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Insecticidal Activity of Bacillus Thuringiensis Strain AH-2 Against Hemiptera Insects Pests: Aphis. Gossypii, and Lepidoptera Insect Pests: Plutella Xylostella and Hyphantria Cunea

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Abstract : In recent decades, climate change has demanded biological pesticides; more Bt strains are being discovered worldwide, some containing novel insecticidal genes while others have been modified through molecular approaches for increased yield, toxicity, and wider host target. In this study, B. thuringiensis strain AH-2 (Bt-2) was isolated from the soil and tested for insecticidal activity against Aphis gossypii (Hemiptera: Aphididae) and Lepidoptera insect pests: fall webworm (Hyphantria cunea) and diamondback moth (Plutella xylostella). A commercial strain B. thuringiensis subsp. kurstaki (Btk), and a chemical pesticide, imidacloprid (for Hemiptera) and chlorantraniliprole (for Lepidoptera), were used as positive control and the same media (without bacterial inoculum) as a negative control. For aphidicidal activity, Bt-2 caused a mortality rate of 70.2%, 78.1% or 88.4% in third instar nymphs of A. gossypii (3N) at 10%, 25% or 50% culture concentrations, respectively. Moreover, Bt-2 was effectively produced in cost-effective (PB) supplemented with either glucose (PBG) or sucrose (PBS) and maintained high aphicidal efficacy with 3N mortality rates of 85.9%, 82.9% or 82.2% in TSB, PBG or PBS media, respectively at 50% culture concentration. Bt-2 also suppressed adult fecundity by 98.3% compared to only 65.8% suppression by Btk at similar concentrations but was slightly lower than chemical treatment, which caused 100% suppression. Partial purification of 60 – 80% (NH4)2SO4 fraction of Bt-2 aphicidal proteins purified on anion exchange (DEAE-FF) column revealed a 105 kDa aphicidal protein with LC50 = 55.0 ng/µ□. For Lepidoptera pests, chemical pesticide, Bt-2, and Btk cultures, mortality of 86.7%, 60%, and 60% in 3rd instar larvae of P. xylostella, and 96.7%, 80.0%, and 93.3% in 6th instar larvae of H. cunea, after 72h of exposure. When the entomopathogenic strains were cultured in a cost-effective PBG or PBS, the insecticidal activity in all strains was not significantly different compared to the use of a commercial medium (TSB). Bt-2 caused a mortality rate of 60.0%, 63.3%, and 50.0% against P. xylostella larvae and 76.7%, 83.3%, and 73.3% against H. cunea when grown in TSB, PBG, and PBS media, respectively. Bt-2 (grown in cost-effective PBG medium) caused a dose-dependent toxicity of 26.7%, 40.0%, and 63.3% against P. xylostella and 46.7%, 53.3%, and 76.7% against H. cunea at 10%, 25% and 50% culture concentration, respectively. The partially purified Bt-2 insecticidal proteins fractions F1, F2, F3, and F4 (extracted at different ratios of organic solvent) caused low toxicity (50.0%, 40.0%, 36.7%, and 30.0%) against P. xylostella and relatively high toxicity (56.7%, 76.7%, 66.7%, and 63.3%) against H. cunea at 100 µg/g of artificial diets. SDS-PAGE analysis revealed that a128kDa protein is associated with toxicity of Bt-2. Our result demonstrates a medium and strong larvicidal activity of Bt-2 against P. xylostella and H. cunea, respectively. Moreover, Bt-2 could be potentially produced using a cost-effective PBG medium which makes it an effective alternative biocontrol strategy to reduce chemical pesticide application.

Keywords : biocontrol, insect pests, larvae/nymph mortality, cost-effective media, aphis gossypii, plutella xylostella, hyphantria cunea, bacillus thuringiensi
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