Ramularia workshop

3–4 October 2018 (Novotel London Stansted Airport)

Presentations, biographies and abstracts

Presentations

Session 1: Introduction to ramularia, emerging problems (regarding current control measures) and perspectives from abroad

1. Neil Havis, SRUC
   Introduction to ramularia

2. Andrew Gilchrist, Scottish Agronomy
   Ramularia: Scottish perspective

3. Michael Hess, TUM (Germany)
   ***Ramularia collo-cygni* in Germany – a regional perspective on occurrence, relevance and control**

4. Silvia Pereyra, INIA (Uruguay)
   Upsurge of ramularia leaf spot in Uruguay

5. Soonie Chng, Plant and Food Research (New Zealand)
   Current challenges and future prospects in managing ramularia in New Zealand

6. Ignacio Antonio Erreguerena, INTA (Argentina)
   Ramularia leaf spot in Argentina: an emerging pathogen in an emerging crop

Session 2: The challenges and opportunities of breeding for resistance to ramularia

7. Clari Burrell, SRUC
   The effects of different stages of *Ramularia collo-cygni* growth on yield formation of barley [confidential presentation]

8. Paul Bury, Secobra
   Breeding for disease resistance in barley: Challenges and opportunities when working on ramularia

9. Joanne Russell, James Hutton Institute
   Genome-wide association and genomic approaches to identifying markers for resistance to ramularia [confidential presentation]

10. James Brown, John Innes Centre
    Genetic and environmental effects on resistance of barley to ramularia and consequences for official trials and AHDB Recommended List ratings

Session 3: Short/medium-term fungicide control measures for ramularia and how to manage fungicide resistance

11. Alexandra Rehfus, BASF
    Status of fungicide resistance in the European population of the barley pathogen *Ramularia collo-cygni*: a further challenge to sustainable barley production in Europe? [confidential presentation]

12. Steven Kildea, Teagasc (Ireland)
    The challenge of controlling ramularia in Irish barley crops

13. Neil Havis, SRUC
    Fungicide resistance issues in Scotland and management of ramularia leaf spot

Session 4: Short-medium term alternative management measures for ramularia (other than fungicides and varieties), including messages to industry

14. Neil Havis, SRUC
    The potential to control ramularia leaf spot using elicitors and biological treatments

15. Michael Hess, TUM (Germany)
    A population genetic approach gives new insights into the epidemiology and control of *Ramularia collo-cygni*

16. Francois Dussart, SRUC
    The rubellins: from biosynthesis to mode of action [confidential presentation]

17. Michael Foged Lyngkjær, University of Copenhagen
    Lessons learned from analysis of gene expression and natural compounds accumulation in barley leaves infected by pathogenic *Ramularia collo-cygni*

18. Simona Radutoiu, Aarhus University (Denmark)
    [confidential presentation]

19. Bart Fraaije, Rothamsted Research
    Testing of antimicrobial properties of (endophytic) bacteria using *Zymoseptoria tritici* as a model [awaiting presentation]

Poster session

20. Andres Mäe, Estonian Crop Research Institute
    Ramularia collo-cygni has invaded barley fields in Estonia

21. Henry Creissen, SRUC
    Biological control of ramularia leaf spot using elicitors

22. Neil Havis, SRUC
    Developing a risk forecast for ramularia leaf spot to aid disease management

23. Joseph Mulhare, TEAGASC
    Ramularia leaf spot of barley in Ireland

24. Kelly Breeds, CenGen (Pty) Ltd
    Development of an improved assay for the detection of *Ramularia collo-cygni* DNA in barley tissues

Biographies and abstracts

Neil Havis
SRUC

Biography

Neil is a Senior Plant Pathologist in the Crop and Soil Crop group at SRUC and has been carrying out research on *Ramularia collo-cygni* since 1999.

His research has been focussed on the understanding of the fungal biology, disease epidemiology and the control of symptoms in the crop. Research at SRUC has produced a number of refereed papers on the fungal life cycle, diagnostics for the pathogen and also an initial paper on the fungal genome. SRUC have contributed to a number of collaborative projects on *R. collo-cygni* (CORACLE and BRCC. Research in the Crop and Soil group is ongoing.

Abstracts

1. Ramularia – an Introduction

Ramularia leaf spot (RLS) in barley is caused by the fungus, *Ramularia collo-cygni* (*Rcc*). The pathogen was first recorded in Italy in the 19th century but has now spread to many countries across Europe, the Americas and New Zealand. More recently there have been reports of the pathogen in South Africa, Tasmania and Canada. Symptoms are characterised by the 5R's and these can be used to differentiate RLS from other barley diseases. The disease appears late in the growing season and reduces yield and quality of barley. Average yield losses in the UK were estimated at 0.5 t/ha. The life cycle of the fungus is being slowly elucidated. The seed borne stage in the fungal life cycle was identified 10 years ago but the influence of airborne spores on disease epidemics is still being evaluated. Analysis of spore release events at two Sottish sites identified a significant relationship between high levels of leaf wetness in barley crops and the movement of spores. Spore levels peak late in the growing season. Although there was a relationship between spore levels and final disease levels in the crop there was no significant evidence of a green bridge effect between late harvested spring barley crops and emerging winter crops.

2. Ramularia leaf spot and resistance management

The initial outbreaks of RLS in Scotland were controlled well by the strobilurin fungicides. Later applications of fungicide (GS59) were seen to be the most effective in controlling RLS. However the field performance of the strobilurins declined rapidly between 2003 and 2005 and by 2006 they were completely ineffective. Analysis showed that all UK *Rcc* isolates examined carried the mutation conferring resistance to the strobilurins. Control was then provided by the azole

and SDHI fungicides. To protect the chemistry farmers were advised to use these fungicides in mixtures. Despite this the fungus has mutated and developed resistance to both of these groups of fungicides. The first mutations in the fungus were recorded in 2015 but by 2017 field performance had declined to minimal levels. Over the course of the last 20 years the multisite fungicide, chlorothalonil has maintained its efficacy against RLS. Studies have indicated no change in the sensitivity of the fungus to this chemical. At the moment late applications of this fungicide give the only reliable control of RLS. Research into the efficacy of other fungicide groups against RLS is ongoing.

3. Alternative control of RLS

RLS is usually controlled by fungicides but field testing of resistance elicitors against the disease has been ongoing at SRUC. Elicitors are simple polysaccharides which induce the defence responses in plants. A cocktail of elicitors was tested initially but they were not as effective against RLS as they were against other barley diseases. A maize cell wall derivative showed some promise as an elicitor I low disease years but when disease pressure was high they were less effective. More recent work has focussed on registered products and although control is variable elicitors may well be able to offer some control as part of an Integrated Pest Management programme. Elicitors in combination with reduced rates of fungicide give consistent control of RLS. A variety of physical and biological seed treatments e.g. steam treatment, heat treatments and bacterial seed inoculants, have also been tested against RLS. Again, results against RLS are variable but work is ongoing to combine the antifungal activity of all of these products into a programme that can be adopted by growers.

Andrew Gilchrist
Scottish Agronomy

Biography

Andrew was brought up on a dairy/sheep farm in the Borders. He completed a degree in Science with Industrial Studies at Napier College, Edinburgh in 1985. During his studies he did work placements with both SAC and BASF trials teams in Scotland, and worked briefly with BASF as a trials officer once he graduated. Scottish Agronomy was founded in the summer of 1985 by Huw Phillips and Andrew accepted the challenge of running the fledgling trials programme. Andrew succeeded Huw Phillips as Managing Director upon his retirement in 2006 and the Cooperative has now expanded to become the largest independent, trials based agronomy organisation in Scotland, serving 200 + farmers and covering over 60,000 ha of combinable crops and potatoes. The trials programme is one of the largest in the UK, currently extending to over 25,000 trial plots. Andrew is a member of the AHDB Barley and

Oats Committee and sits on the Scottish Variety Consultative Committees for oilseeds and cereals.

Abstract

'Ramularia – Scottish Perspective'

Scottish Agronomy are an arable advisory and trials organisation servicing 60,000 ha of combinable crops and potatoes, and with the largest trialling network in Northern UK. The barley crop is the most important arable crop for Scottish growers, amounting to 64% of the combinable crop area. The majority of this is malting spring barley grown for whisky production. Ramularia first became commonly recognised in Scottish crops in the late 90's. At this point it was well controlled by the newly emerging strobilurin chemistry. By 2004 this group of fungicides started to lose efficacy against the disease, and by 2006 provided no control over the disease. Fortunately, through experimentation, it was noted that chlorothalonil (CTL) provided good control of both Ramularia and abiotic spotting. By 2005 it had become established as a key element in protecting the crop, with the most effective timing being awn-emergence (Z49). In a series of trials conducted in 2017, azole and SDHI chemistry provided very poor control of Ramularia. The addition of CTL at the T2 timing provided excellent control and a significant increase in yield in 2 out of 4 trials. The loss of efficacy from systemic fungicides leaves CTL as the only effective treatment against the disease. Other non-systemics such as Folpet have been trialled but are relatively ineffective. The regulatory pressures on CTL mean it is imperative that other solutions are found to control the disease, whether it is by plant breeding, or new crop protection products. The potential loss of CTL would severely impact both yield and quality of barley. This may result in the crop becoming uneconomic for growers and poorer quality grain for processors.

Michael Hess

TUM, Germany

Biography

Receiving his diploma as agricultural engineer and his PhD in phytopathology from the Technical University Munich, Michael has gained 20 years of experience from the management of diverse projects inside and outside the university. His focus lies on disease surveys and designing and conducting epidemiological studies in field and controlled environment. For the past 15 years Michael has worked as a researcher and group leader in "Epidemiology and Integrated Plant Protection" at the chair of Phytopathology, TUM School of Life Sciences. A major project has been the integration of *Ramularia collo-cygni* into strategies for improved disease control. In subsequent collaborations they are conducting population genetic studies and are looking at the impact of climate change on the emerging disease.

Abstracts

Ramularia collo-cygni in Germany – a regional perspective on occurence, relevance and control

About 20 years ago the emergence of a heavy, unspecific leaf spotting complex in barley coincided with a loss of efficiency of well-established disease control strategies. The phenomenon was related to the fungal pathogen *Ramularia collo-cygni* as the biotic cause of Ramularia leaf spots (RLS). Intensive monitoring showed the German-wide occurrence throughout the life-cycle of barley and specific fungicide trials the relevance for control. The high relevance of control is also reflected by the number of fungicides being registered for control of either RLS or PLS (physiological leaf spots), the abiotic part of the spotting complex. By now about 50% of the fungicides on the German market are registered for the control of either RLS or PLS.
For a more targeted approach and optimized control in the terms of Integrated Pest Management (IPM) and in a cooperation between the university and the Bavarian State Research Center for Agriculture (LfL) the new and emerging disease complex was integrated into an established, threshold based decision support system.
On the basis of the high presence and relevance of *R. collo-cygni* found in the monitoring, the improved strategy manages the disease by choice of fungicide, timing and dose respecting the general disease pressure according to the Gerstenmodell Bayern.
The success of the strategy is relying on the availability of effective compounds. Since 2015 we observe an increasing loss of efficiency. The registration is based on 8 AI s from 4 classes, but in the meantime only 2-3 give satisfactory control and only 1 good and reliable control. This is putting the current strategy at question and is making new approaches and a better understanding of the pathogen biology necessary for future control.

A population genetic approach gives new insights into the epidemiology and control of *Ramularia collo-cygni*

Research on the *Ramularia collo-cygni* is facing many limitations due to difficult accessibility for traditional testing systems. In a population genetic approach we generated a new reference genome and sequenced 18 isolates with diverse global origin and from different hosts.
In the genomic comparison to well-studied “model” pathogens to *R. collo-cygni* we calculated a seperation from its nearest sequenced relative *Zymoseptoria tritici* about 27 million years ago, reflected in a remarkably high divergence on protein level. Contrary to our expectations the divergence is not reflected in the comparison of predicted secreted proteins and putative effectors.
Admixture analyses of the 19 sequenced isolates show that *R. collo-cygni* is world-wide genetically uniform and that samples do not show a strong clustering on either geographical location or host species.

Analysis of linkage disequilibrium shows that in the world-wide sample set there are clear signals of recombination and thus sexual reproduction, however these signals largely disappear when excluding three outliers samples, suggesting that the main global expansion of *R. collo-cygni* comes from mixed or clonally propagating populations. We further analysed the historic population size (Ne) of *R. collo-cygni* using Bayesian simulations. The results support the hypothesis of a recent expansion and historic large effective population size.

The uniformity of the population is supporting seed transfer being a part of the global epidemics and the large effective population size could play a role for the quick adaptation to fungicides.

Unlike in many other plant pathogens, further analysis show a very homogeneous genome for *R. collo-cygni*. In consequence the plant side and environmental factors are of great importance for understanding the host pathogen interaction and for developing future, sustainable control.

In an ongoing project on the impact of climate change on Bavarian agriculture (BayKlimaFit) we detected an increasing contamination with *R. collo-cygni* in archived barley samples and could show the effect of drought on a diverse barley panel in their resistance.

Silvia Pereyra
INIA, Uruguay

Biography

Silvia conducts an applied research program in plant pathology with emphasis in the integrated management of wheat and barley diseases. Her main objectives have been: to understand the epidemiology of the economically important diseases affecting wheat and barley in Uruguay and to develop adequate strategies for disease management, integrating production practices, with emphasis on plant resistance and chemical control strategies. Silvia is also involved in training and teaching human resources in the management of cereal diseases.

Abstract

Upsurge of Ramularia leaf spot in Uruguay

Silvia Pereyra[1]; Carlos Pérez[2]; Cintia Palladino[2]; Neil Havis[3]

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[3] SRUC Scotland's Rural College, West Mains Road, Edinburgh, EH9 3JG, Scotland, UK

Ramularia leaf spot (RLS) caused by *Ramularia collo-cygni* (*Rcc*), has become one of the main constraints to barley production in Argentina and Uruguay in the last eight years. Grain yield losses as high as 70% have been reported in susceptible cultivars. The emergence of RLS as a major disease in the last six years has redirected coordinated

research efforts to advance on the understanding of *Rcc*, its epidemiology and best management strategies in our production systems. Based on *Rcc*-DNA quantification, RLS is widely present in seed lots, especially after epidemic years (i.e. 2012, 2016, 2017). Leaf wetness during elongation associated with stress factors in the crop has an impact on disease onset. Commercial cultivars and advanced lines were characterized under intermediate to high disease pressure in nurseries and field trials. Most of the commonly grown cultivars were intermediate to susceptible to RLS. Best timings for fungicide application for RLS control in epidemic years and application at stem elongation (ZGS 32 to 39, depending on the infection severity of other common leaf diseases, especially both forms of net blotch) and awns-peeping (ZGS 47-49). Fungicides containing SDHI (isopyrazam + azoxystrobin and fluxapyroxad + pyraclostrobin + epoxyconazole) and combinations with prothioconazole were the most effective fungicides in controlling RLS improving yield and grain quality. Chlorotalonil is widely used in combination with mixtures of triazoles + strobilurins. Strobilurins are not effective in controlling RLS. Yet, the most efficient and economic options may vary in years with moderate epidemics. Results from these studies emphasize the importance of analysing seedlots for *Rcc* and suggest that both fungicides and timing are major factors in optimizing RLS control in the short to medium term. INIA´s breeding program have a few advanced lines consistently categorised as moderately susceptible to moderately resistant to RLS.

Soonie Chng
Plant and Food Research, New Zealand

Biography

Dr Soonie Chng is a Plant Pathologist and a Programme leader for the Vegetables sector at The New Zealand Institute for Plant & Food Research Ltd. Soonie’s research expertise is in arable and vegetable pathology, with a primary focus on the management of both airborne and soilborne diseases, such as cereal rust diseases, take all of wheat and root rot of vegetables. She works closely with the New Zealand cropping industries (e.g. Horticulture New Zealand and Foundation for Arable Research) and collaborates with international peers on management of major crop diseases. Her current research includes a trans-Tasman collaboration determining New Zealand cereal rust pathotypes, development and co-innovation of a risk predictor tool for root rot of peas with Process Vegetables New Zealand, and the development of Ramularia leaf spot management strategies with the cereal industry. Lately, her research focus has expanded to include management of myrtle rust, a recent incursion into New Zealand.

Abstract

Current challenges and future prospects in managing Ramularia leaf spot in New Zealand

Soonie Chng[1], Rob Craigie[2], Rachael Warren[1], Joanne Drummond[2], Shirley Thompson[1], Ruth Butler[1] and Suvi Viljanen[1]

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Ramularia leaf spot (RLS) of barley was first recorded in New Zealand in 1977 and since then, has become increasingly common. Today, RLS is considered one of the key diseases affecting the country's barley production, reducing grain yield by up to 30%. So far, application of fungicides has been the only measure used in New Zealand to control RLS. However, in common with Europe, New Zealand growers have experienced difficulties in controlling the disease in recent years, even with fungicides, and are concerned by the significant yield losses caused by the disease. Insensitivity to quinone outside inhibitors (QoI) and both insensitivity and reduced sensitivity to succinate dehydrogenase inhibitor (SDHI) fungicides by *Ramularia collo-cygni* was confirmed in New Zealand in the 2017-18 season. Although demethylation inhibitor (DMI) based fungicides are currently effective in controlling RLS, they remain at risk of becoming ineffectual, given that resistance to this group of fungicides in *R. collo-cygni* has already occured in other countries, such as Scotland and Germany. Attempts to develop realiable varietal resistance ratings for the barley cultivars grown in New Zealand has also proven difficult. Many factors including varying crop maturity (early vs. late maturing), amounts of seedborne and airborne inoculum, presence of other diseases and varying seasonal susceptibility have all contributed to the inconsistent ratings. Recent analyses of *R. collo-cygni* DNA in pre-sown and harvested grains from the same plants have shown that, despite the absence of *R. collo-cygni* DNA in the pre-sown seed, the pathogen's DNA was detected in all the offspring (harvested grains) when the disease was present in the field. This suggests that airborne inoculum may contribute substantially towards the infection of the offspring (harvested grains) in the field. When similar seed lines with known amounts of seedborne *R. collo-cygni* DNA were sown at two different sites, RLS developed on the plants at only one of the sites. While there is still a lot to learn about the epidemiology of RLS, we aim to restore the yield potential of barley and minimise fungicide inputs to enhance the financial returns for New Zealand arable growers. Recent and current research on RLS in New Zealand will be discussed.

Ignacio Antonio Erreguerena
INTA, Argentina

Biography

Ignacio is a biologist and plant pathologist graduated at the National University of Mar del Plata, Buenos Aires, Argentina. He is currently doing his PhD on *Ramularia collo-cygni* epidemiological and genetic variability aspects at the Postgraduate School of the Agricultural Sciences College at the same university.

Ignacio has worked in the teaching of Plant Pathology collaborating in courses, undergraduate and postgraduate thesis. He is a phytopathologist and develops research on corn, sunflower, oats, wheat and barley diseases caused by oomycetes, bacteria and fungi on diagnosis and integrated control. He is in charge of the Plant Disease Diagnosis Service and he is part of the research group on Plant Pathology and Disease Resistance of the National Institute of Agricultural Technology.

Abstract

Ramularia leaf spot in Argentina: an emerging pathogen in an emerging crop

Biologist Ignacio Erreguerena

National Institute of Agricultural Technology (INTA Balcarce)

At this moment in Argentina the barley area is about 1 million hectares distributed in and around Bs As province. Almost all grain goes to local malting and brewing beer companies. Malting barley grain that doesn´t reach to malting quality standards is exported to Saudi Arabia, India and Brazil for beer production and animal feeding. About 40 years ago the barley area was about 200 thousand hectares but from 1999 with the introduction of varieties with higher yield and malting quality the barley crop began an expansion to areas where wheat was produced. In 2002 Ramularia leaf spot (RLS) was first diagnosed (Khier et al, 2002) but was in the year 2012 that we had a great outbreak of RLS. These were the three main diseases until RLS appear, Barley net blotch (Drechslera teres sf. teres), Barley Scald (Rhynchosporium secalis) and Bipolaris spot (Bipolaris sorokiniana). The first two are the most prevalent diseases. First, we described the symptoms, and when they can appear. We got flowering spots and grain filling spots, and early spots at tillering stages. We learned how to isolate the pathogen since is a little bit complicated for its low growing rates. Then we use PCR reaction for an early diagnosis and we used it to measure the prevalence of Ramularia in many barley regions of south BSAS province in 2015. We observed a 60% Ramularia prevalence and that all barley varieties are susceptible to RLS.

To know the distribution of Ramularia cases and levels among years. We create a digital platform where farmers and researchers can upload disease data on real time basis called ReTSaVe. It allowed us to observe that RLS high levels cases have been increasing among years being the most important disease in the year of 2017 once again. To establish the protection window to find the best carboxamides spraying time for disease control we developed a protocol that consists in progressive protection and deprotection with fungicide among barley development stages. We determined the protection window from Z33 to Z49 stage (Erreguerena, PhD unpublished). To support the protection window we

had many validation trials where you can see that single or double spraying with carboxamide that includes flag leaf stage had the lowest RLS intensity (Farengo, 2015).

In Argentina barley crop is about 80% of one variety called Andreia. So, disease management strategies depend mainly on fungicides. We developed a fungicide strategy protocol that includes RLS in it and used it in a barley protection network where we could test the different strategies combination in different locations. Seed coated fungicides (SCF) and defense promoters. Disease incidence threshold for non RLS diseases and flag leaf stage for RLS. We observed that SCF plus flag leaf spraying (with carboxamides) was the best strategy regarding all diseases but also the strategy that combines a 20% incidence threshold spraying plus flag leaf spray was good also. In those years with high disease pressure we can combine these two strategies.

In other field trails the use of IPM strategies like delayed planting date and wheat rotation we avoid other diseases, so we can work just with RLS and test if clorothalonil (active ingredient not registered for barley yet) could improve disease control performance of mixtures with prothioconazole and or carboxamides. Indeed, adding clorothalonil to the mixtures improves RLS control.

The major impact of RLS on barley production In Argentina is on the grain quality (weight and size) which is important for beer or malting companies. Regarding yield we've reached up to 1.8 thousand kilos per hectare but around 1.0 thousand kilos per hectare is the average losses. Future prospects on RLS research are: studies on pathogen variability and sensitivity to fungicide active ingredients, disease forecast – epidemiological modelling, Studies on the importance of Inoculum sources and create awareness on the risk of Ramularia and other pathogens to turn resistant or loss sensitivity to fungicides.

Clari Burrell
SRUC

Biography

Clari is a PhD student at SRUC and the University of Edinburgh supervised by Professor Ian Bingham, Dr Neil Havis, and Dr Steven Spoel. Her project title is 'Understanding interactions between *Ramularia collo-cygni* and barley leaf physiology to target host resistance and disease control strategy'. Clari has been investigating the period of growth of *R. collo-cygni* within barley prior to the appearance of visible Ramularia Leaf Spot symptoms as little is currently known about what effect this period may have on the host plant.

Abstract

The effects of different stages of *Ramularia collo-cygni* growth on yield formation of barley

The aim of this work was to ascertain whether the presence of *R. collo-cygni* in its host plant during the long asymptomatic phase could impact yield. Before the appearance of visible symptoms Chlorophyll Fluorescence Imaging did not detect any damage to the photosynthetic apparatus sufficient to effect yield in either seedlings grown under controlled environment conditions or field plants. Further experiments measuring the interception of light by healthy (green) tissue area indicated that the loss of green leaf area due to Ramularia Leaf Spot symptoms is sufficient to account for the yield loss caused by this disease.

Paul Bury
Secobra

Biography

Paul Bury has been a commercial barley breeder for over 30 years, working initially for New Farm Crops in Lincolnshire, which later became part of Syngenta. Being part of the team that bred many successful varieties of barley such as OPTIC, NFC TIPPLE, QUENCH, FLAGON, SY VENTURE and LAUREATE and introduced the world to CMS Hybrid barley, Paul has participated in previous collaborations on *Ramularia collo cygnii* helping with the more practical aspects of investigating this difficult pathogen. Paul is now setting up a new malting barley breeding programme in Lincolnshire for French company Secobra Recherches.

Abstract

Breeding for disease resistance in barley: Challenges and opportunities when working on ramularia

Any breeding efforts to improve disease resistance have to start with a good understanding of the pathogen and what variation in susceptibility exists in nature .When Ramularia Leaf Spotting was first noticed in Scotland in the mid 1990's neither were well known , so a mix of formal and informal meetings of experts were arranged. As breeders we were lucky to spend time with expert pathologists from Scotland, Germany, Norway, Austria and Ireland. Understanding and identification of the disease improved, but we were not able to set up reliable phenotyping methods, or identify stable and consistent sources of resistance. A better funded project followed in 2004 and the CORACLE group improved knowledge a lot, although again , searches for varietal resistance uncovered few leads. From over 20 years' experience Paul Bury outlined the main issues when trying to breed for improved resistance to Ramularia.

- Symptoms are difficult to recognise, often occurring in conjunction with other diseases

- There are no reliable inoculation methods that work on adult plants in the field. That makes it very difficult to screen material for resistance and susceptibility
- Large international studies have looked at thousands of varieties, old and new, and to date none have reported any strong sources of resistance. These studies have often concluded that environmental variation in their data is actually greater than the variation due to variety.
- It is generally agreed that any resistance to ramularia that exists currently is polygenic , with many resistance factors giving small effects

At the end of the presentation Paul suggests that a co-ordinated approach to resistance breeding could work where partners shared what they consider to be reliable phenotype information on as many lines as they have (Disease scores from trials and nurseries) and aim to combine this with Genotype information to try to calculate resistance 'values' which could be used to evaluate new material for potential resistance.

Joanne Russell

James Hutton Institute

Biography

Joanne Russell is a member of the Barley Genetics group at the James Hutton institute in Dundee and her current research explores diversity, at the gene level using genomics and informatics technologies, in the barley genepool. As part of the team she has worked on several translational projects, focused on specific targets such as such as resistance to Fusarium ear blight (INSPYR, RD-2007-3453) and Ramularia (CORACLE, RD-2007-3441) and is currently working on the BBSRC funded IMPROMALT project (BB/K008188/1, RD-2012-37760) to improve the malt extract potential of winter. Joanne has contributed the barley sequencing projects (Mascher et al., 2017) and with colleagues has been involved in the development of the 9K and 50K barley SNP platforms (Comadran et al., 2012; Bayer et al., 2017).

James Brown

John Innes Centre

Biography

James Brown is head of the Department of Crop Genetics at the John Innes Centre in Norwich. His research interests are in pathogen adaption to crops and durable control of disease, especially by breeding for durable resistance. He has worked on Ramularia leaf spot of barley since 2002, particularly on genetics of resistance and genotype-by-environment interaction. He coordinated a LINK project

on Ramularia from 2009 to 2013. He has been the pathology specialist on the Wheat Recommended List Committee since 2010.

Abstract

Genetic and environmental effects on resistance of barley to ramularia and consequences for official trials and AHDB Recommended List ratings

James K M Brown
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The *mlo11* mutation of the wild-type *Mlo* gene is now widely used in spring barley for strong, durable resistance to powdery mildew and may be present in the majority of UK varieties. However, it also makes barley more susceptible to several non-biotrophic diseases (reviewed by Brown & Rant 2013, Plant Pathology), notably Ramularia leaf spot (McGrann et al. 2014, Journal of Experimental Botany). In experiments on seedlings in controlled environment rooms and on adult plants in a polytunnel, *mlo* alleles increased susceptibility to Ramularia by at least 60%. The association of *mlo11* with more severe Ramularia was also apparent in progeny of Power x Braemar and Cocktail x Decanter, but the effect variable, being generally stronger at sites in Bavaria than in Scotland in a series of field trials in 2013. Physiological research supports the hypothesis that the development of Ramularia is strongly influenced by genotype-by-environment (GxE) interactions (McGrann et al. 2018, Annals of Botany). Symptoms were enhanced by abiotic stress in most varieties tested and manipulation of peroxide levels in either direction tended to increase symptoms. These effects were stronger and more consistent in Ramularia-susceptible varieties than in those with partial resistance. This suggests that partially resistant varieties might be less sensitive to GxE effects but QTL mapping has detected no gene apart from *Mlo* which increases resistance. Experiments on a range of barley mutants indicated that high infection by the pathogen, *Ramularia collo-cygni*, is required for disease development but some genes suppress the transition of the fungus from endophyte to necrotrophic parasite.
In field trials of the Power x Braemar F1 doubled-haploid population, there was significant variation in Ramularia within both the *mlo11* and *Mlo*$^{+}$ groups, although *mlo11* lines had substantially more disease on average. This implies that breeders should be able to select lines with improved Ramularia resistance and to combine it with *mlo11*.
However, the GxE effect was reflected in AHDB Recommended List trial data in which the interaction between varieties and sites was so strong that it dominated the main effect of varieties. I have recommended that (1) official and RL trials for Ramularia resistance should be held at sites where this is much the most important disease, so that inference from other pathogens is minimised, (2) ratings should be based only on scores from the best sites, with a wide difference between resistant and susceptible control varieties, and (3) further training of trial managers will assist accurate scoring of Ramularia.

In the early years of Ramularia leaf spot, *mlo11* varieties were consistently more affected than other lines. In the light of the Power x Braemar data, it is conceivable that breeders are already culling *mlo11* varieties which are highly susceptible to Ramularia. If so, it would greatly help us to control Ramularia further if we understood why *R. collo-cygni* turned from being a harmless endophyte to an aggressive pathogen around the mid-1990s.

Alexandra Rehfus
BASF

Biography
Dr. Alexandra Rehfus obtained her PhD degree in Agricultural Science in 2018 from the University of Hohenheim in collaboration with BASF. During her studies she focused on biotechnology and plant protection with a special interest in phytopathology. As part of her internships, diploma thesis and PhD studies Alex had the opportunity to work on several molecular methods (e.g. transformation) in a range of pathogens including the rust fungi, *Zymoseptoria tritici, Pyrenophora teres, Erwinia amylovora* and *Ramularia collo-cygni.* Alex is now working as a post-doctoral researcher in fungicide resistance research in BASF SE, Limburgerhof, Germany. The main focus of her work is the emergence of fungicide resistance in cereal pathogens having already published several peer reviewed papers on the topic.

Steven Kildea
Teagasc, Ireland

Biography
Steven Kildea currently a senior research officer at Teagasc. His research programme focuses on the development of sustainable disease control strategies for Irish arable and potato crops. To achieve these goals, they incorporate detailed fungal population studies with applied field trialling. Currently *Zymoseptoria tritici* on wheat, *Rhynchosporium commune*, *Ramularia collo-cygni*, *Pyrenophora teres* on barley and *Phytophthora infestans* on potatoes are the key pathogens investigated in our lab.

Francois Dussart
SRUC

Biography
Francois' work on *Ramularia collo-cygni* started during my PhD in which he analysed the genome of the fungus to identify gene clusters associated with secondary metabolism. Secondary metabolites such as the rubellins toxin produced by *R. collo-cygni* are often important determinant of the host-pathogen interactions. The findings of this research were presented on a poster during the 2nd International Workshop on Barley Leaf Diseases in Rabat (April 2017) and recently

reported in Molecular Plant-Microbe Interactions (Vol. 31, p.962-975). HIs current research focusses on understanding the host-pathogen interactions at the cell level by investigating the mode of action of the rubellin D toxin *in planta*.

Michael Foged Lyngkjær
University of Copenhagen

Biography
Michael F. Lyngkjær has since 2009 been an associated Professor at the section of Plant Biochemistry in the Department of Plant and Environmental Sciences, University of Copenhagen. Here he teaches and act as research group leader. His work focuses on how natural compounds are synthesized and accumulating in cereals when exposed to stresses, including their role in defence against fungal pathogens and adaptation to severe environmental growth conditions. Before coming to KU, Michael was a researcher at Risø National Laboratory working with barley powdery mildew disease. In 1996, Michael obtained a PhD degree in plant pathology from the Danish Royal Veterinary and Agricultural University.

Simona Radutoiu
Aarhus University, Denmark

Biography
The aim of Simona's research is to understand the molecular mechanisms underlying microbial association with plants. Studies of the interaction between barley and *Ramularia collo-cygni* are focused on fungal components that are regulated during the interaction with barley host.

Bart Fraaije
Rothamsted Research

Biography
Bart Fraaije at Rothamsted Research leads the fungicide research team that has been studying mechanisms for fungicide resistance in arable crop pathogens using the latest advances in phenotyping and genomic tools. The information gained from this work has been used to devise strategies for an optimal, more sustainable disease control while minimising the risk for fungicide resistance development using appropriate crop protection inputs, agronomic practices and host resistance. The current research objectives are: 1) to understand how control agents work (mode of action of new active ingredients, plant defence activators and biocontrol agents) and to identify potential mechanisms employed by pathogens enabling them to cope with such agents; 2) to develop molecular tools/diagnostics to understand the role of inoculum sources (seeds, stubble and alternative hosts), to

study the epidemics of fungal diseases and to monitor the evolution of specific traits in pathogen populations (fungicide resistance and virulence factors); and 3) to evaluate novel strategies for disease control and fungicide resistance management in commercial crops. Bart is also a member of the UK Fungicide Resistance Action Group (FRAG-UK), a forum to discuss and promote practical guidance on the status and management of fungicide resistance issues in the UK.