Due to the high resistance of the spores, the bacterium *Paenibacillus larvae* is the most dangerous bacterial pathogen of the honey bee (*Apis mellifera*). Thanks to its biological properties and restricted pathogenicity, this bacterium can be used as a model organism to study gram positive sporulating aerobic rods. This work is focused on completing information about secreted proteases of this bacterium and in a study of proteases bound in a spore structure. MYPGP medium was used for the cultivation of *P. larvae*. In this medium, lysis of the culture was shown after 40 hours of cultivation. The pH of the medium decreased below 6.4 by lysis. The induction of temperate bacteriophage BLA was detected as a causative agent of this lysis. A new sporulation medium called HCBB agar was proposed for the sporulation of *P. larvae*. In comparison with HCBB agar with MYPGP agar by 31 strains of *P. larvae* stored in our collection, HCBB agar was evaluated as an appropriate sporulation medium with a median of sporulatin 4.2 ´ 106 spores per cm2 in aerobic conditions and 5.65 ´ 106 spores per cm2 in aerobic conditions with 10 % CO2. For purification of the secreted proteases, a one-day culture incubated at room temperature was used. Optimal purification of 87/74 kDa and 42/40 kDa proteases was observed after application of this sample on a DEAE-cellulose column. In the next step, analysis of the activity of purified proteases against bovine serum albumine was done. The secreted proteases have been stable for 14 days at 4 °C, for 3 days at room temperature and only 24 hrs at 35 °C. Proteolytic activity remained stable after heating at 60 °C for 10 minutes. With the increasing temperature, the proteolytic activity decreased. In comparing proteolytic patterns by the different 31 strains, all variants were presented in significantly pathogenic strains, so that there is no protease responsible for the virulence of *P. larvae*. For the extraction of proteases from the external structures of the spores, special electrophoretical equipment was constructed. This apparatus performs the easy extraction of proteases from 108 spores. The extracted proteases are probably located in the spore coat. These proteases were identified as metalloproteases inhibited by 1,10-phenantroline with an optimum of pH in the neutral value. Three combinations of the detected proteases were observed in the 31 strains of *P. larvae*. All these variants were shown in isolates collected from clinical material, so that no one protease is an essential virulence factor. The virulence of the decoated spores was tested *in vitro* and *in vivo*. These experiments proved the same virulence of the decoated and native spores. According to this result, we can hypothesize that the proteases bound in the spore coat do not have an essential function in the virulence of *P. larvae*.