## Pathogenicity of *Nosema* sp. (Microsporidia) in the Diamondback Moth, *Plutella xylostella* (Lepidoptera: Plutellidae)

# Nadia Kermani<sup>1</sup>, Zainal-Abidin Abu-hassan<sup>2</sup>, Hamady Dieng<sup>3</sup>, Noor Farehan Ismail<sup>1</sup>, Mansour Attia<sup>1</sup>, Idris Abd Ghani<sup>1</sup>\*

1 School of Environmental and Natural Resource Sciences, University Kebangsaan Malaysia, Bangi, Selangor, Malaysia, 2 Faculty of Medicine, University Technology MARA, Shah Alam, Selangor, Malaysia, 3 School of Biological Sciences, Universiti Sains Malaysia, Penang, Malaysia

### Abstract

Biological control using pathogenic microsporidia could be an alternative to chemical control of the diamondback moth (DBM) *Plutella xylostella* (Lepidoptera: Plutellidae). The microsporidium *Nosema bombycis* (NB) is one of the numerous pathogens that can be used in the Integrated Pest Management (IPM) of DBM. However, its pathogenicity or effectiveness can be influenced by various factors, particularly temperature. This study was therefore conducted to investigate the effect of temperature on NB infection of DBM larvae. Second-instar larvae at different doses (spore concentration: 0,  $1 \times 10^2$ ,  $1 \times 10^3$ ,  $1 \times 10^4$ , and  $1 \times 10^5$ ) at  $15^\circ$ ,  $20^\circ$ ,  $25^\circ$ ,  $30^\circ$  and  $35^\circ$ C and a relative humidity(RH) of 65% and light dark cycle (L:D) of 12:12. Larval mortality was recorded at 24 h intervals until the larvae had either died or pupated. The results showed that the spore concentration had a significant negative effect on larval survival at all temperatures, although this effect was more pronounced (92%) at  $35^\circ$ C compared with that at 20 and  $30^\circ$ C ( $\approx 50\%$ ) and  $25^\circ$ C (26%). Histological observations showed that *Nosema* preferentially infected the adipose tissue and epithelial cells of the midgut, resulting in marked vacuolization of the cytoplasm. These findings suggest that *Nosema* damaged the midgut epithelial cells. Our results suggest that *Nosema* had a direct adverse effect on DBM, and could be utilized as an important biopesticide alternative to chemical insecticides in IPM.

Citation: Kermani N, Abu-hassan Z-A, Dieng H, Ismail NF, Attia M, et al. (2013) Pathogenicity of Nosema sp. (Microsporidia) in the Diamondback Moth, Plutella xylostella (Lepidoptera: Plutellidae). PLoS ONE 8(5): e62884. doi:10.1371/journal.pone.0062884

Editor: Oscar Zaragoza, Instituto de Salud Carlos III, Spain

Received December 19, 2012; Accepted March 26, 2013; Published May 13, 2013

**Copyright:** © 2013 Kermani et al. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

**Funding:** This research was funded by grant No. (02-01-02 -SF0601) and (05-01-02-SF1019) from the Ministry of Sciences, Technology and Innovation, Malaysia (MOSTI) and Research University Grant (OUP): OUP-2012-043, OUP-2013-043. The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Competing Interests: The authors have declared that no competing interests exist.

\* E-mail: idrisgh@ukm.my

### Introduction

The diamondback moth (DBM) Plutella xylostella L. (Lepidoptera: Plutellidae) causes considerable economic losses worldwide to brassicaceous crops, and occasionally to other crops. Control of this pest is usually achieved through the application of synthetic insecticides that is estimated to cost more than US\$1 billion/ annum to control. Management costs and crop losses caused by DBM account for US\$4-US\$5 billion [1]. The high cost, environmental contamination, development of resistance to chemicals, and pest resurgence [2,3] associated with the current DBM control practices have encouraged the search for alternatives that are more environment friendly. Microbial control is an environmentally sound and valuable option to control this pest. In Malaysia, Nosema bombycis Negali is one of the several pathogens of DBM in the field [4]. DBM mortality is higher in younger instars (first and second instars) than in the older instars. Further, even at low concentrations, infection is remarkably higher for both larvae and pupae in highlands than in lowlands [4].

The effect of temperature on the biology of *Nosema* needs to be investigated because it is one of the most important ecological factors for the development of insect populations. Therefore, this study investigated the effects of *Nosema* spore concentration on the different stages of DBM reared at different temperatures. Establishing a correlation between temperature and pathogenicity of *Nosema* infection would be beneficial in determining whether *Nosema* can be applied as a DBM controlling agent. The optimal temperature at which this pathogen might be more effective in controlling the pest was studied. This information might help in determining whether this pathogen could be used in integrated pest management (IPM) and whether the amount of pesticide required could be reduced considering that *Nosema*-infected populations are more susceptible to the toxicity of the insecticides. In insects, the midgut is a dynamic tissue of the alimentary canal that acts as the route of digestion and allows absorption of digested food. Thus, we studied the effect of Nosema on this active organ. The results are also expected to provide useful information on the histopathology effects on larvae caused by *Nosema*.

#### **Materials and Methods**

#### **Diamondback Moth**

Disease-free DBM larvae of the University Putra Malaysia strain were obtained from the Malaysian Agriculture Research and Development Institute. The stock-culture of DBM used