

The Portuguese Experience with Bee Pathology Laboratories for the Diagnosis of Bee Diseases



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Introduction

In Portugal there are two referenced laboratories responsible for the diagnosis of bee diseases, namely the National Laboratory of Veterinary Investigation (LNIV) which is the National Reference Laboratory and the Bee Pathology Laboratory which belongs to the Agrarian Superior School of Bragança (ESAB). This laboratory was created in collaboration with the Apicultural Association of Montesinho Natural Park (AAPNM) and was certified by the Veterinary Authority in 2006. The Bee Pathology Laboratory is located in the city of Bragança, integrated in the Trás – Os – Montes e Alto Douro region. Is a research and also laboratory that provides a service to individual beekeepers when requested either directly from the laboratory or through the Apicultural Association including those of Controlled Zones. These Zones are defined as areas within the national territory where the systematic control of the bee diseases is undertaken. They are regulated at the national level by the Decree – Law n° 203 of November 25th -2005, which also established which diseases are considered to be of mandatory notification in our country. The diseases referred in this regulation are: American Foulbrood (AFB), European Foulbrood (EFB), Acarapidosis, Varroosis, Small Hive Beetles (SHB) infestation, Tropilaelaps infestation, Chalkbrood (only in Controlled Zones) and Nosemosis (only in Controlled Zones).

Materials and methods

The sample collection (sample of adult bees and/or sample of comb) that are recovered by Apicultural Organizations or beekeepers are placed in a cardboard box and submitted to the Bee Pathology Laboratory. This cardboard box is provided by the Veterinary Services of each region and the rules for sampling are written on each of them. In the laboratory, samples are preserved by freezing them, if there is no possibility of being processed soon. The diagnoses of bee diseases are carried out according to the routine methodologies used by LNIV. The microscopical examination of diseased larvae (which involves the preparation of a stained smear with nigrosin) is commonly used for routine diagnosis of the two most important bacterial diseases (AFB and EFB). For the diagnosis of fungal diseases (like Chalkbrood) we use the microscopic examination with a magnification of 40x to detect the presence of green-brown ascocysts on the surface of the mummies. Also, for the diagnosis of protozoan (Nosema and Amoeba) diseases a routine examination of a droplet of liquid from the macerated abdomen of bees is taken and prepared to be examined under the microscope. In the case of Amoeba disease diagnosis can be made by removing and microscopically examining the Malpighian tubules for the presence of amoeba cysts. For the diagnosis of parasitic diseases, we use the classic technique. Finally, for the diagnosis of pests, such as, Small Hive Beetles, they can be seen under a magnifying glass or by using a dissecting microscope to examine a brood sample or brood comb suspect.



Results

During 2007 and until March 2008, the Varroosis, Nosemosis, Amoeba Disease, AFB, Chalkbrood and Acarapidosis, were diagnosed as the principal honeybee diseases. In the table (I) we can see that the majority of samples submitted to this Laboratory are positive (above 80%). We can also, observe the prevalence of Nosemosis (66.3 and 53.3% between the study period). The positive diagnosis of Varroosis is evident in both years .

Table I – Data of submitted samples

	2007 (%)	January-April 2008 (%)	
Negative Samples (%)	3.2	19.2	
Positive Samples (%)	96.8	80.8	
Total samples (%)	100.0	100.0	
Positive Diagnosis of brood samples	Varroosis	28.4	41.6
	American Foulbrood	15.8	15.0
	Chalkbrood	17.9	9.2
	Wax Moths	4.2	2.5
Positive diagnosis of adult bees samples	Varroosis	20.0	24.2
	Nosemosis	66.3	53.3
	Acarapisosis	5.3	0.0
	Amoeba disease	31.6	9.2
Total of submitted samples (215)	95	120	

Conclusions

The Bee Pathology Laboratory has the objective of providing services to the country for a better control of honey bee diseases and simultaneously intends to develop research in this area. Therefore, we need to carry out new developed methods, such us, molecular methods or immunodiffusion tests, because some diseases (such as, viral diseases) can be diagnosed using these methods and aren't routinely used in our Laboratory .

Fig 1. Location and general view of the Bee Pathology Laboratory