

Sublethal effects of an Argentine *Bacillus thuringiensis* strain on the development and fitness of *Alphitobius diaperinus* (Coleoptera: Tenebrionidae)

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INTRODUCTION & AIM

- Alphitobius diaperinus* (lesser mealworm) is a significant pest affecting broiler and layer poultry facilities.
- Its life cycle (Fig. 1) occurs entirely within poultry litter and manure, leading to structural damage, bird injuries, and serving as a reservoir for microbial pathogens.
- Current control strategies rely on the use of chemical insecticides and the periodic replacement of litter.
- The INTA Mo4-4 strain of *Bacillus thuringiensis*, identified through a nationwide screening of native Argentine strains, has shown high lethality against *A. diaperinus* larvae.

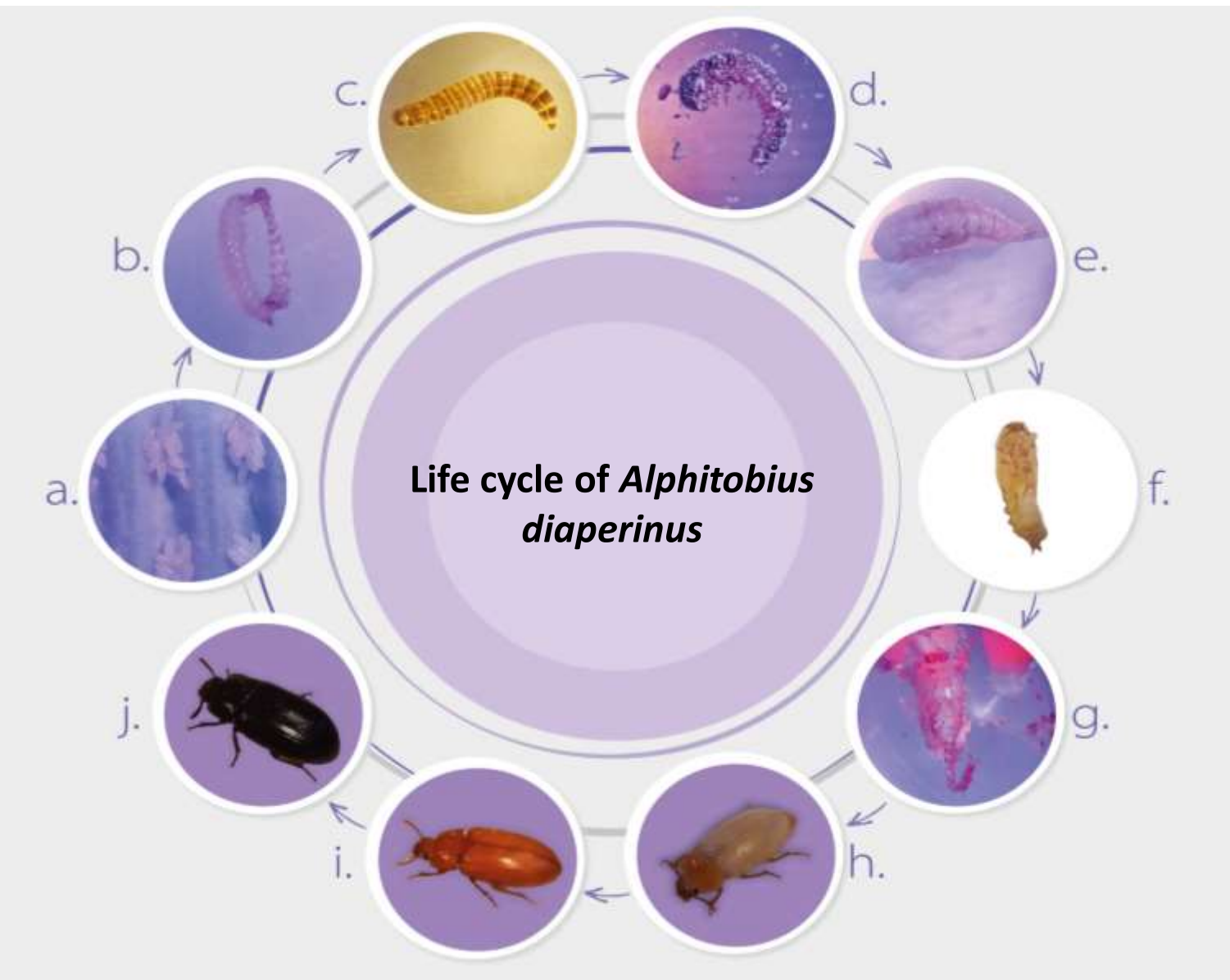


Fig. 1. Life cycle of *A. diaperinus*. (a) Eggs in clusters; (b) early-stage larva with molted exoskeleton; (c) advanced-stage larva; (d) larva preparing to pupate; (e) *A. diaperinus* pupa; (f, g) pupa undergoing progressive molting into an adult; (h) newly emerged whitish adult; (i) sclerotizing adult with a brown coloration; and (j) fully mature black adult.

The aim of this work was to evaluate the sublethal effects of *B. thuringiensis* INTA Mo4-4 on the development, survival, and fitness of *A. diaperinus* to assess its potential as an effective biocontrol agent in poultry facilities.

METHOD

In figure 2, a schematic presentation of the two-stage workflow used to quantify both lethal and sublethal effects of the INTA Mo4-4 strain on *A. diaperinus* larvae is shown.

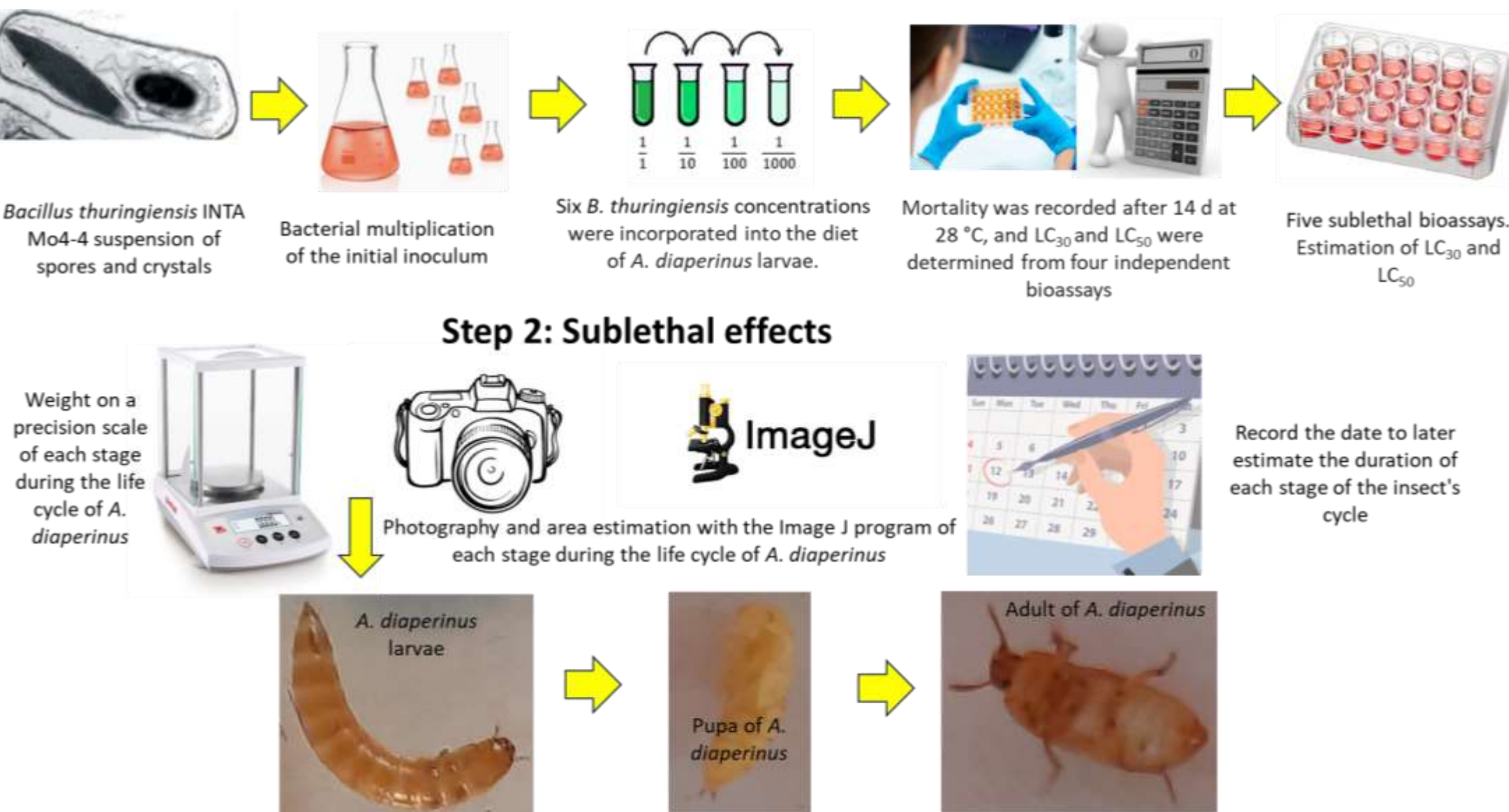


Fig. 2. First series of bioassays to determine lethal concentrations 30 and 50; second series of bioassays to study the sublethal effects themselves. Monitoring of larvae that subsequently pupated and developed into adults. Weight and area were recorded at each stage of the life cycle.

RESULTS & DISCUSSION

- Mortality results:** LC_{30} ($\mu\text{g/ml}$) = 69 [62 – 84] CV=15%; LC_{50} ($\mu\text{g/ml}$) = 136 [118 – 155] CV=12%; Slope: 1.82; χ^2 (4 df) = 4.57.
- Table 1 presents the sublethal effects of different lethal concentrations (LC_{30} and LC_{50}) of *B. thuringiensis* INTA Mo4-4 on key developmental parameters of *A. diaperinus* larvae, pupae and adults.

Table 1. Partial life table of *A. diaperinus*. The average, minimum, and maximum values are indicated for each parameter. Means with a common letter are not significantly different ($p > 0.05$).

Measured variable	CONTROL	LC_{30}	LC_{50}
Larval stage duration from hatching (days)	94.69 ^A [78.31 – 105.00]	101.13 ^A [80.92 -114.48]	122.60 ^A [96.00 – 151.00]
Larval stage duration since the end of Bt intake (days)	76.69 ^A [60.31 – 87.00]	83.13 ^A [62.92 - 96.48]	104.60 ^A [78.00 – 133.00]
Pupal stage duration (days)	5.71 ^A [5.00 - 6.36]	6.35 ^A [5.93 - 6.62]	6.95 ^A [6.29 - 7.60]
Larval area (mm^2)	1.32 ^B [1.17 - 1.67]	0.97 ^A [0.87 - 1.06]	0.82 ^A [0.71 - 0.95]
Larval weight (mg)	0.37 ^B [0.28 - 0.42]	0.25 ^A [0.23 -0.28]	0.20 ^A [0.17 - 0.22]
Pupal area (mm^2)	9.32 ^A [5.90 - 13.50]	8.44 ^A [5.70 - 9.50]	7.80 ^A [6.60 - 8.70]
Pupal weight (mg)	10.08 ^A [6.97 - 14.63]	8.75 ^A [4.44 - 11.21]	8.12 ^A [7.54 - 8.55]
Adult area (mm^2)	9.63 ^A [6.50 - 13.20]	8.54 ^A [4.90 - 10.90]	7.95 ^A [6.90 - 9.30]
Adult weight (mg)	8.97 ^A [6.08 - 13.10]	7.65 ^A [3.47 - 10.37]	6.96 ^A [6.59 - 7.17]

Larval effects:

- LC_{30} treatment: Reduced weight and area compared to control .
- LC_{50} treatment: Further reduced weight and area.
- Statistically significant differences compared to control.

Developmental periods:

- Larval and pupal stages were prolonged under higher treatment concentrations.
- Trends observed, but differences not statistically significant.

Pupal and adult fitness:

- Pupal weights and sizes decreased as treatment concentrations increased.
- Adult weights and body areas followed a similar decreasing trend, with lower values observed at higher concentrations.
- Trends observed, but differences not statistically significant.

CONCLUSION

- B. thuringiensis* INTA Mo4-4 induces both mortality and sublethal effects on *A. diaperinus*.
- Sublethal effects include reduced larval size and weight, with potential implications for pest development and fitness.
- Findings highlight the potential of *B. thuringiensis* INTA Mo4-4 as a bioinsecticide for controlling *A. diaperinus*, with insights for further development.

FUTURE WORK

- Once individual death date records are complete, we will estimate key mortality metrics (LT_{50} and LT_{90}) to evaluate the time-dependent effectiveness of treatments.
- Biochemical analyses will be performed on surviving larvae to assess potential differences in metabolic reserves (e.g., proteins, lipids, glucose, glycogen) across treatments.
- Further bioassays will examine the combined effects of *B. thuringiensis* INTA Mo4-4 and *Enterobacter* sp. INTA AN1-1, the most abundant bacteria isolated from the culturable gut microbiota of *A. diaperinus*.