Clubroot disease is a condition that affects oilseed rape crops and other brassica vegetables such as cabbage or broccoli. It is caused by a microbial pathogen called *Plasmodiophora brassicae* that lives in the soil and can enter the root systems of plants. Once it has managed to infect a plant it multiplies inside the cells of the host, as it grows the roots swell and distort into a large, unshapely gall – hence the name clubroot. These galls place an enormous drain on the host and their growth interferes with the normal development of the plants vasculature, leading to a breakdown in the transport of water from the roots to the above-ground plant. Overwhelmed by the diversion of nutrients into the clubroot gall and unable to properly take up water from the soil, infected plants will wilt and wither. Millions of spores are then released from the rotting gall back into the soil, where they can lie dormant for many years until the opportunity to infect a new susceptible host arrives. The production of oilseed rape for vegetable oil and animal feed is a multibillion euro industry, the cost of clubroot can lead to 15% losses in harvests in some years. Some native brassicas have evolved natural resistance to clubroot disease, the identification of resistance genes from these varieties and their breeding into crop cultivars has been the most important way of tackling the problem so far.

The resistance genes that recognise infection by *P. brassicae* or other microbial pathogens typically have a common structure. They encode proteins that have domains for interacting with and recognising pathogen proteins and domains that transmit this recognition event into an alarm signal. The alarm signal is converted into a defence response with the synthesis of anti-microbial chemicals or the reinforcement of barriers between cells to limit pathogen spread. The relay of the alarm signal following the first contact between the pathogen and the host proteins involves the physical interaction of different signalling components – forming complexes, modifying each other, cascading the signal out into the cell. A great deal of information has been collected about how different disease resistance proteins function at the physical, molecular level. In this project we want to investigate the defence mechanisms for clubroot disease resistance that are mediated by *RPB1* (*Resistance to Plasmodiophora brassicae 1*), a gene that has been identified in *Arabidopsis thaliana*.

We screened 142 different accessions of Arabidopsis collected from around the world, infecting them with a European strain of *P. brassicae* and scoring the disease development. We found 11 clubroot resistant accessions; genetic markers on a region of chromosome 1 correlated with this resistance. Alongside these markers, in the genomes of resistant accessions, we found the *RPB1* gene; we used genome editing tools, the CRISPR/Cas9 system, to delete this gene and generated mutants that were now completely susceptible to *P. brassicae*. The protein that *RPB1* codes for is unlike anything that has previously been characterised in plant disease resistance, it doesn't contain the types of protein domain normally associated with pathogen recognition or signal transduction. We hypothesise that RPB1 acts in concert with other host proteins to interact with specific factors coming from *P. brassicae*. To find these interactors, the experimental approach we plan to take is to add a small tag to RPB1 and make transgenic plants that will have hybrid proteins. If these chimeric proteins can still recognise *P. brassicae* we hope to pull out the proteins that are complexing together by passing them through a matrix that binds the tag added to RPB1. Through affinity purification we can identify any proteins bound up with RPB1 by fragmenting them and using mass spectrometry to capture the molecular fingerprints of specific proteins. We will go on to test the function of any candidate proteins to characterise their interactions with RPB1 and generate mutants to confirm if they are involved in resistance to clubroot disease.

P. brassicae is genetically diverse, across Europe there are many different pathotypes which can infect different subsets of brassica cultivars. We will screen a collection of pathotypes to find those which are restricted by RPB1 and those which are unaffected. By comparing the genomes of compatible and incompatible pathotypes we aim to identify the factors that are being detected by RPB1 or the RPB1 protein complex. Through building up a model of the different host and pathogen proteins that are involved in clubroot resistance we will advance our understanding of the disease dynamics and the evolutionary pressures that are at play in the establishment and break down of immunity.