and beneficial root endophytes, with the ability to switch between these different lifestyles.

Mode of action:

These fungi are able to degrade, penetrate and assimilate the insect cuticle using a combination of cuticle-degrading enzymes and mechanical pressure (Fig. 41 A-E). Onward transmission of *Metarhizium* requires the death of the host as the insect cuticle is breached to release the conidial spores.

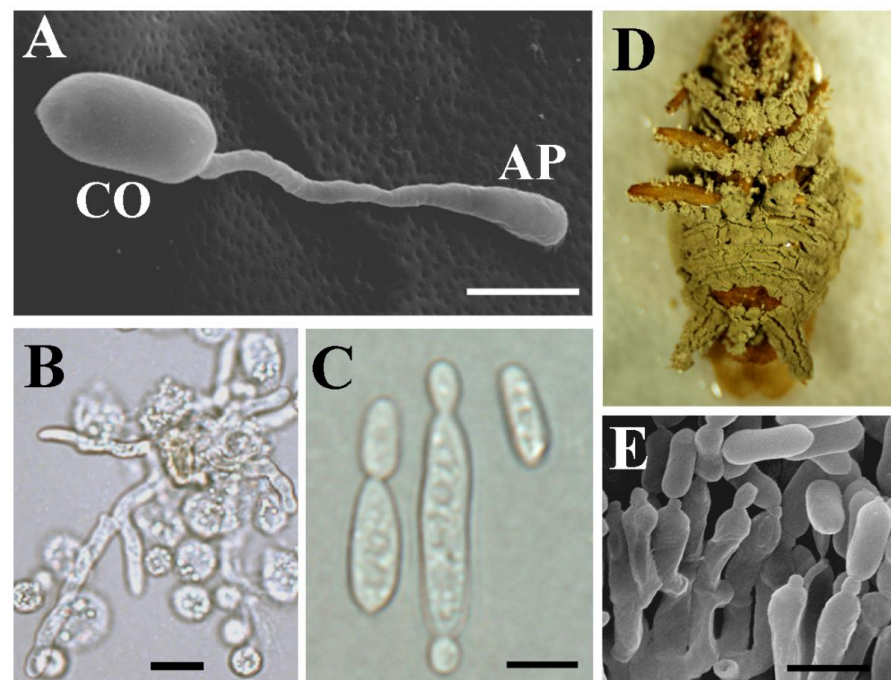


Fig. 41.

42. Natural Resistance Genes against Plant Viruses

Potentially ALL crops X all viruses

What it is:

Virus infections of crops are persistent and cannot yet be combated in the same way as many animal viruses, through provocation of an active immune response. The best strategy is one of avoidance by physical separation of the pathogen and host, or through the deployment of genetic resistance that prevents or limits the extent of the infection.

Mode of action:

To date, the majority of characterized pathogen resistance (R) genes from plants provide monogenic dominant resistance. Those characterized at the molecular level mostly confer resistance to fungal or bacterial pathogens, but there are currently 12 examples of such genes conferring resistance to viruses (Fig. 42).

Gene	Virus	avr*	Plant sp.	Reference(s)
<i>N</i>	Tobacco mosaic virus (TMV) (Tobamovirus)	Replicase/helicase	Tobacco	Whitham <i>et al.</i> (1994); Padgett <i>et al.</i> (1997); Erickson <i>et al.</i> (1999)
<i>Tm2²</i>	Tomato mosaic virus, TMV (Tobamoviruses)	Movement protein	Tomato	Lanfermeijer <i>et al.</i> (2003); Weber and Pfltzner (1998)
<i>Rx1</i>	Potato virus X (PVX) (Potexvirus)	Coat protein	Potato	Bendahmane <i>et al.</i> (1995, 1999)
<i>Rx2</i>	PVX (Potexvirus)	Coat protein	Potato	Bendahmane <i>et al.</i> (2000)
<i>Y-1</i>	Potato virus Y (Potyvirus)	—†	Potato	Vidal <i>et al.</i> (2002)
<i>Sw5</i>	Tomato spotted wilt virus (Tospovirus)	Movement protein	Tomato	Brommonschenkel <i>et al.</i> (2000)
<i>Rsv1</i>	Soybean mosaic virus (Potyvirus)	—	Soybean	Hayes <i>et al.</i> (2004)
<i>RT4-4</i>	Cucumber mosaic virus (CMV) (Cucumovirus)	2a gene	<i>Phaseolus vulgaris</i>	Seo <i>et al.</i> (2006)
<i>HRT</i>	Turnip crinkle virus (Carmovirus)	Coat protein	<i>A. thaliana</i>	Cooley <i>et al.</i> (2000); Ren <i>et al.</i> (2000)
<i>RTM1</i>	Tobacco etch virus (TEV) (Potyvirus)	—	<i>A. thaliana</i>	Chisholm <i>et al.</i> (2000)
<i>RTM2</i>	TEV	—	<i>A. thaliana</i>	Whitham <i>et al.</i> (2000)
<i>RCY1</i>	CMV	Coat protein	<i>A. thaliana</i>	Takahashi <i>et al.</i> (2001)

*Viral avirulence determinant.

†Unknown.

Fig. 42

43. Innovative diagnostic methods used to protect potatoes against potato late blight

Potato X *Phytophthora infestans*

Potato late blight produces symptoms on potato leaves (Fig. 43.1) and tubers (Fig. 43.2). Lower humidity and temperatures above 18°C cause *P. infestans* spores to germinate and infect neighbouring plants. Development of infection is most intensive at temperatures above 20°C, but also at increased humidity. Pathogen spores are spread by wind or rain up to several dozen kilometers (Fig. 43.3).



Fig. 43.1



Fig. 43.2



Fig. 43.3

Mechanism of Action & Use:

RT-PCR provides rapid diagnosis based on the amplification of genetic material using fluorescently-labeled probes. The intensity of the signal produced depends on amount of pathogen present. Response time is greatly reduced. RT-PCR is a diagnostic test that also allows observation of fungal disease development. This technique can be used in routine survey and control measures and is a useful tool to assist development of protection technologies for plants infected with fungal organisms.

44. Application of *Trichogramma* parasitic wasps in maize crops infected with the European corn borer

Maize (raspberry, pepper, hops etc.) X European corn borer (*Ostrinia nubilalis*)



Fig. 44.1



Fig. 44.2



Fig. 44.3

Mechanism of Action & Use:

The biological control of the European corn borer (Fig. 44.1) is through introduction of *Trichogramma* larvae. The female wasp lays eggs into the corn borer's eggs, where the larvae develop and pupate, feeding on the host embryos and destroying them for 8–15 days until the next generation of adult wasps emerge. *Trichogramma* are usually introduced into cultivation using hangers containing larvae and pupae (Fig. 44.2) or by aerial application for larger areas (Fig. 44.3).

45. Use of *Ampelomyces quisqualis* to combat gooseberry mildew

Multiple crops X Powdery mildews (family *Erysiphaceae*)

Powdery mildew (Fig.45.1) is a fungal disease whose characteristic symptom is mould on leaves and fruit, which is initially white, then brown. It affects hundreds of plant species.



Fig. 45.1



Fig. 45.2



Fig. 45.3

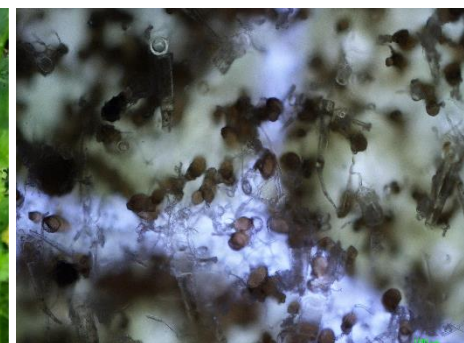


Fig. 45.4

Mechanism of Action & Use:

The hyphae of the fungus *A. quisqualis* penetrate inside the hyphae of the phytopathogenic mycelium and develop there (Fig. 45.4) regardless of external conditions. This parasitization inhibits the development of the mycelia of various powdery mildew species. Application is by foliar spray (Fig.45.2) or directly to the soil (Fig.45.3) as a solution. Commercial preparations of *A. quisqualis* have been certified for use in Italy and Germany.

46. Application of *Trichoderma harzianum* against fungal pathogens

Multiple crops X *Fusarium*, *Sclerotinia*, *Phytophthora*, *Rhizoctonia*, *Cylindrocladium*, *Pythium*



Fig. 46.1



Fig. 46.2



Fig. 46.3



Fig. 46.4

Mechanism of Action & Use:

Trichoderma spp. fungi inhabit the root zone of plants, competing with pathogens for nutrients and living space. They produce metabolites that act antagonistically against a number of pathogens, including *Fusarium*, *Rhizoctonia*, *Sclerotinia* (Fig. 46.1), *Phytophthora* (Fig. 46.2), *Cylindrocladium*, and *Pythium*, and also reduce the occurrence of bacterial diseases that are difficult to combat. Fungi of the genus *Trichoderma* (Fig. 46.3) stimulate growth and induce immune mechanisms in plants. Biopreparations based on *Trichoderma spp.* Are useful in integrated plant protection protocols. They can be applied in several ways: by adding them to the substrate (Fig 46.4), by mixing with seeds (dressing) or when watering or spraying plants. The preparations may be in the form of granules or powders that can be combined with fertilizers and pesticides, but not with fungicides. There are preparations recommended for seed dressing and in seedling production. Others can be used for perennial plants. Among the species used in commercial preparations are *Trichoderma asperellum*, and *Trichoderma harzianum* T-22.

47. Control of *Trialeurodes vaporariorum* in cabbage cultivation using marigolds or limonene Cabbage (and other crops) X Whitefly (*Trialeurodes vaporariorum*)



Fig. 47.1



Fig. 47.2



Fig. 47.3

Mechanism of Action & Use:

Marigold (*Tagetes erecta*) (Fig. 47.2) produces active volatiles including benzaldehyde, linalool, myroxide, piperitone, limonene, ocimene, lagetone, and valeric acid that attract natural enemies of plant pests. Intercropping marigolds with crops provides an environmentally-friendly strategy to reduce pest populations. Volatile limonene alone has also been found to be effective in repelling whitefly (Fig. 47.1) from target crops and was shown to increase yield by 32% during a heavy infestation. Limonene dispensers are extremely effective at repelling whiteflies and offer a low cost and easily-implemented control option, and direct spraying is also commonly used (Fig. 47.3).

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