DIAGNOSIS AND DETECTION OF DIFFERENT GENOTYPES OF *PAENIBACILLUS LARVAE*, THE CAUSAL AGENT OF AMERICAN FOULBROOD DISEASE

Adriana M. Alippi

Laboratorio de Referencia de Loque Americana de la Organización Mundial de Sanidad Animal (OIE) - Centro de Investigaciones de Fitopatología (CIDEFI), Facultad de Ciencias Agrarias y Forestales, Universidad Nacional de La Plata, La Plata, Argentina.

American foulbrood (AFB) is the most contagious and destructive infectious disease affecting the larval and pupal stages of honey bees (*Apis mellifera*) and other *Apis* species. The causative agent, *Paenibacillus larvae,* is a Gram-positive bacterium that can produce over one billion spores in each infected larva. AFB occurs in temperate or subtropical regions throughout the world and leads to huge losses not only in the apicultural economy but also in pollination rates since *Apis mellifera* is the most widely used actively managed pollinator in the world.

Only bacterial spores are capable of inducing the infection. Spores can remain viable for extended periods of time, survive adverse conditions (desiccation, high temperatures, ultraviolet light exposure) and resist contact with standard disinfectants.

Traditional methods such as the recognition of typical clinical symptoms of infection, culture of *Paenibacillus larvae* from diseased brood and microscopy are efficient and inexpensive ways to diagnose the disease. Clinical diagnosis of AFB is based on the identification of the pathogenic agent by microscopic examination of stained smears of dead or sick larvae. Apart from the distinctive clinical symptoms, laboratory confirmation of the presence of *Paenibacillus larvae* is required in most countries where AFB is a notifiable disease.

For laboratory diagnosis, *Paenibacillus larvae* can be isolated and cultured from various sources including ropy larval remains, scales, honey, pollen, wax and adult bees. Routine analysis of honey samples or adult bees for viable spores of the pathogen is strongly recommended to detect not only subclinical infections but also different *Paenibacillus larvae* genotypes.

PCR amplification of repetitive elements present in bacterial DNA (rep-PCR) is useful for genotyping, and ERIC-PCR amplification (rep-PCR by using enterobacterial repetitive intergenic consensus primers) has shown four *P. larvae* genotypes, named ERIC I, II, III, and IV. This typing scheme correlates with phenotypic differences including spore surface configuration, colony morphology, and virulence.

This presentation will focus on current methods for diagnosis and the potential application of various schemes for epidemiological studies of the pathogen.

Keywords: *Paenibacillus larvae;* AFB; diagnosis; genotypes; rep-PCR