FOOD SECURITY
BIOSECURITY
&TRADE
THE ROLE OF
PLANT HEALTH

St Hugh's College, Oxford
12 – 13 September 2016

BSPP

Food Security, Biosecurity and Trade
The role of plant health

St Hugh's College
Oxford
UK

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President's Welcome

I would very much like to welcome you to the 2016 BSPP Presidential meeting at St Hugh's College, Oxford. The theme of this year's meeting considers the impact of plant pathology on issues of food security, biosecurity and trade; challenges of global significance. We have invited a number of international speakers and attendees to present and participate.

Crop losses due to plant disease play a significant role in reducing global food production and associated food security. Restricting the spread of plant diseases between countries and between continents is also an important part of decreasing disease impact. Consequently, improved biosecurity planning is needed in many sectors and countries to reduce the risk of introduction. Trade is one pathway that can facilitate introduction but is vital to the economy of countries and can benefit the individual; increased yields allow producers to trade their surpluses and improve their livelihoods.

The aim for this year's conference is to have a number of general overview papers outlining significant aspects associated with the meeting's themes as well as presentations that illustrate the above challenges with specific technical and scientific examples.

In addition to the main conference, BSPP are organizing innovative outreach activities with Oxfordshire schools. The society is proudly leading the way in increased engagement with schools and the general public to raise the profile of the importance of plant pathology. This is especially important given that plant diseases are now back as part of the national curriculum for schools

BSPP are also proud to be sponsoring the "Grand Challenges in Plant Pathology", another innovation being organized by the University of Oxford in the same week as the Presidential meeting, which aims to bring together a group of outstanding graduate researchers who have the motivation, creativity and skills needed to tackle the most important challenges in plant disease research. Attendees will be challenged to work together in collaborative, interdisciplinary teams to propose novel solutions to real problems posed by industry and non-academic organizations.

We sincerely hope you enjoy the meeting, and the various other activities associated with the meeting. We also hope that you will visit the beautiful, historic city of Oxford during your stay.

The Organizing Committee
Julie Flood, BSPP President 2016
Elizabeth Orton, BSPP Programme Secretary

The BSPP is grateful for support from CABI, the American Phytopathological Society and the Canadian Phytopathological Society. Special thanks are due to all that have been involved in the organization but, in particular, Julie Ellwood, Diane Hird, Sarah Holdgate, Carol Jenner, Will Kay and Elizabeth Orton.

Monday 12 September

Registration

PRESIDENT'S WELCOME

8.45-9.15 Julie Flood.

FOOD SECURITY

Chair Matt Dickinson

9.15-9.45 Sarah Gurr. Fungi challenge global food security

9.45-10.15 Fenton Beed. Food security and plant health

10.15-10.45 Sajid Ali. Protecting global wheat production against worldwide spreading yellow rust pathogen strains and populations

10.45-11.15 Coffee

Chair Rob Jackson

11.15-11.35 Yaima Arocha Rosete. Contributions to plant health by improving diagnosis and management of the coconut lethal yellowing phytoplasma in Côte d'Ivoire

11.35-11.55 Paul Nicholson. Fusarium mycotoxins in cereals: combating a hidden menace

11.55-12.15 Nigel Taylor. RNAi technology imparts high level resistance to cassava brown streak disease across diverse geographical locations and the vegetative cropping cycle

12.15-12.35 Sally Mallowa. Tools of pedagogy and global partnership contribute to war on cassava viruses

12.35-1.30 Lunch

1.30-3.30 P H Gregory Prize competition

Chair Richard Oliver

Kwasi Adusei-Fosu. Detection of possible pathogenicity-associated genes in *Fusarium oxysporum* f. sp. *elaiedis*

James Elderfield. Modelling fungicide resistance management strategies: mixtures and alternation

Oliver Ellingham. Increasing accuracy of powdery mildews (Ascomycota, Erysiphales) identification using previously untapped DNA regions

William Kay. Off-host spore survival in *Zymoseptoria tritici* suggests a route for the maintenance of genetic variability and pathogenicity between crop cycles

Bo Liu. The beneficial effects of silicon on strawberry plants including reduced susceptibility to strawberry powdery mildew

Deepa Paliwal. Aphid-killing bacteria: how do they work?

Douglas Pyott. Engineering of CRISPR/Cas9-mediated virus resistance in transgene-free *Arabidopsis* plants

Anna Tiley. Investigating asexual sporulation in *Zymoseptoria tritici*, a pathogen of wheat

Huan Wang. The role of the Entner-Douderoff metabolic pathway during plant infection by *Pectobacterium* species

3.30-4.00 Coffee

BIOSECURITY

Chair Clive Brasier

4.00-4.30 Nicola Spence. UK Plant Security: Protecting plants and trees using science

4.30-5.00 Gerard Clover. Managing biosecurity risks in New Zealand: relevance to the UK?

5.00-7.00 Poster session and drinks

6.00-7.00 BSPP Annual General Meeting

7.15 for 7.30 Conference dinner

Tuesday 13 September

BIOSECURITY (continued)

Chair Dawn Arnold

9.00-9.20 Paul Beales. The role of the Plant Health and Seeds Inspectorate in plant biosecurity

9.20-9.40 Celia Knight. The Plant Health Professional Register

9.40-10.00 Alexander Mastin. Early detection surveillance for emerging vector-borne pathogens

10.00-10.20 Roy Macarthur. The importance of an agreed seed sampling plan for regulated seed-borne pests and pathogens, to facilitate the international movement of seed

10.20-10.50 Coffee

Chair Murray Grant

10.50-11.10 Rachel Warmington. Biosecurity at the Eden Project – considering local and global threats

11.10-11.30 Gerry Sadler. Scottish Plant Health Service – role in safeguarding food security, biosecurity and trade

11.30-11.50 Guillaume Bilodeau. Biomonitoring of plant pathogens in Canada – technologies and genomic approaches

TRADE

Chair Gerard Clover

11.50-12.20 Clive Brasier. Global trade and increasing forest and environmental bio-insecurity

12.20-12.40 Elizabeth Orton. Population structure of the ash dieback pathogen *Hymenoscyphus fraxineus* in the UK

12.40-1.00 Finn McQuaid. Trade restrictions and control strategies: a modelling case study

Lunch 1.00-2.00

Chair Julie Flood

2.00-2.30 Kenza Le Mentec. International trade in plants and food security

2.30-2.50 Maizatul Suriza Mohamed. Understanding and monitoring *Phytophthora palmivora,* the causal agent of bud rot disease of oil palm

2.50-3.10 Andy Wetten. Cacao swollen shoot virus – quarantine measures and vector biology

3.10-3.30 Jennifer Sjölund. Overcoming challenges in the detection and diagnostics of psyllid vectors of *Candidatus* Liberibacter solanacearum

3.30 Closing remarks

Refreshments and departure

Awards

The Board of BSPP wishes to encourage young plant pathologists to talk about their work. There are two prizes awarded at the presidential meeting:

The P H Gregory prize, awarded for the presentation of an oral paper. The competition is open to (a) members of BSPP who have not previously presented a paper to a meeting of a learned society; and (b) all registered postgraduate students, whether or not they are members of the society, or have presented a paper before to a meeting of a learned society. Contestants should not have entered the P H Gregory competition previously. The winner receives a certificate and a cheque for £250.

Philip Gregory (1907–1986) pioneered aerobiology as a topic for research, combining many disciplines to contribute to better understanding of fungal spore dispersal and plant disease epidemiology. He developed theories of spore dispersal, which was published in his classic paper on the dispersion of airborne spores (Gregory PH. (1945) Trans. Br. Mycol. Soc. 28: 26–72). He became Head of the Plant Pathology Department at Rothamsted in 1958 where he further developed his research on spore dispersal and sedimentation velocities. In his retirement, Philip Gregory continued work on elucidating the epidemiology of black pod disease of cocoa in Nigeria, and maintained enthusiasm for his wide range of scientific interests. He proudly and enthusiastically showed his garden to visitors and they often regarded this as the highlight of their visit to Harpenden (Source: Lacey *et al*. (1997) Ann. Rev. Phytopathol. 35: 1—14).

The John Colhoun Prize, awarded for a poster. The work presented in the poster must form part of a research project conducted by the entrant in support of a PhD or Masters degree, and the entrant must not have been awarded the degree prior to the deadline for abstracts date. Students need not be members of BSPP. The winner receives a certificate and a cheque for £100.

John Colhoun (1913–2002): cryptogamist and plant pathologist. John Colhoun was awarded a MAgr in 1937 at Queens University, Belfast, and then moved to Imperial College, London, working on fungal pathogens of apples for his PhD, awarded in 1940. He returned to Northern Ireland where he worked on flax, an important crop in the province during World War II, leading to a definitive text (Muskett A. E. & Colhoun J. (1947): The Diseases of the Flax Plant). He became reader at Queen's University in 1954. Subsequently, he took up the Chair of Cryptogamic Botany at the University of Manchester in 1960, where he worked on *Fusaria*, *Phytophthora*, *Septoria* and *Phoma*, with hosts ranging from cereals to chrysanthemum, yam, oil palm, and banana. In 1968 he was elected Chairman of the Federation of British Plant Pathologists, forerunner of the British Society for Plant Pathology. He retired from Manchester University in 1980 as Professor Emeritus, having occupied the Barker Chair of Cryptogamic Botany for 20 years. (Source: Epton H. (2003) Mycol. Res. 107: 377–381).

TALKS

Fungi challenge global food security

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Over the past centuries, crop diseases have led to the starvation of the people, the ruination of economies and the downfall of governments. Of the various challenges, the threat to plants of fungal infection outstrips that posed by bacterial and viral diseases combined. Indeed, fungal diseases have been increasing in severity and scale since the mid-20th Century and now pose a serious threat to global food security and ecosystem health.

We face a future blighted by known adversaries, by new variants of old foes and by new diseases. Modern agricultural intensification practices have heightened the challenge – the planting of vast swathes of genetically uniform crops, guarded by one or two inbred resistance genes, and use of single target site antifungals has hastened emergence of new virulent and fungicide-resistant strains. Climate change compounds the saga as we see altered disease demographics – pathogens are on the move poleward in a warming world.

This presentation will highlight some current notable and persistent fungal diseases. It will highlight the evolutionary drivers underpinning emergence of new diseases and allude to the accelerators of spread. I will set these points in the context of recent disease modelling, which shows the global distributions of crop pathogens and their predicted movement and will discuss the concept of crop disease saturation.

I shall conclude with some thoughts on future threats and challenges, on fungal disease mitigation and of ways of enhancing global food security.

Food security and plant health

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Increasing global demand for food requires increased efficiencies in crop production, to avoid encroachment into lands that confer vital services to the survival of our world. A significant mechanism to achieve this is through improved management of pests and diseases. Critical pathogens of globally important staple crops and strategies to mitigate their introduction, establishment and spread will be presented. Solutions include: fit-for-purpose diagnostics, as the first decisive step towards deploying appropriate control interventions; regional surveillance; targeted epidemiology studies; regional pest risk analyses and through networks to facilitate synergy and coordination. Sustainable and integrated disease management practices will be highlighted including crop resistance, agronomic practices and biological control while considering influence of increased globalization, human mobility, trade, climate change, and evolution of pathogens and vectors. The globe's population is currently challenged with greater rates of malnutrition compared to hunger so attention will be paid to the management of vegetable diseases in order to confer nutritional security. This is because common and especially traditional vegetables offer unrivalled profiles of health promoting minerals and vitamins. Further, vegetables generate significant incomes for smallholder farmers from relatively diminutive rural and urban plots. However, for vegetable value webs to be sustained a myriad of pests and diseases must be identified and curtailed through cost-efficient and practical interventions that are durable across contrasting sites and seasons. Similarly food safety must be ensured through judicious pesticide use and implementation of hygiene standards to eliminate human pathogens.

Protecting global wheat production against worldwide spreading yellow rust pathogen strains and populations

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Global wheat production faces a major constraint from rust diseases owing the rapid overcome of resistance genes. Microsatellite genotyping and subsequent population genetic analyses were carried out to identify the centre of diversity of wheat yellow rust pathogen *Puccinia striiformis* and decipher its population structure and migration patterns. At least six genetic clusters corresponding to their likely geographical origin were identified with a predominant clonality and low diversity in most parts of the world in contrast with the Himalayan and near-Himalayan region. Temporal analyses of the Himalayan populations, a high sexual reproduction capacity and sequencing of rust-infected *Berberis* spp. (the alternate host) suggested the region as the centre of origin of the pathogen with potential role of sexual reproduction in generation of novel, aggressive and potentially invasive races/strains. The worldwide spread of the pathogen was deciphered and the sources of recent invasive strains/populations were identified. The Himalayan and near-Himalayan populations were identified as the most ancestral populations and the source of recent invasive races of Europe, "Warrior" and "Kranich"; the Middle East-East Africa as the source of *PstS1/PstS2*; Europe as the source of South American, North American and Australian populations; and Mediterranean-Central Asian populations as the origin of South African populations. The role of both human activities and air dispersal in the worldwide spread of *P. striiformis* from its plausible centre of origin will be addressed in relation to exploitation of available wheat diversity for the durable resistance genes development and deployment at different geographical scales.

Contributions to plant health by improving diagnosis and management of the coconut lethal yellowing phytoplasma **in Côte d'Ivoire**

Yaima Arocha Rosete[1], C Dietrich[2], H Atta Diallo[3], J-L Konan Konan[4], E Kwadjo Koffi[3], P Kouamé Assiri[3], S Koutoua[3], D Kra Kouamé[3], M N Toualy[3], M-P Daramcoum[4], N Isabelle Beugré[4], W Miyasi Ouattara[3], N D Fursy-Rodelec[3], O Noufou Doudjo[5], C Ghislaine Kouadjo[4], A Kouassi[4], N Yankey[6], S Dery[6], A Maharaj[1], M Saleh[1], R Summerbell[1], N Contaldo[7], S Paltrinieri[7], A Bertaccini[7], P Wang[8], J Copeland[8], K Yoshioka[8], W Moeder[8], D Guttman[8] and J Scott[8]

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Côte d'Ivoire lethal yellowing (CILY) phytoplasma is severely impacting the smallholder farmers in Grand-Lahou, Côte d'Ivoire. Specific diagnostics and agroecological approaches are required to prevent disease spread and help farmers to improve their livelihoods. CILY-affected villages were surveyed for the CILY phytoplasma. Plant and hemipteran samples were collected from coconut palms and wild plants growing in the coconut farms. Total DNA extracted from plant and insect tissues were assessed by PCR/RFLP and sequencing of the phytoplasma 16S rRNA, the translocation protein (*sec*A) and ribosomal protein genes. Illumina-based sequencing was used to study the bacterial and fungal microbiome populations from CILY phytoplasma-free and -infected coconut palms. RFLP and SNPs on their *sec*A and ribosomal protein gene sequences distinguished the CILY phytoplasma from the Cape St Paul Wilt Disease and the Mozambican LY strains. Wild plant species from families Poaceae, Verbenaceae, Plantaginaceae, Phyllanthaceae and Cyperaceae were identified as alternative reservoirs for the CILY phytoplasma. A typhlocybine species was confirmed as a potential vector for the CILY phytoplasma. Bacterial and fungal microbiome profiles identified endophytes species to be tested as future potential biocontrols. Findings contributed to develop a more effective management strategy for CILY that has been disseminated to coconut farmers and villagers of Grand-Lahou. This work was carried out with the aid of a grant from the International Development Research Centre (IDRC), Ottawa, Canada, www.idrc.ca, and with financial support from the Government of Canada, provided through Foreign Affairs, Trade and Development Canada (DFATD), www.international.gc.ca.

Fusarium mycotoxins in cereals: combating a hidden menace

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A number of *Fusarium* species along with two *Microdochium* species are causal agents of Fusarium head blight (FHB) disease of cereals. The *Fusarium* species produce mycotoxins in grain that pose a risk to human and animal consumers. *F. graminearum* and *F. culmorum* are the most important pathogens on wheat in many regions. The most important mycotoxins are trichothecenes with deoxynivalenol (DON) being most important on wheat and barley. In contrast other *Fusarium* species predominate on oats and contaminate grain with T-2 and HT-2 trichothecene toxins. While some trichothecenes appear to function as virulence factors in some hosts the role of others is unknown.

Environmental factors play a significant role in determining which species predominate in particular situations and the degree of disease pressure.

Control of FHB is difficult, particularly where conditions are conducive to the disease. Some fungicides have efficacy against the mycotoxin-producing species and can reduce both symptoms and mycotoxin contamination. However, some fungicides cause increased toxin levels in grain, probably as a result of selective inhibition of Fusarium antagonists. Cultural control measures can also reduce FHB and mycotoxin contamination but such measures are too often not applied. Host resistance offers the best means to control both the disease and toxin risk but the genetics of resistance is generally complex in wheat and is poorly characterised in other cereal crops.

This presentation will provide a background to the risks and factors that influence FHB and toxin contamination and outline the progress being made towards countering the threat posed.

Tools of pedagogy and global partnership contribute to war on cassava viruses

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Cassava is a tuberous root crop grown in 105 countries; it is a key food security and industrial crop in Africa and Asia, respectively. Important biotic constraints to its production in Africa include cassava mosaic disease (CMD), caused by eight different geminiviruses and cassava brown streak disease (CBSD), caused by two ipomoviruses. Both diseases are spread by a whitefly vector and by planting infected cuttings. In addition there is the continued threat of wider spread, increased severity or even new viral diseases with global warming. In 2015, our multi-institutional team was funded by the Office of International Programs, of the American Phytopathological Society, to develop a case study teaching resource on CBSD, an integral step to infuse cassava-based perspectives into courses taught in universities in East Africa. The case study was tested and well received at four universities in East Africa. The case study has been submitted to the *Plant Health Instructor*, a peer-reviewed on-line journal. The next step is development of an on-line tool highlighting new research tools in CBSD management. The aim of such initiatives is to strengthen ties with other institutions and strengthen the initiative of the Global Cassava Partnership for the 21st Century GCP21 to attract highly qualified graduates into the cassava workforce. GCP21 looks for gaps in cassava R & D at the global level, and since 2008 has held conferences and strategic meetings that steer communication and aim to interest top labs and transfer modern science into cassava research (www.gcp21.org).

Detection of possible pathogenicity-associated genes in *Fusarium oxysporum* f. sp. *elaiedis*

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Pathogenic isolates of *Fusarium oxysporum* classified as formae speciales based on their host specificity, cause vascular wilts as well as root rots and crown rots in several cash crops across the globe. *Fusarium oxysporum* f. sp. *elaiedis* (Foe) is a major problem to oil palm cultivation in Africa. Three selected Foe isolates from three main locations in Ghana were characterised together with Ivorian Foe isolates using pathogenicity tests, identification and sequencing of effectors and housekeeping genes. A concatenated tree for two housekeeping genes could not distinguish between pathogenic and non-pathogenic isolates. Six effectors were identified within Foe, specifically Secreted-in-xylem (*SIX*) genes, which were reported earlier in *F. oxysporum* f. sp. *lycopersici*. Pathogenicity could not be strongly related to the presence or absence of the *SIX* genes as some effectors were either present or absent in the pathogenic Foe isolates tested and the remaining Foe isolates that were sampled from symptomatic palms in the oil palm plantations. *SIX8*, *SIX9* and *SIX11* were present in all pathogenic Foe isolates tested whereas *SIX1* was only present in four of the Foe isolates. Different *SIX* genes were identified in other *Fusarium* isolates. There were, however, clear differences between isolates unique to Foe compared to the other *Fusarium* species from the individual *SIX* phylogenetic trees generated. A rapid detection assay using loop-mediated isothermal amplification (LAMP) was developed to enable on-site diagnosis of the Foe within a minimum (4:00 min) amplification time targeting the *SIX8* genes.

Modelling fungicide resistance management strategies: mixtures and alternation

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Fungicides play a key role in modern agronomy but pathogen populations often adapt rapidly under the significant selection pressure exerted by these chemicals, threatening continued effectiveness of disease control. Typical advice to mitigate spread of fungicide-resistant strains involves utilising chemicals with different modes of action together in spray programmes. We use mathematical modelling to investigate whether this is best done by mixing or alternating fungicides. We also examine the trade-off between model generality and accuracy in providing practical advice for farmers and agronomists.

We investigate the question of alternation against mixtures as there is an implied trade-off. Mixtures lead to splitting of doses, imposing stronger selection due to the curvature of dose–response curves (e.g. half the dose gives more than half the effect). Alternation avoids this, but surrenders the suppression of resistant strains provided by applying multiple fungicides simultaneously.

We use models of *Zymoseptoria tritici* and *Erysiphe necator*, previously verified against field data, in our study. We rate control strategies by long-term yield and how well the spread of resistant strains is retarded. We consider a hierarchy of models, from the abstract to the biologically detailed.

The expected trade-off is observed, but is dominated by effects driven by fungicide application timing. We find that depending on the shape of the dose–response curves, and the doses being used, either strategy can prove superior. Our results highlight the need for system specificity and biological realism in mathematical models, particularly those intended to generate practical advice for growers.

Increasing accuracy of powdery mildews (Ascomycota, Erysiphales) identification using previously untapped DNA regions

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The powdery mildews (Ascomycota, Erysiphales) are a group of obligate biotrophic fungi found on nearly 10,000 angiosperm plant hosts globally including many that are important horticultural and agricultural plants. Infection can greatly reduce the appearance and vigour of the host therefore reducing attractiveness and yields significantly. A reliable and efficient method is required for unambiguous identification of these often cryptic species such that spread to new areas and/or new hosts can be detected rapidly and controlled early. This research aims to combine currently accepted techniques – host identification, fungal morphological analysis, DNA sequencing of the fungal rDNA ITS region – with sequencing of additional nuclear DNA regions in order to increase the reliability of the identification process via BLAST, DNA Barcoding, and phylogenetic reconstruction. Samples were collected through the Powdery Mildew Survey (a citizen science scheme), begun in 2014 and currently ongoing. Generic fungal DNA primers were found to amplify non-powdery mildew species, some of which were hyperparasites, as well as powdery mildews, and were therefore not a useful technique for accurate identification of powdery mildews. Consequently specific primers were developed for the amplification of the β-tubulin, actin, IGS, chitin synthase, elongation factor-1α, Mcm7, and Tsr1 regions. Initial results indicate that several of these regions could be used alongside ITS to increase identification power (reliability and accuracy). These rapid diagnostic techniques could provide a valuable tool for plant quarantine, particularly for greater security in the movement of plants and plant products in trade.

Off-host spore survival in *Zymoseptoria tritici* suggests a route for the maintenance of genetic variability and pathogenicity between crop cycles

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Zymoseptoria tritici, the causal agent of Septoria tritici blotch in wheat, has evolved resistance to all major antifungal chemistries. The speed at which this has been achieved, and the associated expense in controlling this fungus, has elevated its status to the most economically important pathogen of wheat in temperate climates, now accounting for up to 70% of EU fungicide spending. The ability of *Z. tritici* to evolve resistance so rapidly relies heavily on both the plasticity of its genome, and the diversity of genetic resources available for sexual recombination. It is unknown, however, how such diversity, which apparently includes non-virulent strains, is maintained in the field as opposed to being outcompeted by virulent strains able to undergo multiple asexual sporulation events during a growing season. Evidence in the literature is beginning to suggest a role for host-independent survival of *Z. tritici* spores, thus proposing an alternative survival strategy for any non-virulent strains present. This study, which assesses the ability for off-host spore survival in a nutrient free environment, shows that spore populations can survive autonomously for extended periods, a trait strongly correlated with lipid nutrition. Additionally, somewhat surprisingly, spores from these 'starved' populations suffer no significant loss in virulence, thus potentially allowing for non-virulent strains to infect subsequent crops. Together, these findings suggest a route for the maintenance of high genetic variability and subsequent pathogenicity between crop cycles, and thus have far reaching consequences for how and when we treat *Z. tritici* in field situations.

The beneficial effects of silicon on strawberry plants including reduced susceptibility to strawberry powdery mildew

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Strawberry powdery mildew, *Podosphaera aphanis*, is a major fungal disease affecting strawberry production worldwide. The work reported here assessed the use of silicon (Si) as a nutrient applied via the fertigation tubes in contributing to reduced susceptibility to *P. aphanis* and two-spotted spider mites. Leaf petiole Brix° level, pollen viability and pollinator activity were also investigated.

The field trials were set up at Maltmas Farm in Wisbech, Cambridgeshire under the commercial strawberry polythene tunnels between May and September in 2014 and 2015. The silicon nutrient used was Sirius (OrionFT). Treatments were untreated control, commercial fungicides applied under the farm normal practice, 0.017% Si alone and 0.017% Si plus commercial fungicides. The percentage cover of mycelium and the number of two-spotted spider mites per leaf were assessed fortnightly in 2014 and 2015. The Brix° level of leaf petioles, pollen viability and pollinator presence were assessed fortnightly in 2015.

In both years, 0.017% Si plus commercial fungicides had the greatest level of reduced susceptibility to *P. aphanis*. 0.017% Si alone had lower AUDPC value compared to the untreated control (475 against 662 in 2014 and 267 against 281 in 2015). Treatments with Si had reduced susceptibility to two-spotted spider mites compared to the untreated control in both years. The average Brix° readings of petioles from the untreated control and 0.017% Si alone was 7.36 and 8.52. The pollen viability of the untreated control and 0.017% Si alone was 78% and 80.3%. The presence of pollinators showed no significant difference between treatments.

Aphid-killing bacteria: how do they work?

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Insecticide resistance and on-going legislation changes on the use of insecticides are likely to reduce their availability for use in agriculture; hence there is an urgent need to develop effective biological controls against these plant pests. Aphids are major insect pests of the agricultural and horticultural sectors. We aimed to identify and characterise potential aphid-killing pathogens. We screened a range of phylloplane-residing bacteria for their ability to kill aphids and used alternative insect targets to determine host specificity. An artificial feeding system with a liquid diet was used to devise a high-throughput screening system to identify pathogenic bacteria against the green peach aphid *Myzus persicae* ("wild type" susceptible clone plus insecticide resistant clones). Six bacterial strains were pathogenic to all insecticide susceptible and resistant clones although variation in susceptibility was observed. No single bacterial strain was identified that was consistently more toxic to insecticide resistance clones than susceptible clones, suggesting there was no penalty in resistant clones that makes such clones less fit to bacterial challenge. Genome sequence analysis was used in combination with RNASeq to examine changes in aphid and bacterial gene expression during early stage infection. The altered transcript profiles revealed stress and cell morphogenesis genes in aphids and osmoregulation, while insecticidal toxins, stress and iron efflux transporter genes were differentially expressed in bacteria. These data are being used to understand the molecular basis of aphid mortality to bacterial infection with the aim of utilising them as effective biocontrol agents.

Engineering of CRISPR/Cas9-mediated virus resistance in transgene-free *Arabidopsis* plants

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CRISPR/Cas9 is a new technology that has opened up the possibility to edit the genomes of many model and crop plants with high precision and efficiency. While a number of studies have demonstrated the applicability of this technology for generating mutations in a wide range of plants, few of them have created plants with increased vigour/ marketability. Hence the mechanisms of CRISPR/Cas9 editing in plants has been widely characterised but its usefulness for agricultural applications is yet to be fully explored. Recently, we used CRISPR/Cas9 technology in *Arabidopsis thaliana* to generate novel genetic resistance to *Turnip mosaic virus* (TuMV), which is a major pathogen in vegetable crops. Importantly, the genome-edited, TuMV-resistant plants do not carry any transgenes and hence should not be restricted by current legislation for transgenic GMOs (genetically modified organisms). We hope that this pioneering research will pave the way for a new approach to generate virus resistance in important crops.

UK plant security: protecting plants and trees using science

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Protecting our trees and plants is enormously important for all of us – they supply our food and feed for animals, the air we breathe, support our economy and provide us with enjoyment and recreation. The Plant Biosecurity Strategy for Great Britain, published in April 2014 set out Government's approach to protecting plants from biosecurity threats, building on work undertaken by the Plant Health Services across Great Britain, which comprises Defra (supported by Fera and APHA) together with the Forestry Commission and the devolved administrations in Scotland, Wales and Northern Ireland.

One of the biggest challenges is to scan the horizon and predict what the new threats are; in particular new pest pathways, supply chains and high-risk trades. We need to know the identity, biology, epidemiology, distribution, transmission, hosts, pathways, climate adaptation, economic, social and environmental impacts – it's a massive scientific effort involving specialist natural scientists, economists, statisticians modellers and social scientists. I will present progress on pre-border activities to reduce the risk of pests and diseases arriving from overseas, including work with countries beyond the EU to drive up standards; activities at the border to reduce the risk of pests and diseases entering the EU and the UK and action inland to step up surveillance and improve preparedness. This includes developing a publicly available risk register, novel approaches to detection and diagnosis of pests and pathogens and contingency planning. I will also discuss our overall approach to plant health in England and management approaches to tackle priority pests and diseases.

Managing biosecurity risks in New Zealand: relevance to the UK?

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New Zealand is renowned as having one of the most effective biosecurity systems in the world. The Ministry for Primary Industries is the lead agency but the system incorporates a number of participants (including the trade, research institutes, local councils and the public). The major components of the system are integrated (a "biosecurity continuum" to reduce the likelihood of unwanted pests and pathogens entering New Zealand or to provide effective management should they establish. The system's components range from national and international legislation, pre-import activities, pathway and border management, national surveillance, response and management programmes, and New Zealand's export certification system. This presentation focuses on New Zealand's surveillance, response and management systems and uses a range of examples to illustrate how the system operates to mitigate the risks posed by exotic pathogens. This system and the context within which it operates, is compared and contrasted with the situation in the United Kingdom.

The role of the Plant Health and Seeds Inspectorate in plant biosecurity

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The UK's Plant Health and Seeds Inspectorate (PHSI) play a vital role in biosecurity, protecting the UK from harmful plant pest and diseases. Interacting daily with the trade through facilitation of plant and plant product imports, exports, certification, licensing, surveillance and action activities along with engagement, and biosecurity awareness practices, the PHSI has a wide range of responsibilities in plant health. Various statutory schemes (e.g. plant passporting and tree notifications) are managed by the PHSI to directly meet the challenge of improving plant health biosecurity, and reducing the risk of plant pathogen introductions from other parts of Europe.

Biological and epidemiological scientific knowledge of plant pathogens help understand the threat they pose to UK crops and the wider environment and ensure PHSI activities are focused on high-risk organisms. Plant pathological research into early, in-field diagnostic detection tools is paramount in helping front line PHSI rapidly identify suspect quarantine pathogens at points of entry to the UK and in the wider environment. Following findings, appropriate management practices can be implemented, eradicating the threat or reducing their spread. A number of recent innovations, e.g. LAMP Genie, are now being validated at points of entry to the UK and will be expanded on during this presentation.

The Plant Health Professional Register

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Two reports published in 2014[1,2] initiated an exercise with the inspectorate in UK government departments responsible for the health of plants, seeds and trees. The aim was to create a register that would both assist the chief plant health officer in finding expertise to respond to future disease outbreaks, as well as to acknowledge the skills of this workforce and promote its continual professional development.

The Plant Health Professional Register is administered by the Royal Society of Biology and sits alongside other professional registers, e.g. RSciTech, RSci and CSci. An advisory board comprises representatives from the Animal and Plant Health Agency, Fera Science, Defra, Science and Advice for Scottish Agriculture, the Department for Agriculture and Rural Development in Northern Ireland, Welsh Government and the Forestry Commission. The board developed competencies to recognize the skills of employees and satisfy the Plant Biosecurity for Great Britain strategy.

The pilot exercise was highly successful, resulting in a staged plan to register 300 government inspectors by 2018.

Government alone cannot protect the UK landscape from attack from pests and pathogens and on-going work will look at the value of the Plant Health Professional Register, with appropriate amendments to competencies, to other sectors e.g. industry, agronomists, academia and others.

This talk will present the state of play of the register in September 2016.

[1]Defra/GoS (Dec 2014) Animal and Plant Health in the UK: Building our science capability.
[2]Defra (April 2014) Protecting Plant Health-a Plant Biosecurity Strategy for Great Britain.

Early detection surveillance for emerging vector-borne pathogens

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Emerging plant pathogens are an increasing source of economic loss and a risk to food security in many countries. Due to the considerable costs of control once these pathogens become established, scientifically informed and economically justified surveillance activities aimed at early detection are required. Sampling for detection of vector-borne pathogens is complicated further by the need to consider whether to sample from vectors, hosts, or both populations. Despite this, current strategies rarely take the epidemiology of the pathogen into consideration when making this decision. Using an epidemiological modelling approach we show that the sampling effort required from either hosts or vectors in order to detect a specified mean prevalence can be presented as a linear function of the sampling effort from the other population (host vs vector). This linear relationship suggests that the total sampling effort required will not be minimised when sampling from a combination of hosts and vectors. Therefore, the optimal course of action in order to minimise total sampling effort will be to sample exclusively from either the host or the vector population. Using our method we show how to determine which population to sample from in order to minimise total sampling costs (assuming a linear cost function), based upon the epidemiological characteristics of the pathosystem under question and the relative costs of sampling hosts versus vectors.

Biosecurity at the Eden Project – considering local and global threats

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The Eden Project is a botanic garden, educational charity and visitor attraction in Cornwall, which has been open to the public since March 2001. It was designed to celebrate the interdependence of plants and people, and to educate people about the need to care for the world. In the fifteen years since opening there have been many pest and disease management challenges following the first plantings in the rainforest biome. Even at the early stages, when a small team of horticulturists started procuring and growing plants from around the world in preparation for the biomes, it was clear that there was a significant risk of importing pests and diseases. This was despite ensuring that all plants were imported with their relevant plant passports and phytosanitary certificates.

At the Eden Project, biosecurity is taken very seriously and is included in our risk management plan. As a member of the International Plant Sentinel Network we are concerned not only with protecting our plant collection, but also protecting the wider environment by identifying emerging pest and pathogen threats globally. Our Plant Health Procedures outline our quarantining and inspection procedures, as well as restrictions on plant movements and importation of other organic materials into the gardens. In addition, our Plant Records and Acquisition Policy ensures responsible and legal sourcing of all new plant material. By having strict protocols in place we hope to safeguard our plant collection for the future, whilst helping others to do the same.

Scottish Plant Health Service – role in safeguarding food security, biosecurity and trade

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Scottish Government (SG), as part of the UK Plant Health Service, undertakes many activities to protect the health of plants and to facilitate the safe trade in plants and plant products. SG recently published the Scottish Plant Health Strategy 2016–2021, which aims to safeguard agriculture, horticulture, forestry and the wider environment from plant pests. This will involve an integrated approach and effective collaboration between all interested parties.SG undertakes many activities to ensure food security and to prevent the spread of pests. Examples of the different types of activities undertaken in Scotland will be given during the presentation and these include:

- Inspection of plants and plant products being moved in trade for freedom from quarantine pests
- Working with the rest of the UK Plant Health Service to assess risks from new and established plant pests
- Running the UK Potato Quarantine Unit on behalf of the UK Plant Health Service, testing potatoes from outside the EU for plant pests prior to planting in the UK
- Certification of crop species to ensure high health planting material is available for farmers, growers and the public
- Surveillance and contingency planning for pests and their control or eradication
- Commissioning research to underpin official plant health activities
- Providing publicity material on new pest threats so that all are informed of risks and can take appropriate actions

However, protecting plant health is not solely a responsibility of government and we are working with stakeholders to ensure we all play our part.

Biomonitoring of plant pathogens in Canada – technologies and genomic approaches

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Movement of fungi between continents has caused major ecological, agronomical and economic disasters. In an era where international trade is continuously expanding, preventing and mitigating new pathogens is a major challenge, and has led to international treaties to reduce risks while maintaining trade. The spread of phytopathogens by wind, rain and insects can cause the introduction of exotic plant diseases. Since it is hard to trap and investigate these organisms, new technologies such as qPCR and metagenomics with Next Generation Sequencing (NGS) are being investigated for rapid detection and monitoring of fungal biomes in environmental samples but these raise major issues in a regulatory context. Most quarantine fungal species have close relatives that are innocuous to crops or the environment. When the fungal biome is monitored with NGS using barcode amplification, a small percent difference between barcode sequences of these closely related species may generate results that are misleading in the regulatory context. The situation is even more precarious when the closely related species are unknown, a problem particularly acute for obligate plant pathogens. Our results using biosurveillance tools show the potential of these methods to identify potential sources of entry, abundance, and biodiversity that can be used to determine the level of risk associated with these species for growers and trade. Case studies on biomonitoring methods of plant pathogens from database generation, molecular tool development in Canada will be presented on how internationally accepted risk analysis protocols are in place for NGS data.

Global trade and increasing forest and environmental bio-insecurity

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Damage to trees and forests from introduced non-native pests and pathogens is a growing global phenomenon with profound community, ecosystem and economic impacts. The number of infestations for any given country is significantly correlated with trade and is not related to forest area. Many organisms arrive on living plants or plant products including wood packaging. In the UK, an increasing threat from exotic pathogens were first noted in the 1990s. Since then, the emphasis of UK forest protection research has largely shifted from management of endemic pests to often unsuccessful post hoc attempts to control outbreaks of introduced pests across rural and urban landscapes, woodlands and plantations.

Current international regulations aimed at reducing the risks of introduction are oriented to promotion of trade and take insufficient account of modern science, e.g. lack of any meaningful assessment of the scale of the threats, and reliance on named organisms when many organisms spread via international trade are largely unknown to science. Often their geographical origins, and hence their natural behaviour, are also unknown. In practice, the regulations have proved inadequate and relevant institutions are often conflicted.

The UK is in something of a crisis over how to future-proof its forests and ecosystems against unknown pathogen threats and climate change. Recent epidemics have brought the issue into sharper focus, but the changes in international biosecurity protocol required to prevent further pest incursions have yet to emerge. For the UK specifically, imposition of stricter border controls may suddenly and unexpectedly have become an option.

Population structure of the ash dieback pathogen *Hymenoscyphus fraxineus* in the UK

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Common ash trees, *Fraxinus excelsior*, have been subject to heavy dieback and mortality in their native range caused by the pathogenic fungus, *Hymenoscyphus fraxineus*. *H. fraxineus* was first observed in the UK in 2012 in eastern England, although it is likely to have been present for many years before that. It is thought that the pathogen has infected trees in the UK via two routes, through airborne sexual spores from continental Europe and by planting infected saplings imported from continental nursery stock.

We determined the population structure of *H. fraxineus* in the UK at sites where the disease was considered to have been initiated either by airborne spores (established sites) or from planted stock (planted sites). Samples were obtained from three established and three planted sites in 2014-15. Vegetative compatibility (VC) among the isolates was tested to determine the population structure. A total of 30 SNP markers based on KASP™ genotyping chemistry were developed using the sequence information of 43 *H. fraxineus* isolates from across the Europe including the UK to study genetic differentiation at the sampled subpopulations.

VC tests and SNP marker data both showed a high level of genotypic diversity in local populations. They indicated that all subpopulations originated from the same source population and that each subpopulation was founded by several individuals. Genetic differentiation between planted and established sites was small, indicating that planting stock and airborne infection have contributed similar amounts of genetic diversity to the newly established population of *H. fraxineus* in the UK.

Trade restrictions and control strategies: a modelling case study

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Cassava is a key staple crop for subsistence growers in East Africa, where it is often relied upon for food security. Cassava brown streak disease is a major threat to this, causing yield losses of up to 75%. One dispersal mechanism for the pathogen is the informal trade of planting material, which can be particularly difficult to regulate.

Creating a mathematical model of the system, we show that this trade is likely to be vital to long-range dispersal of the pathogen, while the vector of the pathogen is more important in local amplification of the disease. An agricultural police unit is currently being set up in Uganda to try and restrict the informal trade of untested material, and our model suggests that any such efforts will reduce the dispersal rate of disease.

However, to effectively reduce disease, other control methods are required. One such method in the early stages of establishment in Tanzania and Uganda is the clean seed system, for distribution of disease -free, improved planting material. A key consideration is the importance of informal trade of the improved material between growers, extending the reach of the introduction of clean seed beyond the direct beneficiaries. We show that restrictions in trade cannot always be effectively combined with clean seed systems; the success or otherwise of restrictions depends on the introduction strategy used.

International trade in plants and food security

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Threats to plant health exacerbate the food production shortfall that already prevails in various regions of the world due to unfavourable environmental and social conditions. Numerous endemic disease agents wreak havoc on such staples as cassava, maize, rice and wheat, and inflict substantial losses on pastures, threatening the livelihoods of vulnerable farmers and severely undermining the calorie intake and the nutritional status of millions in developing countries. Prevention and control of endemic phytopathogens is challenging, given climate change-induced alterations in the host–pathogen–vector interactions that lead to more frequent outbreaks and upsurges and the continuous appearance of new pathotypes. Plant health disciplines contribute to building resilience in farming systems, including through providing the necessary knowledge for the selection of resistant breeds. This helps address food shortage. However, to achieve global food security additional food supply needs to be moved from surplus to deficit areas. The WTO Agreement on the Application of Sanitary and Phytosanitary (SPS) Measures ensures the safe transboundary movement of goods, including foodstuffs. It aims to limit the risk of introduction, through the trade pathway, of pathogens that could otherwise severely damage plant, animal and human health or the environment. Importing plants and plant products requires a science-based risk assessment to establish adequate risk management measures that guarantee the freedom of imported products from quarantine pests and diseases. Plant pathology is essential to accurately assess the risk and identify appropriate measures for its management, allowing food to reach its consumption area while protecting the ecosystem from disease dissemination.

Understanding and monitoring *Phytophthora palmivora*, the causal agent of bud rot disease of oil palm

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Palm oil is a key component in Malaysian commodity trading and production of palm oil in 2015 was recorded at 19.97 million tonnes with an export value from the palm oil and other oil palm products of approximately RM 60.17 billion. Keeping exotic diseases out is crucial for minimizing the losses of oil palm production to diseases. Bud rot, triggered by *Phytophthora palmivora*, is one of the exotic diseases that threatens Malaysian production. The disease has cause significant losses to the industry in Colombia, but no incidence has been reported in Malaysia or other Southeast Asian countries. Several aspects of research need to be conducted to understand why this pathogen causes problems in oil palm in South America but not in Southeast Asia.

This study aimed to analyse variation between the Colombian *P. palmivora* isolates that cause bud rot disease in comparison with Malaysian isolates and other isolates gathered from different hosts and regions. Our hypothesis was that *P. palmivora* isolates from the different regions and/or hosts have different molecular characteristics and have dissimilar levels of pathogenicity. Sequence alignments of several genetic markers (from housekeeping genes) failed to distinguish between Colombian oil palm isolates and *P. palmivora* from different hosts and regions. However, a new marker named as HPAVR along with AFLP analyses, separated the Malaysian and Colombian isolates into distinct clades. Pathogenicity assays using the zoospores for artificial inoculation are currently being conducted, but not enough evidence has been collected to confirm that pathogenicity correlates with these distinct clades.

Cacao swollen shoot virus – quarantine measures and vector biology

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The International Cocoa Quarantine Centre run by the University of Reading serves to reduce the risk of pest and disease transmission during the movement of germplasm between cocoa-producing countries. Studies of the genetic diversity of *Cacao swollen shoot virus* (CSSV), the most damaging virus affecting cacao, allow for more robust screening of quarantine material while analysis of the feeding behaviour of the insect vectors of CSSV inform the design of protection measures for the crop.

Since the effectiveness of mealybugs (Hemiptera: Pseudococcidae) as CSSV vectors is thought to be species-dependent, electrical penetration graph analysis is used to characterise their behaviour as they feed on cacao. Identification of electrical waveforms indicative of distinct stylet events means that time spent by cacao-feeding mealybugs in particular activities can be quantified. In this way, differences were observed between species for the salivation events linked to virus transmission. There are over 60 species of mealybugs found on cacao and their identification requires a high degree of expertise, depending as it does upon characterisation of microscopic structures that are, *in vivo*, often obscured by wax exudates. A novel histological approach involving detailed morphological characterisation of mealybugs combined with high resolution melt-based DNA barcoding of those same samples allows for the identification of CSSV vectors in regions without access to DNA sequencing facilities. This approach has identified the most abundant mealybugs in CSSV-affected areas of West Africa and these species are being prioritised for analysis of their vector potential. Feeding trials have confirmed the ability of these mealybugs to transmit CSSV between plants and also demonstrated that this capacity is not restricted to those species known to be found on cacao.

Overcoming challenges in the detection and diagnostics of psyllid vectors of *Candidatus* Liberibacter solanacearum

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The bacterium, *Candidatus* Liberibacter solanacearum (CaLsol) infects solanaceous and apiaceous crops and is vectored by several species of psyllids. Infected potatoes develop zebra chip disease, which has led to economic losses throughout Central and North America, and New Zealand. In Europe, the CaLsol haplotypes have been found to primarily infect apiaceous crops, and in carrots lead to stunted growth and leaf discolouration. The high species diversity of psyllids (>90 species in the UK; approx. 3800 worldwide) and the lack of taxonomically useful characteristics for some species presents a challenge in the identification of vectors. Species are frequently misidentified and the current distribution of several important species is uncertain.

We present an on-going project in collaboration with Rothamsted Research, SASA, the NHM, and SLU that aims to review and survey psyllid vectors and sampling techniques. To ensure accurate species identification we use a combination of classical and molecular taxonomy to develop molecular diagnostic assays allowing rapid and accurate species identification. We selected species for assay development using a set of criteria to determine whether they could potentially vector CaLsol if the bacterium were to be introduced into the UK. Specimens were sequenced and cross-referenced with voucher specimens obtained using non-destructive DNA extraction techniques. We tested assays on bulk suction trap catches and assess the use of a regional suction trap network as a means to monitor psyllid migration and dynamics to ensure the detection of vectors should they occur.

POSTERS

**J Colhoun competition entrant*

*The effector complement of *Phytophthora fragariae*

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Strawberries are a valuable fruit crop both for the UK and worldwide, resulting in a UK home retail value of over £500 million in 2015. However, with the increased threat from unpredictable weather conditions and the withdrawal of many preventative chemical fungicides and soil fumigants, this industry is at risk. One of the major threats to the industry is the water-borne disease strawberry red core, caused by the oomycete pathogen *Phytophthora fragariae*. This work aims to identify the genes within the pathogen that encode effectors, which when identified by the corresponding resistance gene in the host trigger a resistance response. Resistance to *P. fragariae* is known to be based upon a gene-for-gene model, where a resistance protein in the plant detects a single secreted protein from the pathogen and triggers a resistance response.

In order to aid the identification of candidate genes, the genomes of ten isolates of *P. fragariae* were sequenced by Illumina short read sequencing and one isolate by single molecule real time sequencing using PacBio. The gene complements of these genomes were predicted and analysed to provide a list of predicted proteins containing key features of avirulence genes: secretion signal peptides and the RxLR, EER and WY motifs. By comparing the gene complements between strains of known race types, we generated a list of candidate avirulence genes. Further investigation will identify which of these genes are key to the resistance response in the plant.

*Reducing epidemics of strawberry powdery mildew using a silicon nutrient to give reduced susceptibility to *Podospheara aphanis*

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Work carried out at the University of Hertfordshire has shown that the regular use of a silicon nutrient in the fertigation tubes at a commercial strawberry farm results in reduced susceptibility to pests and diseases. Powdery mildew of strawberry (caused by *Podospheara aphanis*) is the most important disease of the protected strawberry crop in the UK and has to be controlled by frequent use of fungicides. The work reported here aims to investigate the action of the silicon nutrient delivered via fertigation tubes on the plant resulting in reduced disease susceptibility.

The effect of a silicon nutrient on levels of strawberry powdery mildew was monitored fortnightly from May to August in a trial in a commercial crop. Fifty leaves were sampled from each treatment at each assessment date. Treatments were: control (no silicon and no fungicides), commercial fungicides without silicon, silicon plus commercial fungicides, silicon without fungicides, silicon twice a week plus commercial fungicides, and silicon twice a week without fungicides respectively. The silicon alone reduces disease susceptibility, and the lowest disease levels are found in the silicon with fungicide treatment. The twice a week application of silicon could result in further reduced disease susceptibility compared with a single treatment of silicon per week.

*Interaction between nitrate supply and severity of spot blotch of wheat caused by *Bipolaris sorokiniana*

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Spot blotch caused by *Bipolaris sorokiniana* (*Cochliobolus sativus*) is a serious disease of wheat grown in warm climates. The impact of plant diseases is influenced by plant nutrition, and it has previously been reported that nutrient stress affects spot blotch severity. We are investigating the interaction between nutrient supply, spot blotch disease severity and regulation of defence gene expression in Iraqi wheat. The susceptibility of seven Iraqi wheat varieties to spot blotch was tested. Among them, Dor29 was the most susceptible and Latifia the least susceptible. These varieties were selected for further investigation. Seedlings were grown in sand watered with Hoagland's solution modified to contain 0.75 mM, 3.75 mM or 7.5 mM nitrate. Eighteen-day-old seedlings supplied with 0.75 mM nitrate had significantly fewer leaves and tillers than those supplied with 3.75 mM or 7.5 mM nitrate. Fourteen-day-old seedlings were inoculated with spot blotch. Four days after infection there was a trend to higher disease severity at the higher nitrate levels, particularly for the more susceptible variety Dor29 at 7.5 mM nitrate. In contrast, some – but not all – previous studies, which were mostly with field-grown plants, found that adequate nitrate supply reduces spot blotch severity. Thus, effects of nitrate most likely depend on environment and growth stage. There is a complex pattern of changes in expression of genes coding for WRKY transcription factors in response to nutrient stress and infection by *B. sorokiniana.*

*Fine mapping, gene identification and characterisation of resistance to *Rhynchosporium* on 6H in barley

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Rhynchosporium commune, causing leaf scald, is one of the most destructive pathogens of barley worldwide, leading to yield losses of up to 30–40%. Despite advances in molecular marker technology and the sequencing of the barley genome, little is known about barley resistance to *Rhynchosporium* at the molecular level.

The major resistance gene *Rrs13* has been mapped to the short arm of chromosome 6H in a BCline 30 × Clipper population. A large resistance QTL on 6HS has also been identified in CIho3515 × Alexis and Steptoe × Morex double haploid (DH) populations. The latest version of the barley genome assembly revealed that the large QTL identified in the Steptoe × Morex and Clho3515 × Alexis populations is located at a different locus to that of *Rrs13*, and has subsequently been named *Rrs18*.

Fine mapping using selected Steptoe × Morex BC1 F3 lines has been combined with exome capture variance calling to narrow down the interval containing *Rrs18* and to identify candidate genes for *Rrs18.* Further fine mapping will shorten this list of candidates and enable development of diagnostic molecular markers for use by breeders. Sequencing of RNA from resistant and susceptible lines will be used to identify genes or alleles specific to resistant lines that are not present in the Morex genome annotation. Candidate genes will be validated by overexpression in susceptible cultivar Golden Promise to identify the first barley resistance gene to *Rhynchosporium*.

Characterization of barley host and non-host interaction with aphids

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Aphids are phloem-feeding insects that cause substantial yield loss on a wide range of crops, including important cereal crops such as barley. Most of the aphid species are limited to one or few host species, but some species are able to reproduce in many plants belonging to different families. While feeding, aphids probe the leaf surface secreting proteins regardless of the plant species confronted. This observation points to different molecular events take place during host and non-host interactions. These molecular interactions between the aphid and the plant potentially determine the aphid host range. We aim to develop control strategies for aphids by investigating mechanisms of resistance (including non-host resistance) and susceptibility. In this project, the responses of the different barley cultivars–aphid species interactions are investigated. First, we determined colonization efficiency, aphid probing behaviour and elicitation of barley defences for three aphid species, defining three types of interactions host, poor host and non-host. A microarray experiment has revealed that each species elicits a unique barley response. Analyses of barley transcriptional changes showed a more pronounced response during the poor-host versus host interaction, and few genes affected by the non-host interaction. We identified gene sets specifically up-regulated during the poor-host interaction. These include several members of the thionin antimicrobial peptide family, detoxification or water stress response. Our current goal is to further implicate these genes in aphid host range using functional assays. Our work will thereby provide novel insights into host and non-host defences in a monocot crop to aphids.

*Ultraviolet light induced photocatalytic removal of ethylene in post-harvest storage, for the control of post-harvest pathogens on fruit crop

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Ethylene is a gaseous phytohormone released by climacteric fruits during the ripening process. Although it is well established that ethylene plays a significant role in the defence against various pathogens, much research suggests potentially negative effects with regards to increased pathogen susceptibility within post-harvest storage environments.

Pathogens such as *Colletotrichum gloeosporioides* and *Botrytis cinerea* can utilise quiescent stages in their infection cycles. They can land, and remain dormant, on a suitable unripe host prior to switching to a necrotrophic lifestyle at the onset of ripening, when the host environment becomes more favourable for disease onset. Due to ethylene playing a major role in the ripening of fruit, it is suggested to be a causing factor in post-harvest disease. Moreover, research has shown ethylene to induce spore germination and multiple appressorial formation.

Previous research on the effects of ethylene removal on fruit–pathogen interactions has largely revolved around 1-MCP, a compound that blocks ethylene at receptor level. Whilst some studies show disease reduction, others demonstrate increased susceptibility. We are utilising a method of UV photocatalytic ethylene removal using titanium dioxide nanoparticles.

It is hypothesized that UV photocatalytic ethylene removal will prevent disease in two ways: (1) ethylene removal will delay the ripening of the fruits, delaying pathogen virulence from a quiescent stage, preventing disease symptoms and therefore spread, and (2) the external ethylene removal causing ripening delay will stimulate the fruits to up-regulate ethylene biosynthesis genes, which are known to play a major role in defence against necrotrophic pathogens.

*The molecular basis of pathogenicity of *Neonectria ditissima*

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European canker, caused by the phytopathogenic fungus *Neonectria ditissima*, is an economically important disease in almost all pome-producing regions of the world. This fungus is able to infect a wide range of apple varieties, causing canker and die back of young shoots. Plant resistance is a promising alternative to largely ineffective cultural control, but is time-consuming to deploy due the long breeding cycle in apple. A deeper understanding of the host–pathogen interactions, and how host resistance and pathogen avirulence are linked, is key to the deployment of durable resistance in the field.

The genome of *N. ditissima* facilitates the identification of putative effectors and pathogenicity genes through bioinformatics analysis of the secretome and through comparisons to other pathogens. We annotated a previously sequenced *N. ditissima* genome assembly using RNA-Seq data and have now improved this assembly using PacBio sequencing allowing us to present an updated analysis of the predicted secretome of this pathogen.

In due course RNA from infected host tissue will be extracted and further RNA-Seq data will be generated. Differentially expressed transcripts will be sought. The identification of specific candidate genes controlling pathogen virulence will allow a better understanding of the mechanism of infection. Similar comparisons will be carried out on the host, utilising the apple genome to identify putative resistance genes, differentially expressed between resistant and susceptible cultivars.

What is the origin of the initial inoculum for strawberry powdery mildew epidemics?

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In the last 25 years strawberry growing in the UK has been transformed, with yields going up whilst the number of hectares used has gone down. The UK now grows 80% of the strawberries eaten here. This has been achieved by precision choice of varieties, the use of fertigation and also the use of fleece in the spring and polythene tunnels, primarily during harvest. Everbearer strawberries are harvested from May through to the end of September, so crops will be under polythene tunnels for up to 5 months. The conditions of temperature and humidity in these tunnels are ideal for the development of strawberry powdery mildew.

Controlling strawberry powdery mildew is a major cost for strawberry growers, and the bigger the epidemic, the higher the cost. Growers tend not to see early disease development and then be taken by surprise when the epidemic builds up very suddenly. The question is often 'where did that come from?' Since 2011 we have followed the overwintering of the fungus on strawberry plants in the ground for 3 winters and also all the plants coming onto the farm for single-season planting. Growers source these plants from the UK and Europe. Results from propagators, and between propagators range from 6.5% plants showing infection to 90% on one delivery in 2015. The results displayed will indicate where the propagators are situated, and the possible impact of these disease levels both on disease developments and on costs to the grower.

BAK1 contributes to basal resistance against the wheat fungal pathogens *Zymoseptoria tritici* and *Fusarium graminearum*

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The two ascomycete fungi *Zymoseptoria tritici* and *Fusarium graminearum* are economically important pathogens of wheat worldwide, causing Septoria tritici leaf blotch (STB) and Fusarium head blight (FHB) diseases, respectively. Both diseases can have major impact on overall crop yield, whilst *F. graminearum* infection of wheat ears can also lead to contamination of the grain with harmful mycotoxins. Although the infection biology of both pathogens has been well characterised, the mechanisms behind either resistance gene-mediated or basal defence against either pathogen are not well understood.

In *Arabidopsis*, the leucine-rich repeat receptor-like kinase (LRR-RLK) BAK1 is an important regulator of pattern recognition receptor (PRR)-triggered immunity (PTI) and is known to be involved in defence against a range of bacterial, oomycete and fungal pathogens. BAK1 regulation of PTI depends on interactions with a number of other LRR-RLKs, including BIR1 and SUPPRESSOR OF BIR1 (SOBIR). BAK1 also appears to be required for the function of LRR-receptor-like immune receptor proteins (LRR-RLPs). It is currently unknown whether functional homologs of these genes exist in wheat.

We have identified at least 3 candidate wheat BAK1 genes, two of which (TaBAK1-2L and TaBAK1-6L) are up-regulated during *Z. tritici* infection. Using BSMV-VIGS to silence these genes individually, we show that leaves and ears of susceptible silenced plants support higher levels of *Z. tritici* and *F. graminearum* infection, respectively. These data suggest that BAK1 and therefore probably as yet unknown interacting surface receptor(s) also contribute to basal resistance in wheat against these fungal pathogens.

Sibling rivalry: the growing dominance of *Globodera pallida* and novel control measures for potato cyst nematodes

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Potato cyst nematodes (PCN; *Globodera pallida* and *G. rostochiensis*) are sedentary endoparasitic nematodes of potato and are some of the most economically important of all plant-pathogenic nematodes. They have an economic impact in the EU of €600 million p.a.

Surveys of PCN populations in the UK and mainland Europe have indicated that *G. pallida* is outcompeting *G. rostochiensis* in mixed populations. This appears to be caused by the use of conventional control measures such as nematicides, crop rotation and genetically resistant cultivars, each of which tends to be less effective at controlling *G. pallida* than *G. rostochiensis*. However, in Ireland, where such control measures are not routinely used, this laboratory has shown that the incidence of *G. pallida* is also increasing.

The BIOLOG Ecoplate™ system was used to construct a community level physiological profile of soil bacterial communities in a potato field. Measurements of soil bacterial activity and biodiversity indicated that potato ridge soil had significantly higher levels of both than unplanted (bulk) soil.

The tritrophic relationship between PCN, potato and rhizobacteria was examined with the aim of developing novel, biological control measures for PCN and in particular for *G. pallida*. Conducting experiments with potato root leachate has shown that microorganisms present in potato ridge soil have a role in PCN hatch.

G. pallida is emerging as the primary target for new PCN control measures. The reason for its dominance over *G. rostochiensis* in mixed populations requires further examination, particularly concerning the role played by soil microorganisms.

*Investigations of the barley powdery mildew effector (CSEPs) in non-host plant wheat

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As a highly host-specific and biotrophic fungal pathogen, *Blumeria graminis* f. sp. *hordei* (Bgh) can only survive and cause powdery mildew disease on living barley plants. Wheat is resistant to Bgh, which is a form of non-host resistance. To date, over 500 candidate secreted effector proteins (CSEPs) have been identified in Bgh. Among these effectors, CSEP0064 and CSEP0264 were found to be similar to RNAses and are up-regulated during infection. Recently, a *Pseudomonas fluorescens*-based effector-to host analyser strain (EtHAn) was developed as a modified T3SS, which is able to deliver effectors into plants. In this project, we deliver two of the barley powdery mildew effectors, CSEP0064 and CSEP0264, into wheat using the EtHAn strain. We are using this system to screen for responses to the effectors in a mapping population of wheat-(WAGTAIL). After screening, we finally observed differential responses in some of the wheat lines. Ultimately, we hope to localise the gene/genes that are involved in effector recognition and which may be important for non-host recognition and resistance.

Coexistence of *Leptosphaeria* spp. on oilseed rape crops in the UK

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Phoma stem canker of oilseed rape is associated with *Leptosphaeria maculans* and *L. biglobosa*, which coexist on oilseed rape crops. Generally *L. maculans* is considered more damaging than *L. biglobosa*[1]. However, substantial yield losses due to *L. biglobosa* have been reported in Poland[2] and the disease in China is associated only with *L. biglobosa*[3]. One main method used for controlling phoma stem canker is by deployment of cultivars resistant against *L. maculans*. However, there are no breeding programmes for cultivar resistance against *L. biglobosa.* Samples of airborne inoculum were used to examine the abundance of the two *Leptosphaeria* species in air samples at three sites in the UK. Their abundance was also investigated on cultivars differing in their resistance against *L. maculans* over three cropping seasons (2011/12 –2013/14). The proportions of the two species were assessed at the phoma leaf spotting and the canker stage. The results indicated that the abundance of *L. biglobosa* was less on oilseed rape cultivars with extensive colonisation by *L. maculans* than on those with little colonisation by *L. maculans*. Effective control of *L. maculans*, i.e. by using cultivars resistant against this pathogen, may provide a niche for *L. biglobosa*, increasing the possibility of severe epidemics caused by *L. biglobosa* in future.

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[2]Jedryczka M, 2007. Phytopathol. Polon. 45: 73–75.
[3]Zhang X *et al.*, 2014. Plant Pathol. 63: 598–612.

*Stem base temperature in oilseed rape and implications for disease progress models

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Phoma stem canker causes yield loss of oilseed rape due to development of cankers at the stem base. Cankers restrict nutrient and water uptake late in the season leading to premature seed ripening and pre-disposing plants to lodging. Temperature experienced by the casual *Leptosphaeria* species growing at the base of oilseed rape stems differs from air temperature. Temperature in the stem base is buffered from air temperature extremes. The relationship between mean stem base temperature and air temperature is linear but depends upon plant growth stage. Stem base temperatures are more strongly correlated with near-surface soil temperature. These findings have implications for the efficiency of disease models for phoma stem canker that depend upon a linear relationship between air temperature and canker growth.

*Quantum pathogen evolution by integron-mediated effector capture

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Plant pathogenic Pseudomonads are responsible for the loss of millions of pounds in crop revenue each year. Pseudomonads export effector molecules into the plants' cells in order to supress immune responses such as the plant hypersensitive response (HR). The plants immune system can recognise certain effector molecules and trigger the HR preventing bacterial infection. Pseudomonads can evade HR by potentially gaining different effector molecules captured within integrons, mobile pieces of DNA that have the ability to capture and express genes. There are genes conserved within integrons that can be identified such as *xerC* and the UV damage repair operon *rulAB* appears to be a hotspot for integron insertion. Using these features it has been possible to identify a number of integron-like elements (ILEs) within *Pseudomonas syringae* pathovars. The three regions used to identify potential ILEs were the *rulAB* operon, the *xerC* gene and the ILE insertion junction, *rulB-xerC*. The screening revealed 22 new uncharacterised ILEs with at least one carrying different cargo genes from previously identified ILEs within plant pathogens. The screening also revealed that the XerC integrase is conserved across multiple plant pathogens at different loci. UV tolerance tests have also been performed to ascertain whether the disrupted *rulAB* operon is still functional after ILE insertion. Ongoing work includes examining the conditions required for ILE gene expression, assessing the mobility of the identified ILEs and investigating the identified ILE sequences further.

*Structural basis of the mode of action of RNase-like effectors in cereal powdery mildews

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Food security is a growing issue due to continuously increasing world population, agricultural space limitation and changing climate. Moreover, currently cultivated crops are under constant abiotic and biotic stresses that lead to harvest losses. *Blumeria graminis* is a major fungal pathogen that causes powdery mildew disease in wheat and barley. It utilises hundreds of CSEPs, small, secreted effector proteins in order to subdue host immune defences and establish infection. Effectors with RNase-like structure seem to be particularly prevalent within CSEPs. This study investigates BEC1054 (syn. CSEP0064) previously shown to be an essential *B. graminis* effector in establishing successful infection. We hypothesise that BEC1054 is involved in preventing host cell death via competing with certain ribosome inhibiting proteins (RIPs) for plant ribosome binding. Analysis of total RNA extracts from transgenic wheat induced with methyl-jasmonate to stimulate expression of jasmonate-induced proteins (JIPs) revealed that heavy *B. graminis* infection protects plant rRNA from degradation in comparison to uninfected plants. Expression of transgenic BEC1054 also reduces in plant rRNA degradation in comparison to control plants. Moreover, co-agroinfiltrations of *Nicotiana benthamiana* with BEC1054 and JIP60, a RIP, showed that the effector is able to alleviate the necrotic effect of JIP60. In conclusion, BEC1054 seems to be a crucial effector in establishing *B. graminis* infection by stopping the programmed cell death initiated by ribosome inhibiting protein JIP60.

*Detection, identification and quantitation of oomycetes in imported nursery plants

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International trade in hardy ornamental nursery stock has increased the dispersal of many plant pathogens, including highly damaging oomycetes. Potted plants with soil substrates are now recognised as the major pathway of introduction of new pests and pathogens in Europe. We screened the diversity and pathogen loads in soil from potted woody plants, in roots and in water using baiting assays. Classical isolation techniques were used as well as molecular assays, including TaqMan PCR chemistry and Next Generation sequencing (NGS) approaches using Illumina MiSeq. Approximately 90% of tested plants contained at least one species of oomycete (*Phytophthora*, *Pythium*, *Phytopythium*), while 86% of asymptomatic plants tested positive for oomycetes in the growth substrate. In total, 10 *Phytophthora* species, 17 *Pythium* spp. and five *Phytopythium* spp. were isolated with classical techniques. TaqMan assays for three different loci, two of them *Phytophthora* species-specific, revealed higher pathogen average densities in roots in comparison with the soil and filter samples. NGS also showed the wide diversity of oomycetes in nursery plots. These preliminary results provide worrying evidence of plant pathogens being moved 'silently' between countries in nursery plants, and highlight the need for stronger regulation to reduce plant biosecurity risks in Europe.

Macrophomina phaseolina infection on stomatal features of leaves of *Ipomoea batatas*

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The association of *Macrophomina phaseolina* with leaf spot diseases of *Ipomoea batatas* was found to negatively affect the chlorophyll content and the plant yield. Using Koch's postulate with different concentration of the causal organism's spore suspension (10 cfu/mL; 13.3 cfu/mL; 20 cfu/mL) and anatomical techniques (a light microscopic study), the effect of the leaf spot diseases on the stomatal size, density and index was assessed. Results reveal a reduction in stomatal number and distribution thereby resulting in significant reduction in the stomatal size, density and index on adaxial and abaxial surfaces of affected plant leaves compared to healthy plant leaves. The plants leaves sprayed with spore suspension concentration of 20 cfu/mL showed the smallest stomatal size value of 1.08 µm at both adaxial and abaxial surfaces compared to the control plant leaves (healthy plant leaves) with 1.17 µm at both surfaces. Stomatal index value was 8.33/ mm^2 and 9.63/mm^2 at adaxial and abaxial surfaces compared to the control plant leaves with 11.28/mm^2 and 11.96/mm^2 respectively. Stomatal density of 3.6/mm^2 and 4.5/mm^2 at adaxial and abaxial surfaces compared to the control plant leaves with 5.0/mm^2 and 6.0/ mm^2 respectively. The results from this study also showed that degree of severity of the leaf spot diseases vary with the fungal spore suspension concentration.

A comparison of UV-C and high intensity pulsed polychromatic light sources as elicitors of hormesis in tomato fruit, *Solanum lycopersicum*

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Hormesis is a dose response phenomenon where low doses of a stress bring about a positive response in the organism undergoing treatment. UV-C hormesis has been known for over 20 years and has a broad range of benefits including increased nutritional content, delayed chlorophyll degradation and disease resistance. The beneficial effects of UV-C hormesis have been observed on many varieties of fresh produce including both climacteric and non-climacteric fruit, tubers, salads and brassicas.

Previous studies have performed postharvest treatments with conventional low-pressure mercury UV-C sources. Commercial application of these treatments has been prevented, in part, due to lengthy exposure times. Here we show that the use of a high-intensity pulsed source, rich in UV, reduces treatment times by 97.3%.

Understanding the pathogen distribution and evolution to develop resources and strategies for effective management of the finger millet blast disease in East Africa

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Finger millet is an important cereal crop for millions of people in East, Central and Southern Africa. It is highly nutritious compared to various cereals, and gluten-free. The production of this crop continues to be threatened by blast disease caused by the fungus *Magnaporthe oryzae* impacting upon food security.

The specific aim of this research is to investigate the population diversity and reproductive behaviour of the finger millet blast pathogen in East Africa in relation to time and space, and develop insights into genome level differences. Geographic focus of the study is key cropping districts of Kenya, Uganda, Ethiopia and Tanzania. A set of samples representing more than 300 isolates previously collected from Uganda and Kenya has been utilised to develop sequence-based molecular markers. Next-generation sequencing methodologies have been tested to generate genome level data. This was underpinned by bioinformatics of genome sequences available at NCBI for the blast pathogen isolates mainly from rice. We have identified molecular markers including novel loci suitable to assess the pathogen genetic diversity and identify the haplotypes from finger millet crops in East Africa. Genome sequence of a key *M. oryzae* isolate has been generated using Illumina MiSeq 600 system. These resources will enable comparative analysis of finger millet blast pathogen isolates representing historical (collected during 2000–2003) and contemporary populations along with assays to assess the reproductive behaviour.

Knowledge of the pathogen evolution and the resources generated will feed into host resistance development to enhance productivity of finger millet in East Africa.

NOTES

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