

Blotches and spots

Understanding blemishes of faba bean

Over the last decade, grain legumes such as faba beans have been cultivated widely in the Nordic and Baltic countries. Faba bean is the world's second-highest grain-yielding legume, yet the occurrence and spread of specific diseases of faba bean grown in Europe have not been studied in great detail. Dr Biruta Bankina and colleagues Dāvids Fridmanis, Gunita Bimšteine, Jānis Kaņeps, Ieva Plūduma-Pauniņa, Zinta Gaile, Līga Paura, Fred L Stoddard, and Ance Roga, at the Latvia University of Life Sciences and Technologies, have been examining specific fungal pathogens of faba bean. The team has identified species of *Botrytis* that cause chocolate leaf spot and an *Alternaria*/*Stemphylium* complex, causing leaf blotch of faba bean. Identification of these pathogens will lead to further understanding of how to breed unblemished beans.

Faba bean (also known as broad bean, *Vicia faba*) is an important legume crop known for its benefits as a source of protein for food and as green manure in fields. In Europe, compared with farming of cereals and oilseed rape, which have the highest profit margin, the cultivation of legumes as a popular crop of choice has been limited. Only in the last decade has the importance of growing legume grain crops increased rapidly in Europe.

However, like many crops, faba bean can be devastated by several diseases, which greatly affect yields. There has recently been an increased interest in identifying specific pathogens of faba bean in order to understand how to better control the occurrence and spread of diseases. Dr Biruta Bankina and her team at the Latvia University of Life Sciences and Technologies, in cooperation with Helsinki University and the Latvian Biomedical Study and Research Centre, have recently identified fungal species that cause leaf diseases of faba bean. A deeper understanding of the pathogens and their lifestyle on the plant will enable better ways to control these diseases.

SPILL THE BEANS ON FABA

Faba beans are well known as an important source of protein in human and animal diet. As a legume, these plants also have the specialised ability to change atmospheric nitrogen into a more useful form for plants, through a close association with root-living bacteria. This symbiotic relationship allows the soil to be rich in nitrogen compounds which are necessary for healthy plants. In this way, legumes such as faba bean are used as green manure by farmers in between rotation of cereal crops.

Despite this, farming of faba beans and other legumes has not been prevalent

in Europe until the last decade. Since then, the cultivation practices in Nordic countries changed, resulting in a 15-fold increase in faba bean growth. Increased cultivation of this crop is also accompanied by threats that reduce its yield; disease is one of the most important factors affecting legume yield worldwide. Although the application of fungicides (chemicals that kill fungi) partially reduces the spread of some pathogens (fungi that cause disease), a more effective disease-management strategy is essential in order to maximise yield. Genetic identification of specific pathogens of faba bean and studying their mode of infection in the plant will advance progress in this field.

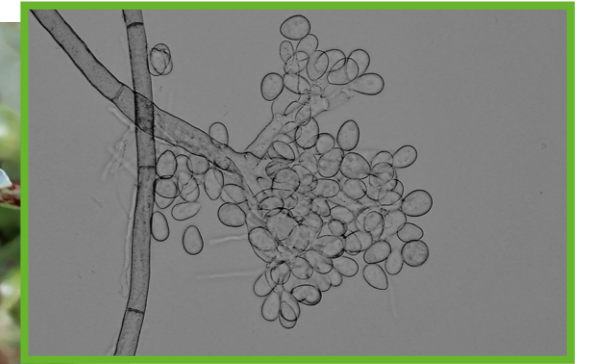
BEAN BLEMISHES

Plant-pathogen interactions are described as an arms race, with each organism trying to outcompete the other. A plant gets diseased when it is attacked by a specific pathogen, and the plant cannot defend itself successfully through its various immune responses. Failure to recognise the pathogen, or the pathogen overcoming or suppressing the barriers and defence pathways put up by the plant, results in a disease outcome. For a leaf-attacking pathogen, symptoms appear as blemishes on the leaves.

Plant disease occurrence not only depends on the specific pathogen and the plant but is also influenced by cultivation practices and the climate. For example, while some pathogens proliferate in high humidity and moisture, drought stress weakens the plant, thereby allowing pathogens to take advantage. Seasonal spread of pathogens is therefore quite common. In addition, intercropping, ie, growing two or more crops in the same field, reduces pathogen spread, while high cropping density increases disease spread.



Symptoms of chocolate spot on faba bean.



Botrytis spp. is a necrotrophic fungus that causes chocolate spot disease in many plant species.

At the Latvia University of Life Sciences and Technologies, Dr Biruta Bankina aims to understand the specific pathogens that cause leaf diseases in faba bean. Her team has been investigating the science behind the occurrence of chocolate spot disease and leaf blotch, as well as rust and downy mildew, on faba bean. Each of these diseases is caused by unique pathogens, and the focus of Dr Bankina's research is to identify the specific organisms that cause each of these diseases.

SURVIVAL BY DEATH

One of the most common diseases of faba bean is chocolate spot, caused by different species of the fungus *Botrytis*. *Botrytis* is a necrotrophic fungus, which means that it needs to penetrate tissues

and kill the cells in order to survive. Necrotrophic fungi obtain their nutrition by consuming the contents of dead plant cells. The fungus attacks soft plant tissue and produces enzymes that digest the plant cell walls, allowing it to enter cells and kill them via toxins. *Botrytis* thrives when there is high humidity and moisture, causing maximum damage to the plant.

Dr Bankina's team performed a study where chocolate spot was detected on faba bean leaves. Over a five-year period, chocolate spot symptoms were identified

on faba bean leaves every year. The spots were either small, reddish-brown, or large blotches of dead tissue with red margins (hence the name, chocolate spot). Disease severity was associated with months of heavy rainfall. Across three different cultivars of faba bean tested, none could be deemed as being resistant, although there were differences in disease progression.

Given that there are a number of *Botrytis* species affecting different plants, the researchers wanted to identify the specific species that infected faba bean in Latvia. In recent work, Dr Bankina's team has extended

Dr Biruta Bankina and her team have recently identified fungal species that cause leaf diseases of faba bean.

these findings to identify four unique *Botrytis* species that infected faba bean leaves and caused chocolate spot disease. As the appearance of *Botrytis* species alone cannot be used to distinguish them, the team used DNA sequencing to identify these unique fungal species (*B. fabae*, *B. cinerea*, *B. fabiopsis*, and *B. pseudocinerea*). Preliminary results from the study indicated that of these four, *B. fabiopsis* may be the least infectious on faba bean leaves. Identification of these unique *Botrytis* species will allow dissection

of the sensitivity of different faba bean cultivars to each of these pathogens.

BLACK BLOTCHY BLIGHT OF BEAN

In addition to chocolate spot disease, faba bean is affected by another fungal disease, resulting in black leaf blotches, also referred to as blight. Fungi from two different genera – *Alternaria* and *Stemphylium* – are known to cause the same disease on faba bean. Climatic conditions with high humidity and moisture and not very high temperatures allow these fungal pathogens to thrive.

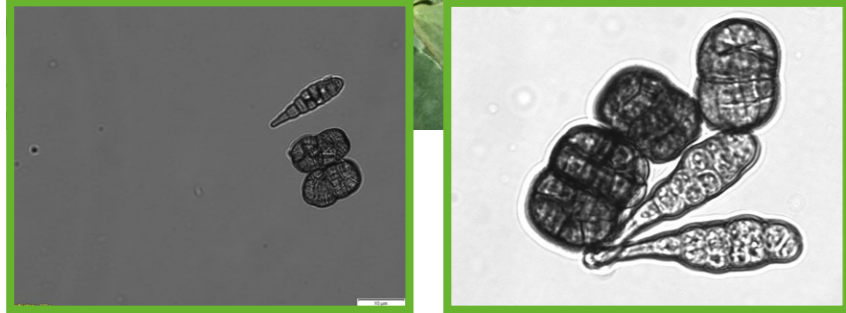
When Dr Bankina's team investigated diseases in faba bean over five years, the occurrence of leaf blotch was also common every year. Large, grey-to-black, dead leaf blotches were observed that, by eye, appeared symptomatic of infection by either *Alternaria* spp. or *Stemphylium* spp. The scientists observed the infected leaves under a microscope and were able to detect

spores of both *Alternaria* and *Stemphylium* together in the same leaf blotch. Although they did not find much

difference in disease sensitivity of different bean cultivars, leaf blotch had the most severe symptoms compared with other diseases of faba bean. Understanding this disease complexity and the collaboration between two fungal species to cause this disease poses a challenge, as each species would have a different mode of infection, making it harder to breed plants for disease resistance.

FURTHER FUNGI ON FABA

In addition to chocolate spot and leaf



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blight, faba bean also gets affected by disease causing rust and downy mildew. Rust, caused by the fungal species *Uromyces viciae-fabae*, is spread through its spores by the wind. Rust can cause huge devastation of faba bean and thrives under humid and warm conditions. Symptoms appear as small, orange-brown pustules on leaves surrounded by a yellow halo. Severe infection causes premature leaf fall and reduction in seed size.

Downy mildew is another disease affecting faba bean, caused by the microorganism *Peronospora viciae*. The microorganism enters leaves through stomata (microscopic openings that allow leaf transpiration) to infect the plant, and appears as leaf blotches with a red tint on the underside of the leaves. The microorganism could also result in stunted growth of seedlings.

Dr Bankina and colleagues identified the presence of rust and downy mildew on faba bean grown in Latvia over a five-year



period. While rust appeared in all but one year during pod development, downy mildew was only detected in a single year when there was low temperature and high humidity. Furthermore, while there was no cultivar of faba bean that was resistant to these pathogens, they exhibited differences in disease sensitivity. Investigating the lifestyle of both rust- and downy-mildew-causing fungi and their mode of attack on faba bean will reveal insights into targets of disease control.

CONTROLLING FABA BEAN DISEASE

All the above diseases of faba bean were tested for their sensitivity to fungicides by Dr Bankina and her team. A combination of two active ingredients of fungicides caused partial reduction of chocolate spot, leaf blotch and rust, but not of downy mildew, as it is known that these fungicides do not target the microorganisms causing downy mildew. Although fungicides caused a decrease in disease occurrence, this is not an effective way to control faba bean diseases, as it is not a long-lasting solution. Moreover, fungicides are not favourable to the ecosystem, as they cause soil pollution. Therefore, finding new targets for disease resistance in faba bean is a priority. Identification of specific pathogen species causing each of these diseases and their mode of action on faba bean will no doubt lead to unravelling the genetic basis of disease occurrence. Further work will inevitably result in cultivation practices for healthy faba bean.



The team identified the presence of rust (left) and downy mildew (right) on faba bean grown in Latvia.

Behind the Research



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Research Objectives

Dr Bankina and her team study plant diseases affecting faba bean (*Vicia faba* L.), one of the most important grain legumes.

Detail

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Bio

Dr Biruta Bankina is professor at the Faculty of Agriculture, Latvia University of Life Sciences and Technologies. The most important directions of her research are fungal diseases of crops, their life cycles, and biological traits of their causal agents.

Dr Zinta Gaile, professor, works at the Institute of Soil and Plant Sciences, Latvia University of Life Sciences and Technologies. She has worked as a principal researcher in several projects studying factors affecting field crops' yield formation.

Dr Fred Stoddard, from the Department of Agricultural Sciences at the University of Helsinki, Finland, is one of Europe's leading legume scientists. His research centres on faba bean and includes genomics and genetics, and agronomy and cropping systems.

MSc Ieva Plūduma-Pauniņa, agronomist, works at the Research and Study farm 'Peterlauki', Latvia University of Life Sciences and Technologies. She has worked as researcher in several projects, for example, ERANET SusCrop 'LegumeGap' (2019–2022).

Dr Dāvids Fridmanis is Senior Researcher at the Latvian Biomedical Research and Study Center, Head of the G-Protein-Related Receptor and Exotic Microbiome Research Group.

Dr Līga Paura is professor in Bioinformatics at the Faculty of Information Technologies, Latvia University of Life Sciences and Technologies. She has long experience in agricultural data analysis including crop and animal sciences.

MSc Ance Roga is research assistant and PhD student with experience in molecular biology. Her background includes participation in various research projects involving fungal DNA isolation and various

genetic analyses of fungal isolates.

Dr Gunita Bimšteine, senior researcher, works at the Institute of Soil and Plant Sciences, Latvia University of Life Sciences and Technologies. She has gained experience in field plant pathology area in several projects on fungal diseases in winter wheat, field beans and vegetables.

MSc Jānis Kaņeps, researcher and PhD student at the Agriculture faculty, Latvia University of Life Sciences and Technologies. Main fields of interest are diversity and biology of *Botrytis* spp., *Pyrenophora tritici-repentis* and other fungal pathogens.

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Collaborators

- Latvian Biomedical Research and Study Centre
- University of Helsinki

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Personal Response

Once you have identified the specific pathogens causing each disease in faba bean, what will be the immediate priority for your work?

“ We still have a lot of unanswered questions because the supposed spectrum of pathogens is significantly larger than described before. The next step is to clarify the virulence differences between known and unknown pathogens, their life cycle, and sensitivity to fungicides. Another interesting question is the interactions between different fungus, including pathogens, possible endophytes, etc, which are components of plants' biome.

Can the identification of disease resistance genes in faba bean lead to the generation of healthy plants through classical breeding methods?

It can, but so far it hasn't, so we don't know. We may need to look more widely than just at 'disease resistance' genes – the resistance (such as it is) may be associated with a more general stress response (there is a hint that way from GWAS in another project). If we need to alter its expression, that may require deeper technology such as gene editing rather than simple classical breeding. ”