19_EFB Fera_BBKA master 11/04/2012 16:29 Page 19

# Biology and Spread of European Foul Brood: Some Preliminary Results

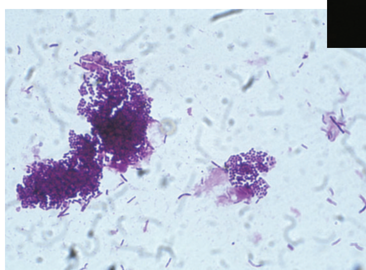
European foul brood (EFB) of honey bees is arguably the most important brood disease of honey bees in the UK, and is caused by the bacterium *Melissococcus plutonius*. It is far more common than the other bacterial disease of honey bees, American foul brood (AFB); in 2011 there were 366 cases of EFB in Great Britain, compared with fifty cases of AFB. On top of that its symptoms are probably more serious for a hive than other brood disorders like Chalkbrood, caused by the fungus *Ascosphaera apis*. I have written in *BBKA News* in February 2011 and October of the same year about my PhD project, in which I am trying to understand the biology and spread of EFB. Here I will give a brief update about my most recent progress.

## Bioinformatics

Most of my recent work has been in bioinformatics; the use of sophisticated computer software to analyse DNA sequences. My main aim has been to arrange our identified sequence sections of the *M. plutonius* type strain genome into the correct order (the type strain genome being the complete DNA sequence found in the first strain of this bacterium). This task is proving a lot more complicated than envisaged, as we now have identified large sections of DNA whose internal sequence we are confident about, but we are not completely certain in what order these large sections are arranged.

Despite not being completely finalised this genome sequence has been very valuable for other investigations into the biology of *M. plutonius*. For example, some work I did a while ago on the comparative metabolism of *M. plutonius* and another bacterium has been taken forward by undergraduate and Masters students in my laboratory at the university. This research has the potential to produce insights into how the bacterium is able to survive and thrive inside a honey bee larva. In addition to looking at the type strain genome I have been analysing sequences from some of the strains that I isolated in the UK in 2010 as well as some sequences from co-infecting bacterial species.

*EFB larva with yellow infected gut.*



*Melissococcus plutonius.*

Another aspect of my work with DNA sequences has been identifying genes that have slightly different sequences of DNA letters in different samples of *Melissococcus*. Knowing this will allow us to differentiate between strains of the bacterium. This work has been going on for some time now and I am gradually moving towards a scheme I am happy with. As with any scheme the amount of variation that I can detect is fundamentally limited by the variation present within the species.

In *M. plutonius* there are not huge differences between isolates, but I do seem able to pick out trends from isolates from different countries. This is something I hope to confirm with more samples from a broader geographical range.

## Apiary epidemiological investigations

In terms of working out some of the ways that EFB spreads around, my work has not solely been limited to making inferences from sequence data. I have taken samples (and had samples sent to me) from several case study apiaries from the Midlands and the north of England. The samples taken covered all the possible sources of EFB infection we could think of; soil, water, wild pollinators, bees from the apiaries, as well as swabs from all sorts of structures and equipment at the sites.

These samples were sent to Fera where I subdivided them; one portion of each sample was preserved to keep any bacteria present alive and the other portion had all the DNA present extracted. This DNA was then tested for sequences that were specific to *M. plutonius*. DNA from the EFB pathogen has been found on a range of equipment and in several samples of insects, and these provisional results will inform my sampling strategy for the coming beekeeping season.

*EFB-twisted and discoloured larvae. All photos supplied courtesy of The Food and Environment Research Agency (Fera), Crown Copyright.*

*EFB scale.*

## Future directions

I am now in the third year of my PhD and this coming beekeeping season will be the last I take samples from. One of the most important things now is to have a clear idea of what aspect of the work from last year I want to investigate further, and how I need to go about this in order to make the results rigorous and defendable. Other avenues of work will be the completion of the typing scheme, the finalisation of our genome sequence, and analysis of the data generated.

## Acknowledgements

My work is funded by a BBSRC CASE (Council for Advancement and Support of Education) studentship, in partnership with BDI (Bee Diseases Insurance). I am supervised by Dr Thorunn Helgason and Prof J Peter W Young (University of York), Dr Richard Thwaites and Dr Giles Budge (Fera), and Bernard Diaper (BDI). In addition, I would like to give thanks to the beekeepers and Bee Inspectors who have been an invaluable help with sample collection.

**Edward Haynes**, Fera and the University of York