Testing the efficacy of *Trichogramma minutum* in the context of an 'Early-intervention Strategy' against the spruce budworm using different release methods

Véronique Martel¹, Simon Trudeau¹, Rob Johns², Emily Owens², Sandy Smith³, Guy

¹Laurentian Forestry Centre, Canadian Forest Service, Natural Resources Canada, Qc; ²Atlantic Forestry Centre, Canadian Forest Service, Natural Resources Canada, NB; ³Department of Forestry, University of Toronto, ON; ⁴Horticultural Research and Development Centre, Agriculture and Agri-Food Canada, Qc

Industry Partners: Anatis Bioprotection; Canopée dronautique

Abstract

This report presents the second year's results (2017) of the research project aiming to assess the efficacy of *Trichogramma* releases against SBW. In 2017, eleven blocks were set up in three areas in Quebec: seven blocks had a control, a release by card and a release by drone plots. The other four blocks had a control and a release by card plot. In each plot, branches were collected in the spring and fall to evaluate population densities using L2, and in summer for SBW egg parasitism. Moths were collected using pheromone traps from the Budworm Tracker project (http://budwormtracker.ca) in each region to synchronize the release with oviposition. While in 2016, results showed that egg parasitism was higher in all plots treated with cards, in 2017, the results were not as clear. The block with the lowest egg density showed a higher egg parasitism for both treated plots (cards and drones) compared to the control. However, parasitism was low in the other blocks. Several factors could have decrease the impact of our parasitoids' releases: the cold and rainy weather, the presence of another moth species in abundance that could have come into competition with SBW eggs for parasitism, and predation by slugs.

Résumé

Ce rapport présente les données de la 2^e année (2017) du projet visant à évaluer l'efficacité des lâchers de trichogrammes contre la TBE. En 2017, onze blocs ont été installés dans trois régions du Québec : sept blocs avaient un site témoin, un site avec lâchers par cartes et un site avec lâchers par drone. Les quatre autres blocs avaient un site témoin et un site avec lâchers par cartes. Dans chaque site, des branches ont été collectées au printemps et à l'automne afin d'évaluer les densités de populations à l'aide des L2, et à l'été pour les taux de parasitisme des œufs. Les papillons étaient piégés par des pièges phéromone du projet **Pisteurs** de tordeuses (http://pisteursdetordeuses.ca/) pour synchroniser les lâchers. Alors qu'en 2016, les résultats montraient que le parasitisme était plus élevé dans les sites traités, en 2017, les résultats étaient moins clairs. Le bloc avec la plus faible densité d'œufs a obtenu un parasitisme plus élevé dans les sites traités (drone et tricho-cartes) que dans le témoin. Par contre, le parasitisme était bas et non concluant dans tous les autres blocs. Plusieurs facteurs semblent avoir diminué l'impact de nos traitements : la météo froide et pluvieuse, la présence d'une autre espèce de papillons abondantes qui pourrait avoir été en compétition pour le parasitisme des œufs, et la prédation par des limaces.

Introduction

With a spruce budworm outbreak currently spreading through eastern Canada, there has been increased interest in developing new approaches to mitigate potential damage. The 'Early-intervention strategy' (EIS) has been alternative/complementary approach to the traditional 'Foliage protection strategy'. EIS focuses on using products to treat spruce budworm 'hot spots' while populations are still relatively low, ideally halting or slowing the local rise and spread of outbreak. A significant challenge to developing the EIS is that female moths often migrate in from surrounding unsprayed areas to lay eggs, potentially offsetting local population reductions caused by insecticide application [1]. None of the currently available control agents (i.e., Mimic, Btk, and pheromone) can be used to target spruce budworm eggs and it remains uncertain whether additional egg mortality could aid in managing low-density populations near to ongoing outbreaks.

Trichogramma minutum is a generalist egg parasitoid commonly used in agroecosystems [2] and has been tested previously as a biological control agent against spruce budworm. Among the advantages of using this biocontrol agent is the ease of its rearing for commercial purpose and the fact that it kills its host before the damaging stage, i.e. before caterpillars. Although it is used in orchards [3] and has been previously tested as a biological control agent against spruce budworm, it is not currently used in forestry in Canada. Past studies found that proper deployment of T. minutum in areas of high spruce budworm density increased egg parasitism by 14-83% and reduced larval populations from 42 to 82%, thus providing significant foliage protection benefits [4]. In the context of EIS, T. minutum application densities would be much lower and cost less than those required for treating high-density populations. Moreover, the wide range of established deployment methods (e.g., aerial application, ground application, etc.) [5] and negligible environmental impact could make it a versatile tool for application in a conventional spray program as well as in areas not suitable for chemical insecticide application, such as some woodlots, conservation and residential areas, and parks. As a complementary extension of the ongoing ACOA Early Intervention project (ACOA 2.2.5), we propose to examine the potential of T. minutum as a biological control agent for reducing egg densities in low-density spruce budworm populations in small private wood lots, using two different release methods. This control strategy would be the only one currently available where the immigrating moths could be targeted through their egg progeny and one of the few control methods available for private owners. This mortality would also be added to the natural parasitism, and eventually to any larval insecticide used.

For the second year of this project, we focused on comparing the efficacy of two release methods: the installation of cards on the ground vs. drone.

Objectives

- 1) Determine whether the use of *T. minutum* significantly increases egg mortality in low to moderate density spruce budworm populations.
- 2) Determine the impact of *T. minutum* releases on the populations' growth (L2-L2).
- 3) Compare the efficacy of both release methods (cards vs. drones).

Methodology

Study sites

Based on the L2 and defoliation levels evaluated by the MFFP, we identified three areas (Charlevoix, Mauricie and Laurentides – Sainte-Adèle) where SBW is present with either low defoliation or in a restricted area. Within each area, we selected respectively four, six and one 1 ha blocks with one (Charlevoix) or two (Mauricie and Laurentides) treated plots (30m x 30m) and one control plot (30m x 30m) (Fig. 1). It is important to not that in Sainte-Adèle, the municipality had decided to treat all their forest and wood lots with Btk to decrease SBW populations.



Figure 1. Map showing the four study areas in Quebec: Sainte-Adèle (2017), Mauricie (2017), Charlevoix (2016-17) and Kamouraska (2016). Coloured areas represents the observed defoliation by aerial surveys made by MFFP in 2016.

Trichogramma releases

Release timing is of prime importance in biological control, and especially when releasing *Trichogramma* [4]. To make sure that the first *Trichogramma* release was carried out at the beginning of the moth flight periods, we were in contact with citizens from the Budworm Tracker project to be informed of when the first males were captured. The first release was conducted a few days after the first capture and the second release one week after the first one.

Ground releases (using commercial cards provided by Anatis Bioprotection, thereafter called 'Tricho-cards' on which about 6,000 *Ephestia kuehniella* eggs parasitized by *T. minutum* are glued – Fig. 2) were made in each treatment plot. For each release, 100 Tricho-cards were installed at eye level on balsam fir or spruce branches, as evenly as possible within the plot. At the second release, the first card was left in place to allow for

SERG Interim Report (Year 2, Jan. 2018): T. minutum against SBW

late emergences and a second card was installed next to the first one. The Tricho-cards were removed about a month later, during branch sampling.



Figure 2. Tricho-cards, provided by Anatis Bioprotection, containing about 6,000 eggs parasitized by T. minutum

Drone releases (using a modified drone developed by Canopée Dronautique; Fig. 3) were done in one plot/block in Mauricie and Ste-Adèle (but not Charlevoix), with the same release rate (3M/ha, i.e. 1.5M females/ha) and on the same day as the cards installation.



Figure 3. A. Picture taken from the drone, by Canopée Dronautique, showing the parasitized eggs mixed with vermiculite being released in Sainte-Adèle; **B.** Drone take-off for the release in Mauricie (Alexandrais Maltais piloting the drone).

Egg parasitism and population growth

Local population densities were assessed using L2 assessment by branch collection (15/plot) before (April 2017) and after (October 2017) the growth season as well as egg assessment by branch collection (25/plot) about 10 d after the end of oviposition period as predicted by BioSIM [6]. The egg collection was also used to assess the effectiveness of the *T. minutum* treatment and the natural occurrence of *Trichogramma* in the control blocks by measuring parasitism rates.

Results

Population levels

The L2 density before the treatments varied between a little below 1 to 15 L2 per branch in the different sites (Fig 4). The L2 density after the treatments are not yet available as SBW is currently in diapause – results will be available in spring 2018.

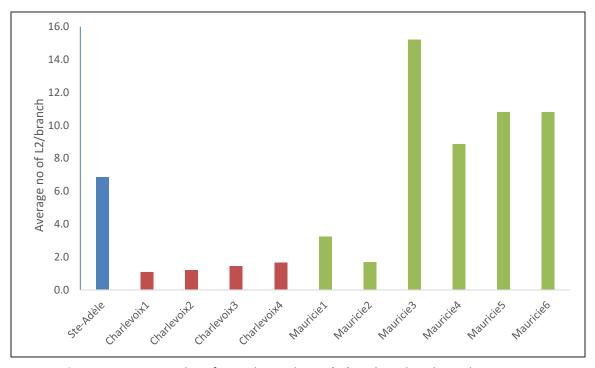
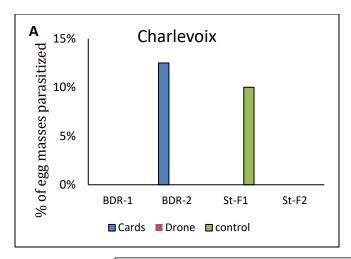
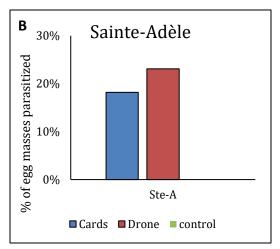


Figure 4. Average number of second instar larvae (L2) per branch in the study sites.

Egg parasitism

There was some parasitism in a few plots, but there is no clear pattern (Fig. 5A-C). When putting the percentage of parasitism in relation with the egg density on the site, it appears that the highest parasitism rates were found when egg density was lower, which would be expected (Fig. 6).





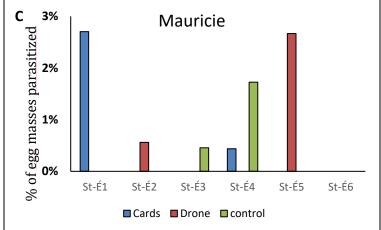


Figure 5. Percentage of SBW egg masses parasitized by T. minutum in the control and treated plots for each area.

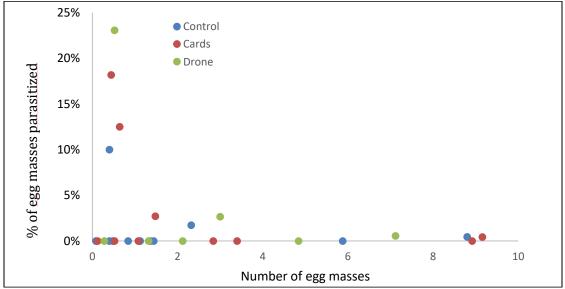


Figure 6. Percentage of SBW egg masses parasitized by T. minutum in both 2016 and 2017 in relation with the density of eggs on the site

Discussion and tentative conclusions

The results make it difficult to draw clear conclusions on the efficacy of *Trichogramma* releases against SBW. In both year (2016 and 2017), the block with the highest efficacy was the one with the lowest egg density. That probably indicates that the release rate should be increased for plots with higher SBW density, to compensate for the lowest efficacy of *Trichogramma* at high host density. In 2017, for the Sainte-Adèle block where the egg density was the lowest, plots with releases by both drone and tricho-card show similar efficacy, with about 20% of SBW egg masses parasitized, while there was no parasitism in the control plot. Although only on one site, these results indicate that this method, under optimal conditions, can give promising results. Sainte-Adèle probably had the lowest egg density, even though the L2 density was initially higher than in Mauricie, because of the Btk treatment performed at the larval stage. This suggest that the use of two control methods might increase the overall efficacy in decrease SBW densities. The L2 data after treatments will confirm, or infirm, this.

However, results from the Charlevoix and Mauricie blocks are not as clear. In Mauricie, there is some parasitism in different plots, with no clear pattern between treated and control sites. Nevertheless, the important aspect is that no parasitism was above 3%, which mean that it was negligible everywhere. In Charlevoix, where no drone release were done, we had parasitism in only 2 plots: one treated and one control. Among the possible factors that could have decrease our efficacy: (i) the cold and rainy weather during the release period could have delay the *Trichogramma* emergence, extending the duration of their exposure to predation; (ii) in Mauricie, at the time of the release, two other moth species were found in higher abundance than SBW – these moths' eggs were probably in competition with SBW eggs to be parasitized by *Trichogramma*, decreasing our efficacy; (iii) in Charlevoix, some of the Tricho-cards collected after the emergence show important signs of slug predation – if the predation occurred before the *Trichogramma* emergence, the actual release rate was much lower than intended.

The L2-L2 population growth data, available next spring, will be necessary to confirm if the treatment had an impact on the population level for the Ste-Adèle block. Although not so clear, these preliminary data show that it is possible to increase egg parasitism using *T. minutum* releases under good conditions (SBW density, *Trichogramma* release rate, weather, etc.), and that it might be more efficient in low-density populations. However, additional study is necessary to determine more precisely these conditions. For any further study, I would recommend using the drone release method instead of the Tricho-cards one: the release are much faster and show the same efficacy for the block where it worked.

Acknowledgements

We thank the Ontario Ministry of Natural Resources and Forestry for financial support and the Ministère de la forêt, de la faune et des parcs du Québec (MFFP) for their support and for providing data from the L2 and defoliation surveys. We also thank the Canadian Forest Service, especially the SMA Innovation Funds for providing financial and in-kind support, Anatis Bioprotection, especially Silvia Todorova and Mylène St-Onge, for providing insects and precious advices, as well as Canopée Dronautique (Frédéric Jean et

SERG Interim Report (Year 2, Jan. 2018): T. minutum against SBW

Alexandre Maltais) for the drone services. We are also grateful to the private lot owners and the Saint-Siméon and Sainte-Adèle municipalities for providing us with access to their land and property during this study. Numerous people also provided support in the lab and the field: we thank S. Bélanger, P. Huron, M. Labrecque, M. Gorla, L.-P. Gélinas and C. Rochon.

References

- 1. Béchard, A., Delisle, J., Johns, R., Labrecque, A., Martel, V., Pureswaran, D., Régnière, J., Royer, L., Thompson, D., van Frankenhuyzen, K. (2014) SERG-I Project FPL-786 Dynamics and management of rising spruce budworm outbreak. Interim Report.
- 2. Consoli et al. 2010. *Egg parasitoids in agroecosystems with emphasis on* Trichogramma. Springer.
- 3. Smith SM (1996) Biological control with *Trichogramma*: advances, successes, and potential of their use. *Annual Reviews in Entomology* **41**: 375–406.
- 4. Smith, S.M., Wallace, D.R., Howse, G., Meating, J. (1990) Suppression of spruce budworm populations by *Trichogramma minutum* Riley, 1982-1986. *Memoirs of the Entomological Society of Canada* **153**: 56-80.
- 5. Smith, S.M., Wallace, D.R. (1990) Ground systems for releasing *Trichogramma minutum* Riley in plantation forests. *Memoirs of the Entomological Society of Canada* **153**: 31-44.
- 6. Régnière, J., Saint-Amand, R., Béchard, A. (2014) BioSIM 10 User's manual.
- 7. Quayle, D., Régnière, J., Cappuccino, N., Dupont, A. (2003) Forest composition, host-population density, and parasitism of spruce budworm *Choristoneura fumiferana* eggs by *Trichogramma minutum*. *Entomologia Experimentalis et Applicata* **107**: 215-277.