

Exploring the biocidal properties of *Bacillus thuringiensis* INTA Mo1-10 through genomic and phenotypic analysisCamila Rojo¹, José Niz¹, Germán Dalinger¹, Leila Ortíz¹, Augusto Salas^{1,2}, Diego Sauka^{1,2}.¹Instituto Nacional de Tecnología Agropecuaria (INTA), Instituto de Microbiología y Zoología Agrícola (IMYZA), Hurlingham, Buenos Aires, Argentina; ²Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), Argentina.

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INTRODUCTION AND OBJECTIVES

Bacillus thuringiensis is a Gram-positive, spore-forming bacterium widely recognized for its potent insecticidal properties. It is recognized as a key biological control agent and a more sustainable alternative to chemical pesticides. This study focuses on the INTA Mo1-10 strain, preserved in our collection and originally isolated from grain milling residues in Argentina. The objective was to elucidate the relationship between its genetic profile and biocidal activity through comprehensive genotypic and phenotypic characterization.

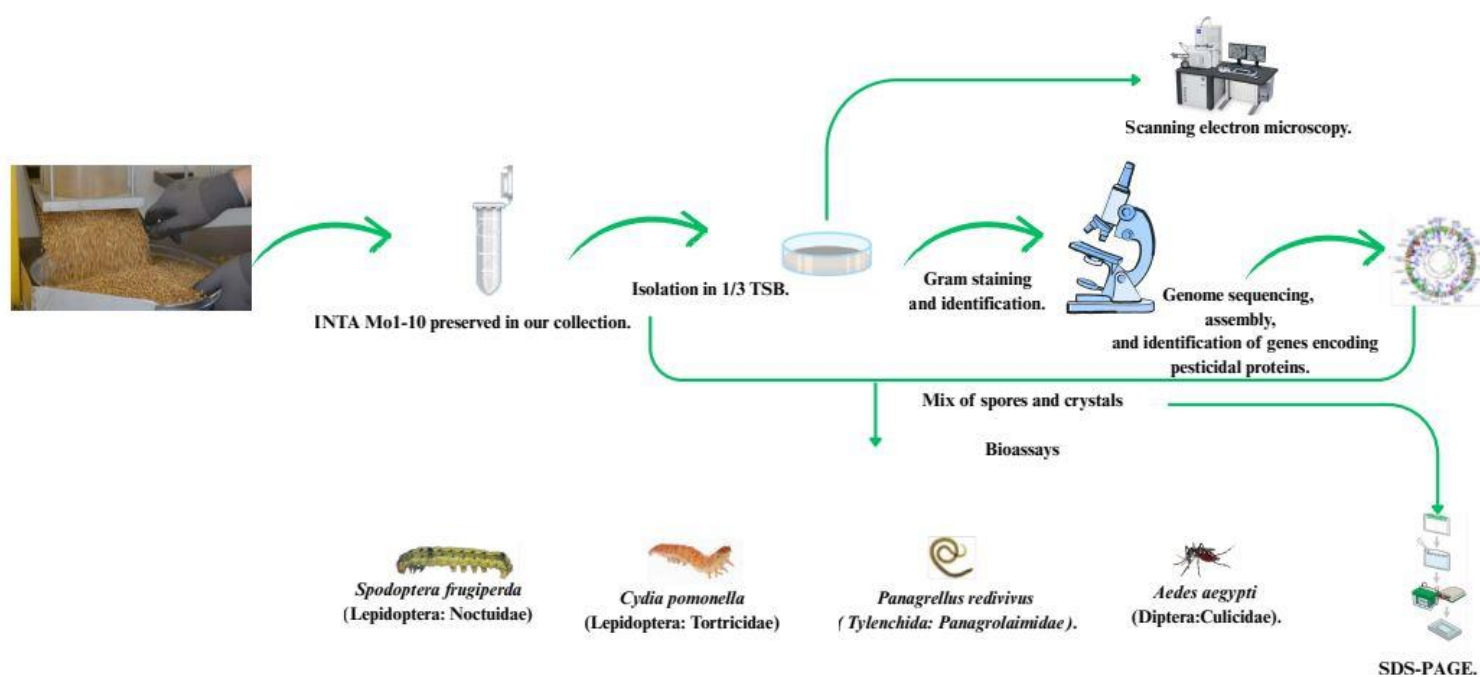
METHOD

Fig 1. Schematic representation of the work plan, from the isolation of the strain from grain milling residues in Buenos Aires Province, Argentina.

RESULTS & DISCUSSION

Surface streak isolation of INTA Mo1-10 allowed us to observe whitish, shiny colonies with regular edges. Gram staining confirmed that it is a Gram-positive bacterium. The genomic sequence showed a total size of 6,298,872 bp in 215 contigs with a G+C content of 34.7%. Genomic sequence analysis (**Table 1**) revealed the presence of 9 coding sequences with significant similarity to known pesticidal proteins from *B. thuringiensis*.

Contig	Pesticidal proteins	Percent identity (%)	Completeness	Target order
29	Cry1Da1	100	Yes	Lepidoptera
	Cry1Ca15	100	Yes	Lepidoptera Diptera
	Cry1Ia10	100	Yes	Lepidoptera
	Cry1Aa3	100	No	Lepidoptera
36	Spp1Aa1	77.4	No	Blattodea Lepidoptera
85	Cry2Ab1	100	No	Lepidoptera Diptera
	Vip3Aa10	100	No	Lepidoptera
154	Cry9Ea11	100	No	Lepidoptera
162	Cry1Ab1	100	No	Lepidoptera

Table 1. Putative pesticidal proteins identified in INTA Mo1-10: contig, pesticidal protein, % identity, completeness, and predicted target (BPPRC database).

The presence of a parasporal crystal protein band of approximately 130 kDa was confirmed by SDS-PAGE analysis (**Fig. 3**). The literature links the mentioned band with bipyramidal crystals and the Cry1 protein with lepidopterocidal activity, which is confirmed in INTA Mo1-10.

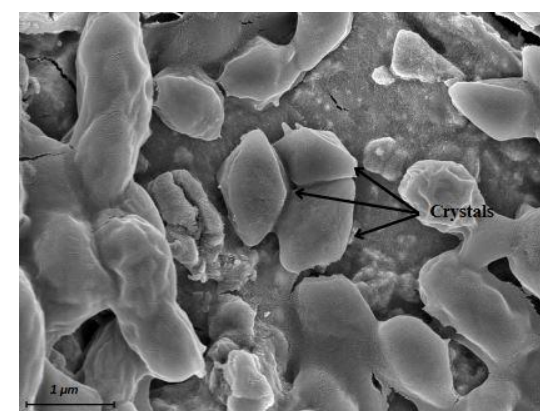


Fig 2. Visualization of bipyramidal crystals of INTA Mo1-10 by scanning electron microscopy.

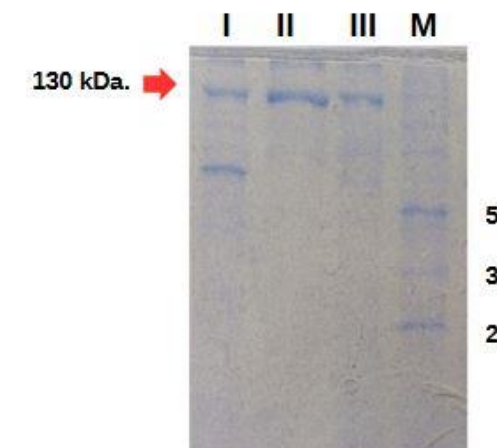


Fig 3. SDS-PAGE analysis of crystal protein components of INTA Mo1-10 and different *B. thuringiensis* strains. Lanes: I, HD1; II, HD137; III, INTA Mo1-10; M, molecular weight marker.

Qualitative bioassays

Target species	Target order	Family	Concentration (µg/mL)	Mortality (%)
<i>Spodoptera frugiperda</i>	Lepidoptera	Noctuidae	250	83.5 ± 6.4
<i>Cydia pomonella</i>	Lepidoptera	Tortricidae	10	83.3 ± 4.2
<i>Aedes aegypti</i>	Diptera	Culicidae	10	45.0 ± 0.0
<i>Panagrellus redivivus</i>	Tylenchida	Panagrolaimidae	ND	84.0 ± 1.0

Table 2. Mortality rates of INTA Mo1-10 spore–crystal preparations against target species at the indicated concentrations. Values are expressed as mortality (%) ± standard deviation (SD).

Quantitative bioassays

Target species	Target order	Family	LC ₅₀ (µg/mL)	CV	Slope	χ ²
<i>Cydia pomonella</i>	Lepidoptera	Tortricidae	2.8[2.3 - 3.4]	0.1	1.8	3.0
			2.9 [2.3 - 3.5]		1.7	2.1
			2.4[2.0 - 2.9]		2.3	2.8
Mean			2.7			

Table 3. LC₅₀ values (µg/mL) with 95% confidence limits, coefficient of variation (CV), slope, and χ² from mortality assays of *Cydia pomonella* using INTA Mo1-10 spore–crystal preparations.

CONCLUSION

INTA Mo1-10 exhibits a clear link between its genetic composition and biocidal activity. Cry1 proteins account for activity against Lepidoptera, while Cry1C may underlie mosquitocidal effects. Nematicidal activity was not linked to crystal proteins.

FUTURE WORK / REFERENCES

Complete *Spodoptera frugiperda* LC₅₀ bioassays and deepen genomic analysis to identify nematode virulence factors.