Report for the Canadian Bee Research Fund (2021-22)



International Honey Market

Research
bulletin:
Tracking
temperatures
of nuc
shipments
from
Vancouver to
Whitehorse in
June, 2021

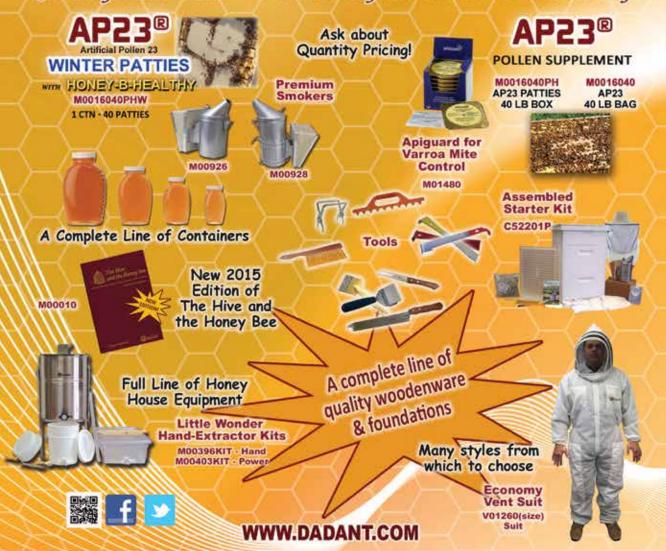
EurBeST – a pilot study testing varroa resitant bees under commercial bee keeping conditions





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Cover picture: Bee collecting resin from sea buckthorn Photo Credit: Cherie Andrews, Chinook Honey Company & Chinook Arch Meadery

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Canadian Honey Council Report





Rod Scarlett, Executive Director, CHC

he onset of spring normally encompasses renewed optimism and a sense that "this is the year" when everything works out. High honey prices and improved pollination contracts have certainly given cause for hope, but this has been tempered by ever increasing input costs and supply chain issues. As I am writing this, it is becoming increasingly apparent that an even more daunting issue is facing the sector, that of stock replacement. Beginning in mid-March reports were coming into the office of large over-wintering losses. While this is not unusual in regional circumstances, it was apparent the losses were more national in nature. Higher than average losses were reported across the Prairies and in Ontario and Quebec. While packaged bees were still coming into Canada it became clear that demand greatly exceeded supply.

While there seemed to be consensus that varroa was the main cause for the high losses, it was apparent that many beekeepers were faced with the fact that there was little or no replacement stock available either internationally or domestically. Naturally, there were calls for "quick fix" solutions the most obvious being opening the US border for package bees. In the years that I have served as Executive Director, I can honestly say that this is the one issue that divides individual beekeepers and provincial associations. Perhaps, given the circumstances this spring, it is time to discuss, in rational terms, the elephant in the room.

I have no strong opinion on this matter one way or the other as I see the arguments on both sides. One of the problems is that this is often an emotional argument and beekeepers are often diametrically opposed. There seems to be no attempt to compromise nor to strategically offer alternatives. Both sides say they rely on science, but interpret the science to match their perspective. In very simple terms, one side has beekeepers who insist there is no health risks, and even if there were, the ability to access stock outweighs any risk. The other side is worried about bee health risks and maintains that domestic stock production needs to be emphasized and supported.

Certainly, that is an over-simplification but it really is the crux of the matter. From a Canadian food security perspective, we need bees for pollination. We also need bees for honey production. What is interesting is that as far as I can determine, bees may be the only livestock in Canada that relies on other countries to supply a large percentage of replacement stock. A vast percentage of queens are imported and packages serve to recoup a good number of colonies lost over winter. This is certainly at odds with the idea of domestic self-sufficiency.

Still, if beekeepers were to suffer similar national losses next year, it may be time now to have that serious discussion on the pros and cons of the border opening, to put together an emergency plan that could be instituted on a very short timeline. Hopefully, it wouldn't be needed but for the health of the national industry, the discussion should not be avoided. Just food for thought.

The Mite-A-Thon is a North American effort to collect Varroa mite infestation data in honey bee colonies. All beekeepers can easily participate by testing their hives for mites, creating a rich distribution of sampling sites in Canada, the United States, and Mexico. This initiative has provided a platform for beekeepers' associations to engage newer members and re-familiarize long-time members with proper Varroa monitoring techniques. It has also opened the conversation on treatment practices, initiating a dialogue between beekeepers. The Mite-A-Thon runs from April 30 to May 15 and from August 13 to 28.

Visit www.mitecheck.com to submit data. ■



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Atlantic





It's the beginning of April and there is already panic in the beekeeping and blueberry industry. Early reports from many provinces are above average to heavy losses. Beekeepers are scrambling to find ways to replace winter losses but are finding it hard given the current global situation. Blueberry growers are also very nervous and it seems there is going to be a fairly large shortfall of pollinators this season. With the current price

of blueberries this is going to heavily impact the blueberry industry. This also may impact long term growth as the blueberry industry looks to open up new land but are starting to have a reality check on trying to figure out where the pollinators are going to come from. Even through good winters for the bees, the current projections for berry land expansion is currently exceeding the amount of bees that can be raised between the respective pollinating provinces. Some outside the box solutions that do not put the health of local hives in jeopardy are going to be required if the blueberry industry wants to continue to expand. Needless to say with the scarcity of available hives for pollination this season the price for pollination is at a record high. CHC has been involved in a pollination task force with beekeepers and growers from PEI, NB, Quebec, NS and BC to try and come up with some ideas to deal with the pollinator shortage this season and in future seasons.

The bees out east are actually looking quite good considering what seems to be happening to the rest of the country. It's still early and a lot can happen but several large New Brunswick producers are currently looking at under 10% loss. Bees seem a bit light coming out of winter but weather seems to be coming around and getting feed to them shouldn't be an issue. As of right now I have not heard of any large disasters. A few beekeepers in Nova Scotia are reporting slightly higher than normal losses but nothing to be worried about. If nothing to crazy happens in the next month or so, 2022 is shaping up to be a pretty decent season for eastern beekeepers.

I hate to do this but I'm going to end this column on a sad note. Alex Crouse, the president of the Nova Scotia group passed away suddenly on April 6th. I had the pleasure of working with Alex over the years with the NS group and our Atlantic Beekeeping joint group. Alex was full of energy and had so many great ideas. He was great for the industry and was always willing to learn. He was a great leader for the Nova Scotia beekeepers and their loss will be felt for years to come. I believe Alex would have been a future CHC representative for Atlantic Canada and who knows how many more great contributions we will now miss out on in Eastern Canada. Alex was humble, kind

hearted and always willing to talk bees. I was lucky to have known him as a friend and work closely with him. Alex you will be missed...

Québec





Trop de déception ce printemps au Québec. Les ruches sortent par centaine des caveaux depuis quelques semaines et plusieurs apiculteurs constatent plus de mortalité que ce qui était anticipé. Malheureusement, cette situation est généralisée au Canada.

L'automne dernier, les prévisions de notre équipe de recherche tournaient autour de 40 % de

Maggie Lamothe Boudreau perte provincialement. Nous en sommes mainte-

nant à des prévisions de tout près de 60 % de prévisions. Ce dossier à occupé énormément les administrateurs cet hiver puisque ces pertes massives ont des impacts considérables sur les entreprises apicoles concernées et sur des cultures dépendantes de la pollinisation par l'abeille domestique. Vous devriez recevoir sous peu un sondage du Conseil canadien du miel afin de connaître les diverses raisons pour lesquelles certains apiculteurs ne participent pas à la pollinisation du bleuet. Plusieurs comités ont été mis sur pied dans celui du « Task Force Pollination » du Conseil Canadien du Miel et le comité pollinisation de l'association québécoise Les Apiculteurs et Apicultrices du Québec. D'ailleurs plusieurs négociations ont été réalisées avec la financière agricole afin d'obtenir une assurance catastrophe. Nous avons appris lors de l'assemblé générale annuelle que cette assurance entrera en vigueur cette année et quel défrayera seulement la portion des pertes entre 70% et 100 % chez les apiculteurs inscrits à l'assurance hivernent.

Les producteurs de Bleuets sont toujours à la recherche d'apiculteurs intéressés à participer à la pollinisation. Si jamais vous démontrez un intérêt communiqué avec votre représentant CCM qui pourra me transférer vos coordonnées qui seront transmises à l'association des producteurs de Bleuets du Québec. Plusieurs dossiers continuent d'être travaillés tels que celui de l'ARLA où nous souhaitons qu'un apiculteur puisse aussi j'ai sur le comité technique. Les négociations avec les producteurs de grain vont bon train. Plusieurs projets à venir en collaboration avec l'UPA sont en préparation, je vous reviendrai avec plus de détail lors du prochain rapport. Api Bleu Max est maintenant en fonction pour les trois prochaines années et visera a évalué le plus possible de paramètre concernant la pollinisation et sont impact sur les ruches d'abeilles. Ce projet est essentiel afin de solidifier le partenariat étroit entre les apiculteurs et pour les producteurs de Bleuets.

Ensuite, l'organisation travaille a collaboré avec le CCM et le CRESAD afin de réaliser la mise su pied du fameux projet de nourrissage automnal des ruches en régie biologique. Ce projet souhaite com-



parer le nourrissage au sirop de sucre comparativement au nourrissage au sirop de sucre biologique ou au miel. En effet pour le moment la majorité des études que nous possédons stipule qu'il est essentiel de nourrir les ruches au sirop de sucre conventionnel afin qu'elle passe l'hiver adéquatement. Ce projet permettra une bien meilleure compréhension de cet enjeu crucial pour tous les apiculteurs canadiens. Nous remercions l'Alberta de participer activement au projet.

Une demande est d'ailleurs en cheminement afin que le président du Comité biologique du Conseil canadien du Miel puisse siéger au Comité technique qui réalise l'évaluation des normes biologiques canadienne.

Notre Assemblée générale Annuelle (AGA) a eu lieu le 18 mars en personne. Je tiens à féliciter les membres du conseil d'administration élue qui contribueront à l'avancement de plusieurs dossiers importants à l'apiculture québécoise.

En voici la liste:

Président : Raphaël Vacher

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2e vice-présidente : Julie Fontaine

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Administrateur et président du comité du Nord-Ouest : André Talbot Administrateur et président du comité de la région de Québec : David Lee Desrochers-Croteau

Administrateur(-trice) et président(e) du comité Mauricie-Estrie-Centre-du-Québec : Steve Michel

Administratrice et représentante de la relève : Sophie Roy

Administrateur et représentant de la catégorie « petite échelle » : Julien Levac Joubert



Je souhaite donner du courage pour la saison 2022 à chacun d'entre vous qui n'avez pas obtenu la survie escomptée. Battonsnous pour protéger l'abeille, cet être essentiel à notre agriculture canadienne.

Too many disappointments this spring in Quebec. The hives have been coming out of the wintering vaults by the hundreds for a few weeks and several beekeepers are seeing more mortality than expected. Unfortunately, this situation is widespread in Canada.

Last fall, our research team's forecast was around 40% loss provincially. We are now at a forecast of almost 60% and spring isn't over. This issue has occupied administrators enormously this winter since these massive losses have considerable impacts on the beekeeping companies concerned and on crops dependent on pollination by honeybees.

Several committees have been set up within the "Pollination Task Force" of the Canadian Honey Council and the pollination committee of the Quebec association Les Apiculteurs et Apicultrices du Québec. You should soon receive a survey from the Canadian Honey Council to find out the various reasons why some beekeepers are not involved in blueberry pollination.

Moreover, several negotiations have been carried out with the ministry of agriculture in order to obtain catastrophe insurance. We learned at the Annual General Meeting that this insurance will come into effect this year and it will only pay the portion of the losses be-

tween 70% and 100% among beekeepers registered for wintering insurance

Blueberry growers are still searching for beekeepers interested in participating to pollination. If you ever show an interest, contact your CCM representative who can transfer your contact information to me which I will then send to the association of blueberry producers of Quebec.

Several other topics continue to be worked on such as that of the PMRA where a request was sent to get a beekeeper to sit on the technical committee. Negotiations with grain producers are also well underway. Several upcoming projects in collaboration with the UPA are in preparation, I will come back to you in more detail about these during hopefully my next report. Api Bleu Max is now in operation for the next three years and will aim to evaluate as many parameters as possible regarding pollination and impact on beehives. This project is essential in order to solidify the close partnership between beekeepers and Blueberry producers.

Then, the organization worked with the CCM and CRESAD to carry out the implementation of the famous project concerning autumn feeding of hives in organic management. This project aims to compare feeding with sugar syrup versus feeding with organic sugar syrup or honey. Indeed, for the moment the majority of studies we have stipulate that it is essential to feed the hives with conventional sugar syrup so that it spends the winter adequately. This project will provide a much better understanding of this critical issue for all Canadian beekeepers. We thank Alberta for its active participation in the project. Let's add that a request is in process for the Chair of the Canadian Honey Council's Biological Committee to sit on the Technical Committee that evaluates Canadian organic standards.

Our Annual General Assembly (AGM) was held on March 18 in person. At last, it felt so good to see all of you beekeepers in person. I would like to congratulate the members of the elected Board of Directors who will contribute to the advancement of several important issues for Quebec beekeeping.

Here is the list:

President: Raphaël Vacher

1st Vice-President: Maggie Lamothe Boudreau

2nd Vice-President: Julie Fontaine

Director and Chair of the Montérégie Committee : Alexandre Mainville

Director and Chair of the Northwest Committee: André Talbot

Director and Chair of the Quebec City Region Committee: David Lee Desrochers-Croteau

Director and Chair of the Mauricie-Estrie-Centre-du-Québec Committee: Steve Michel

Director and emerging representative: Sophie Roy

Director and representative of the "small scale" category: Julien Levac Joubert



I want to give courage for the 2022 season to each of you who have not achieved the expected survival. Let us fight to protect the bees, an essential part of our Canadian agriculture.

Ontario





John Van Alten

My name is John Van Alten. I have been asked to represent Ontario at Honey Council. Following in the footsteps of Albert Devries, I have big shoes to fill. I hope to be able to have a positive influence on some of the issues facing our industry. I appreciate the opportunity to reacquaint myself with beekeeping across Canada.

Ontario is finally getting some good flying conditions and some early pollen is starting to come in. That being said, I am hearing a wide va-

riety of overwintering reports, but in general it looks like we are experiencing even higher than normal wintering losses. Some commercial outfits are reporting very high losses, and the colonies that are alive are generally small clusters. While there are some positive reports, it looks like this will be a rebuilding year for a lot of beekeepers. I am still awaiting the final data from the overwintering survey sent out by the Ontario Beekeeper's Association, but the deadline for my report submission won't allow me to wait.

Poor overwintering is being reported across Canada and while there are probably a lot of different reasons why this is happening, one thing is clear. If we are to remain a viable agricultural industry and a dependable source for the pollination of fruit and oil seed crops, we must find a way forward to stop this from happening. This will involve beekeepers, researchers, government agencies and the grower groups working together.

I hope to be part of finding solutions. From what I've seen, your Canadian Honey Council delegates and Administrator are all working hard on this issue. Take care.

Saskatchewan





Jake Berg

Spring is slow in coming to Saskatchewan. Indoor winter hives are still sitting inside and as I'm writing this there is a spring storm forecasted for 5-15 centimetres of snow. So far this spring is shaping up to be one of the latest times we've moved bees out which is getting more concerning with each passing day with higher than normal winter loses being reported across the country. Also, there is a rising fear that apivar is not working as effectively as it once did so there is some

anticipation to get our hives out of storage and checked over to reveal what our survival rate is.

Although this is yet to be proven, I do believe the sun is beginning to set on the usefulness of apivar in Canada. But the question is; where do we go from here? Can we extend its life if we use other or multiple types of treatments such as formic acid, oxalis acid, thymol or brood breaks. With the combination of other treatments possibly apivar's life can be extended for a few more years.

With higher than normal over wintering losses, it is apparent that beekeeping in Canada has a mite and a bee health issue. Most of the higher than normal winter loss can be explained by higher than normal verroa loads last fall and the viruses that come along with that. Rather than importing more sick bees into Canada from around the world, I believe it would prudent to fix the current bee health situation in Canada as a more long term solution.

Hopefully in the next couple weeks, spring will actually begin in Saskatchewan and we can begin our beekeeping season and make plans for the summer ahead.

Alberta





Curtis Miedema

It seems like another season is upon us. Winter isn't quite done sending a few last blasts before we can enjoy spring. Most beekeepers have been through their bees by now in our area and it seems that losses are defiantly higher than normal. Some producers reporting absolute wreaks with the main assumed culprit is the varroa mite. It seems the current treatments are becoming less effective. Hopefully we will see new control options come to the market soon. With the chal-

lenges of sourcing replacement stock locally and internationally this will put a large strain on the industry. As reports are saying high losses across the country it may be time to consider looking at package bees from the mainland United States as a viable option in times of disaster.

Beekeepers are resilient and we have been through high losses before. Hopefully everyone can find ways to build back their numbers and capitalize on the favorable honey prices.

Wishing everyone a fantastic season.

BeeMoid

Bee Maid Honey Limited Scholarship and Research

In 2004, the Fiftieth Anniversary Scholarship was created to commemorate the formation of Bee Maid Honey Limited in 1954. The Scholarship recognizes and encourages children and grandchildren of members and staff as they enter post-secondary study. The Scholarships are designed to stimulate the pursuit of excellence by rewarding outstanding achievement. The scholarships are also intended as a process by which young people can bring fresh ideas and attitudes to the Bee Maid family.

Bee Maid Honey Limited is pleased to announce that the winners for the 2022 Fiftieth Anniversary Scholarships are Abigail Peace from Saskatoon, Saskatchewan and Aprille Colipano from Stoney Plain, Alberta. Abigail has been accepted by the University of Saskatchewan and is planning to pursue a Bachelor of Science in Biomedical Sciences. Aprille is registered with the Northern Institute of Technology and is pursuing Business Administration, specializing in Accounting. Congratulations to them both and we wish them the best of luck in their studies.

The Bee Honey Research fund was established to support research in any area of apiculture, hive health or honey production. A call for proposals was sent out in late 2021 and the committee was pleased to see so many high quality applications. The Bee Maid Honey Research Committee is pleased to announce the following project was selected for funding assistance.

• "Development of Two Novel Miticides into Field Application to Control Varroa mites in Honey bees" conducted by Dr. Medhat Nasr.



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Report for the Canadian Bee Research Fund (2021-22)

Field Trials of a New Acaricidal Compound Against Varroa destructor in Honey Bee Colonies

Stephen Pernal (Agriculture and Agri-Food Canada, Beaverlodge, Alberta) Erika Plettner (Simon Fraser University, Burnaby, British Columbia)

Progress Toward Objectives and Results:

ield work to evaluate our experimental compound 3c {3,6} against the honey bee parasitic mite, Varroa destructor, started in September 2021 at AAFC's Beaverlodge Research Farm and in Surrey, British Columbia. In preparation for the work in Alberta, Robert Lu was recruited as a new Masters' student in the Department of Biological Sciences at the University of Alberta. Robert was resident in Beaverlodge during the fall of 2021 to conduct the experiment at that location with assistance from AAFC technicians Abdullah Ibrahim and Rassol Bahreini. Field work in the lower mainland of BC was led by Jorge Enrique MacIas-Samano of the Plettner lab with the assistance of beekeeper Carolyn Essaunce.

In both of the parallel trials in Alberta and British Columbia, there were three experimental treatments: 1) Compound 3c{3,6}, 2) Thymol, and 3) a Negative Control. We chose a thymol-containing treatment as a comparison because thymol has physical properties similar to our experimental compound. Each treatment was replicated ten times across colonies in each location, with compound 3c{3,6} being delivered on wooden wafer release devices (15.3 cm x 5.1 cm). The compound was applied to the release devices similar to our earlier studies in 2019, using isopropanol with 2% glycerol (v/v). The dose in 2021 was 4 g/device, applied in layers, in 40 mL of isopropanol with 2% glycerol. The negative control was comprised of release devices receiving solvent only. The thymol treatment consisted of one application of Thymovar®, used per instructions, however only for a 4 week treatment period. As such, the latter treatment consisted of two half-strips of Thymovar®, having a total thymol content of 15 g, applied to the top bars of colonies in a manner similar to compound 3c{3,6}.

Prior to the experimental treatment, single brood chamber colonies were equalized with regard to brood and food stores, as well as mite loads. Four days prior to the experiment, natural mite drops were counted by installing a sticky board under each colony. The board was replaced on the first experimental day when the treatment devices were installed (Day 0), this being 3 Sep 2021 at both locations. Also on Day 0, we performed an alcohol wash of workers to check phoretic mites on adult bees, and examined 100 cells of capped brood to determine infestations of varroa on pupae and levels of mite reproduction. In addition, we measured the areas of pollen, honey, capped brood and adult bees. After 28 days of experimental treatment, the devices were

retrieved, a second alcohol wash was taken and the same set of assays performed. Apivar® strips were installed as a 6-week finishing treatment (Fig. 1), and the phoretic mite infestations were again determined.

2021 Timeline

Fall Varroacide Experiment



Figure 1. Timeline of field experiments at AAFC in AB and SFU in BC, testing compound 3c(3,6) against a negative

Results of mite drops onto the sticky boards are shown in Fig. 2 (BC) and Fig. 3 (AB). There were higher numbers of mites on the sticky boards in the treatments, compared with the negative control, but the effect was not as strong as we had observed in 2019. Furthermore, during the Apivar® finishing treatment, we saw large numbers of mites dropping in both the treated and control colonies, unlike in 2019 where we did not see such a mite surge during the Apivar® treatment for the $3c\{3,6\}$ treatment (only in the control).

In 2021, the efficacy of the $3c\{3,6\}$ treatment was lower than anticipated (Figs. 2, 3), $39\pm6\%$ in BC and $47\pm6\%$ in AB. The Thymovar® treatment had higher efficacy in BC ($59\pm3\%$) and similar efficacy in AB ($49\pm7\%$) compared with the $3c\{3,6\}$ treatment. In both locations, the natural mite drop in the negative control was similar over the 4-week treatment period ($23\pm3\%$ in BC, and $24\pm3\%$ in AB).

The main difference between the trial in 2019 and the one in 2021 was the delivery method of compound $3c\{3,6\}$. In 2019, a set of 10 tongue depressors were used to deliver a combined 5 g dose of the compound, whereas in 2021 a single wooden wafer was used to deliver a 4 g dose of the compound. The reason we chose to decrease the dose for the 2021 trial was that, in 2019, ~ 1.2 g of material were recovered from the tongue depressors after the trial. This meant that ~ 3.8 g of material had been transferred into the hive.

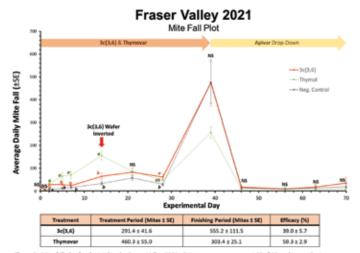


Figure 2. Mite fall plot for the trial in the Fraser Valley 2021. Points represent means ± 5E of 10 replicate colonies.

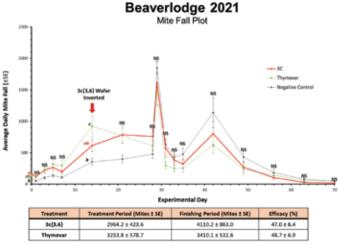


Figure 3. Mite fall plot for the trial in Beaverlodge 2021. Points represent means ±5E of 10 replicate colonies

At the start of our 2021 experiments, colonies had a 2.0% mite load on adult bees in BC and 14.8% in AB. There were no differences in mite loads among the three treatments, per location, as observed throughout the experiment (Table 1, data for Washes 1-3 on experimental days 0, 28 and 70).

Table 1. Data for mites on adult bees (washes), brood and food stores

Location	Tr.	Alcoho	washes		Day 0		Day 28		Day 0		Day 28	
		(mites/100 bees)			No. of 4x	No. of 4x4" squares			No. of frame sides			
		Wash	Wash	Wash	Cappe	Open	Capped	Open	Honey	Pollen	Honey	Pollen
		1 (%)	2 (%)	3 (%)	d brood	brood	brood	brood				
BC	3с	1.9 ± 0.3	5.1 ± 3.2	0.8 ± 1.5	9.2 ±	9.7 ±	0.4 ±	0.0 ±	5.0 ± 0.4	0.6 ±	9.4 ±	0.3 ±
	_				2.0	2.6	1.0	0.1		0.2	1.9	0.3
	Thy	2.0 ±	4.3 ± 2.3	0.0 ± 0.1	10.9 ±	6.5 ±	0.8 ±	0.9 ±1.2	4.9 ± 0.7	0.6 ± 0.2	10.1 ± 3.1	0.2 ± 0.1
	Con	2.0 ± 1.5	6.4 ± 3.5	0.3 ± 0.7	10.5 ± 2.4	6.0 ±	0.6 ± 1.1	0.5 ± 1.3	6.1 ± 0.7	1.6 ± 0.9	8.6 ± 3.6	0.4 ± 0.3
AB	3с	15.0 ± 5.7	21.1 ± 9.1	0.1 ± 0.3	21.6 ± 7.7	6.8 ± 4.2	6.1 ± 5.8	1.9 ± 3.6	3.0 ± 2.4	2.0 ± 1.1	3.3 ± 1.3	2.1 ± 1.2
	Thy	18.0 ± 8.7	19.3 ± 6.7	1.9 ± 2.4	19.1 ± 5.0	7.0 ± 3.0	4.5 ± 3.5	1.2 ± 1.0	3.8 ± 2.3	1.7 ± 1.1	3.7 ± 2.1	2.1 ± 1.3
	Con	12.2 ± 6.6	22.4 ± 13.4	0.6 ± 0.7	25.1 ± 6.1	15.1 ± 6.1	7.5 ± 5.0	2.4 ± 2.8	6.5 ± 3.1	1.9 ±	3.8 ± 1.6	1.9 ±

Averages ± SD

There were no differences among the three treatments in terms of brood areas (capped or open), or between honey and pollen stores. During the course of the 28-day treatment, overall, there was a significant decrease in the amount of brood and a significant increase in the quantity of honey or syrup stored. These are both indications that the colonies had stopped growing, in preparation for winter. Consequently, it was not possible to obtain meaningful data for mites in brood on Day

28. Nevertheless, mite levels in brood were similar across treatment groups prior to day Day 0 of the experiment.

Colonies were overwintered in their treatment groups. In BC, as of March 3, 2022, all but two of the colonies survived. Cluster sizes were measured in November 2021 and in early March 2022, and there were no significant differences among the three treatment groups (Table 2). The two colonies that perished were in the Thymovar® and control sets. Colonies in Beaverlodge, AB are still wintering.

Table 2. Overwintering data (BC)

Location	Treatment	Cluster size (f	rames covered)	Cluster size	Cluster size (frames covered) March 2022		
		Nov 2021		March 2022			
		Top view	Top view Bottom view		Bottom view		
BC	3c{3,6}	9.0 ± 1.1	5.1 ± 1.2	8.6 ± 1.0	4.4 ± 1.2		
	Thymovar	9.4 ± 1.0	5.4 ± 1.0	8.0 ± 2.9	3.9 ± 1.5		
	Control	9.1 ± 1.0	5.4 ± 0.8	7.3 ± 3.1	3.1 ± 1.7		

Averages ± SD Note: two dead colonies were counted as zeros in their respective sets.

Impediments and Unexpected Events Encountered:

There were two unexpected aspects of the experiment performed in 2021: 1) the need for a large surface area of the release device for 3c {3,6} and 2) difficulty building up mite populations in the early part of the summer in BC, thereby causing a later start of the experiment.

- 1) The difference between the release devices used in 2019 (10 tongue depressors) and 2021 (one wooden wafer) was the surface area. The tongue depressors used in 2019 had a combined surface area of 900 cm², whereas the wafers used in 2021 had a surface area of 156 cm². Another difference between the two trials was the placement of the wafers: between brood combs in 2019 and on top of the combs in 2021. The results from 2021 have taught us that it is critical to obtain sufficient evaporation of the compound in the brood nest, in order to have a large mite drop at the beginning of the treatment. Therefore, we have decided to design new release devices with a larger surface area that will be placed between brood combs instead of on top of them.
- 2) The slow start in building up mite populations in colonies in BC, despite efforts to seed colonies and to add extra mites in, caused us to delay the experiment until early September. In 2019, we started one month earlier in British Columbia and, therefore, still had brood that could be assessed by the end of the experimental treatment period. Though the start of the experiments in BC and AB was completely synchronized by date in 2021, the colonies in Alberta ended up having higher initial infestations of mites than desired. In retrospect, these trials would have been better started before fall so as to present more realistic mite treatment levels.

Future Short-Term and Long-Term Goals:

Short-term, our goal is to perform a field trial of 3c{3,6} in the Fall of 2022 with the following improvements over the one in 2021: 1) we will test new release devices that have a combined area similar to the tongue depressors used in 2019, 2) with a higher dose (8 g/ brood box), 3) for 6 weeks (as opposed to 4 weeks). We will test two substrates for the release devices: porous wood (same product as in 2021) and cardboard.

In 2022 we will be starting with apiaries where bees with mites have been overwintered. In 2021, we started from mite-free imported package bees, in BC. As such, we anticipate that mite populations will build up sufficiently by mid-to late summer. Starting the experiment in August will place the evaluation work after the experimental treatment in September, when there is still some brood present. This will enable us to get more meaningful data on mites in brood, to assess the effect

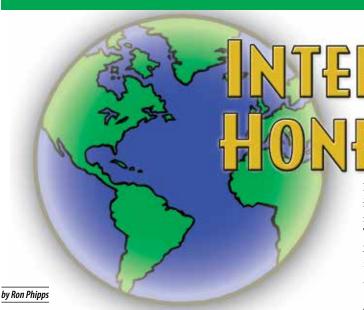
of the new acaricide on mite reproduction. We also plan to perform a laboratory incubator bioassay to test the effects of compound 3c{3,6} on the mortality of mites within individual sealed brood frames. This assay will provide meaningful data for the efficacy of the compound under controlled settings.

Medium-term, we plan to fully validate and use a method of residue analysis in wax and honey of compound 3c{3,6}. Wax samples will be taken immediately after treatment, after wintering and during honey production. Honey will be drawn immediately after treatment (for spring trials), after wintering and during honey production.

Long-term, we plan additional field trials in 2023. Additionally, we plan to find the target protein(s) compound 3c{3,6} binds to in the mites. Knowledge of the target site will be helpful in registration and also in future improvement of the technology.

Summary:

In conclusion, we have conducted two large-scale field trials in 2021 that have produced valuable data for the efficacy of compound 3c{3,6}. Our results were also very consistent between two very different geographic regions. We have learned that the method of application of our experimental compound needs to be modified, the dosage increased and the exposure period lengthened to 6 weeks. This exposure period is consistent with most other commercially-registered products. We also found no negative effects of compound 3c{3,6} on the size or health of honey bee colonies. Also gained were valuable insights into managing experimental mite populations at a field level, to better position us for success in our fall 2022 trials. ■



The international honey industry is in an unprecedented situation with a confluence of major developments.

Honey antidumping rates

The U.S. Department of Commerce's preliminary calculations of honey antidumping duties have sent shock waves throughout the world, especially in the U.S., Canada, Mexico, Europe, Argentina, Brazil, Ukraine and Vietnam. The countries in the antidumping case are currently among the top ten global honey exporters.

Preliminary Honey Antidumping Duty Rates (announced as of Nov. 17, 2021):

	Country Rate	Exporter Rates*			
Argentina	16%	7.8%, 24%, 49%			
Brazil	20%	7.8%, 29%			
India	6.48%	6.24%, 6.72%			
Ukraine	18%	18%, 32%			
Vietnam	412%	410%, 413%			
*Assigned to specific exporters					

This means that six of the largest honey exporters in the world are subject to U.S. antidumping orders. The antidumping order for Chinese honey has been in effect for 20 years. According to FAO statistics, China is the largest world exporter of honey. Within the U.S., the countries' rank by import volume is India, Vietnam, Argentina, Brazil, and Ukraine. Therefore what happens in the final determination, scheduled for April 2022, in respect to final rates and critical circumstances, will have a profound effect on the international honey market and on beekeepers in America and in the world.

U.S. law requires that the Department of Commerce conduct verifications to validate information and data submitted by foreign producers, and that interested parties may comment on the verification reports.

Brazil successfully argued that there were mistakes in the DOC's calculations, so that the rate for one exporter was reduced to 10.52% and the rate for all others came down to 9.38% in mid December. Interested parties have the legal right to raise comments during the verification period regarding the comprehensiveness and accuracy of the evidence, the integrity of the documents, and the representations. It is anticipated that exporters, importers, beekeepers and lawyers will be presenting through proper channels their arguments for changes in respect to both rates and critical circumstances.

Before 2021 ended, there were reports that Argentina and Vietnam had critical circumstances. In the case of Argentina, the report is that the company with the lowest rate was exempt because their export volume did not create the type of surge that results in critical circumstances. As of this writing, Brazil did not receive critical circumstance designation, and it is not known whether India or Ukraine will. Since critical circumstances involve retroactive duties, this is a very significant question for importers.

The threat of an antidumping petition has been a concern as early as the 4th quarter of 2020, and international honey exporters and U.S. importers have been creating a surge of shipments presumably to beat the date that preliminary antidumping duties would be collected by U.S. Customs. Cash payments were imposed in November 2021. Total U.S. imports for year-to-date September 2021 increased by 65,000,000 lbs.

(21%) compared to 2020 and reflect an overall surge in shipments.

The Final Determination of duty rates is scheduled for April 8, 2022. Until then, imports will be assessed cash duties at the preliminary rate. After the Final Determination is made, there could be a retroactive increase in duties to be collected.

	YTD Sep 2020	YTD Sep 2021	% Change
Argentina	66,059,571	73,741,248	12%
Brazil	46,902,726	56,141,804	20%
India	68,400,062	100,298,658	47%
Vietnam	82,139,703	99,385,528	21%
Ukraine	9,025,924	7,401,119	-18%
Total Imports	316,757,130	382,048,128	21%

U.S. Imports in Pounds Jan. to Sept. 2021 vs. 2020

There is an axiom in mathematical logic that states that if P implies Q and Q is false, then P is also false. Given the fact that Indian Light Amber and Vietnamese Light Amber have sold at similar low prices for many years, and, if as reported, India was used as a comparable country in the Vietnam antidumping duty calculations, it is hard to understand the 406 point (6.48% for India and 412% for Vietnam) disparity in the Preliminary Antidumping Duty Rates, with Vietnam's being so high and India's so low.

Shares of U.S. Imported Bulk Honey from Five Major Exporting Countries, 2015-2021(thru Aug.)									
	2015	2016	2017	2018	2019	2020	Jan Aug. 2021		
Argentina	16%	21%	18%	20%	21%	21%	20%		
Brazil	9%	12%	12%	13%	14%	18%	18%		
Canada	5%	8%	8%	8%	4%	2%	1%		
India	21%	18%	23%	24%	28%	20%	27%		
Vietnam	22%	24%	18%	21%	21%	26%	23%		
Other Countries	28%	17%	21%	14%	11%	13%	10%		
TOTAL	100%	100%	100%	100%	100%	100%	100%		

It is noteworthy that India, with a population of 1.3 billion which enjoys honey and uses it as a medicinal product, has increased their exports to the U.S. from zero in 2000 to claim a market share of nearly 30% of the U.S. market. India has received a dramatically lower antidumping rate compared to countries with mature beekeeping and honey exporting industries and good, internationally recognized beekeeping practices. Beekeepers in advanced honey producing nations have suffered declines in productivity which have been correlated with environmental degradation of soil, water and atmosphere. In China and India the environmental degradation and stress are even more severe, increasing the challenges to productivity rates for authentic honey.

Many beekeepers in North and South America were amazed and baffled to see a 6.48% duty rate for Indian honey, and no critical circumstances, when India's import volume surged 82% in 2021 compared to the same period in 2020.

Underlying the concerns of accuracy are questions around what is actually being bought, authentic honey or economically adulterated honey. It is becoming increasingly clear that that question cannot be answered without reference to the modes of production and the full traceability of the products and their authenticity.

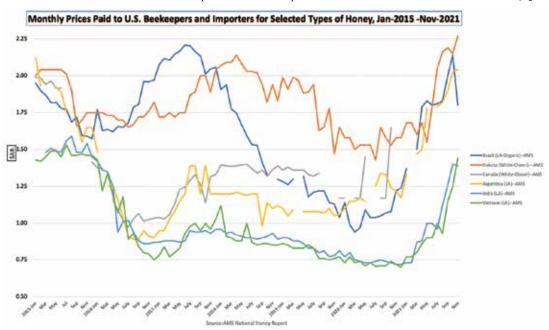
Honey market tendencies

In the period leading up to the announcement of the antidumping findings, the market was basically frozen. Many exporting countries had low inventories and at the same time there was complete uncertainty as to what the duties would be. That included which countries and which exporters within those countries would achieve comparative advantage. After the announcement, along with other events, honey prices rose, in some cases dramatically. For those countries which have exported at extraordinarily low prices during recent years, the high antidumping rates are serving as an umbrella, allowing fresh offers from traditionally low-priced exporters to almost double. The whole tenor of the market has changed.

The confluence of events, including the promulgation of the USP standard and widespread use of Nuclear Magnetic Resonance testing, will most likely compel a shift in the export of honey, and a strengthening of prices. This is a market which will not be easily manipulated. That in turn will help prevent economically motivated adulteration. Countries which have a history of exporting adulterated honey may have to find other markets in a new environment in which adulteration, in its various modes, may become very difficult.

U.S. and import prices

Honey prices were trending downward until March 2021, and after the filing of the antidumping petition they turned sharply upward. In 2020, prices for clover were above \$1.50/lb., then climbed over \$2.00/ lb. after March 2021. Indian and Vietnamese prices were below \$0.75/ lb. in 2020, and remained below \$1.00/lb. through 2021. All import prices rose after April 2021. ▶ pag. 14



2021 Honey Prices						
	March	July 2021				
• India	\$0.73/lb.	\$0.99/lb.				
• Vietnam	\$0.75/lb.	\$0.90/lb.				
Argentina	\$1.25/lb.	\$1.75/lb.				
• Brazil	\$1.29/lb.	\$1.79/lb.				
• US White	\$1.60/lb	\$2.00 plus				
• Canada	\$1.25/lb.	\$1.97 (Sept.)				

In the 4th quarter of 2021, the escalation of prices has become steeper and honey is being offered at \$1.80-\$2.25/lb.

The value of the global honey market was \$7.84 billion (U.S.) in 2020, experiencing significant growth of 9%, despite weak prices. Consumer demand for natural sweeteners, and the perception of honey as an immunity-boosting product, are helping to fuel the increase during a global pandemic.

Public Health England recommends honey for treatment of acute cough, one of the common symptoms in COVID patients. Honey is being incorporated into more and more pharmaceutical products globally.

Argentina

Argentina has received substantial antidumping rates, but within Argentina there is a major exporter with comparative advantage with respect to the antidumping rate and critical circumstances. Other exporters are subject to higher rates and critical circumstances. The amount of retroactive critical circumstances duties could be financially very significant for importers who bear liability for those duty payments. Honey exporters in Argentina, as well as in other countries, are concerned about terms of sale in this uncertain context.

At the beginning of the crop there were good rains and suitable temperatures in the areas producing the major spring honey crops. This is attributed to it being a La Nina year. If things continue with reasonable weather, the 2022 crop should be in the upper range. The majority of Argentine white honey was shipped to the U.S. in 2021, which received 70% of total exports. Japan prizes the quality of Argentine honey, and their requirements are strict. By November, Argentina had exported 60,567 metric tons, and by the end of December the quantity may reach 65,000 metric tons. Most quantities that remain in stock at the end of 2021 are dark honey.

The crop began in September-October with citrus and lemon in northern Argentina. Thus far the weather conditions are reasonable. Argentina expects a normal crop, which generally ranges from 55,000 to 75,000 metric tons. Because Argentina is a large country ranging over many longitudes with different climates, the national crop is stable. Since fields have been converted over past years from clover and alfalfa for dairy and cattle production to soybean production, honey crops have gone down.

Argentina is at the vanguard of promoting proper modes of production and beekeeping practices. The Argentine honey industry also promotes sophisticated and ongoing research on honey.

Argentina and Brazil both suffered serious economic and human consequences from the pandemic. Some internal transportation restrictions impeded movements of both bees and honey.

Brazil

In December, small amounts of honey were being produced in the Serrano region and parts of the Northeast, at high prices. Marmeleiro, some Angico, and polyfloral Extra Light Amber and White precede the main crops that will come in the first quarter of 2022. The condition of the bees is good. Not many offers were being made in early December, as beekeepers were awaiting clarity on the market and the U.S. antidumping rates.

Brazil Honey Exports Jan-November 2021							
Destination	U.S. Dollar Value						
World	45,507,967	\$157,019,658					
USA	32,773,333						
Europe	8,650,877						
Canada	2,546,729						

Canada

About 13,000,000 pounds of honey were exported from Canada in the first 10 months of 2021, according to Canada's government export data. The demand for Canadian honey appears to be considerably greater than the supply left in beekeepers' hands. Producer prices climbed from \$1.25/lb. (U.S.) in June 2020 to 2.18/lb. in January 2021. After the antidumping suit was filed in the U.S. in April, prices went up to \$2.40-2.70/lb. to Japanese and U.S. buyers.

Canadian beekeepers reacted to the antidumping rates with concern that there could be massive increases in volumes from countries that have never been suppliers to North America. There was shock at learning the preliminary determination rates and their anomalies.

There are reports of heavy bee losses in some regions, and weak bees that may not make it through the winter. The number of healthy colonies for one of the large beekeepers was 70% of normal as winter arrived. Northern regions of Canada have had major swings in temperature variation from day to day, and more wind through the season.

Canadian Honey Exports Jan-October 2021							
Destination Kilos U.S. Dollar Value							
World	5,875,016	\$35,008,000					
Japan	3,469,331	\$21,429,000					
USA	2,091,691	\$11,725,953					

U.S. imports from Canada in 2020 totaled 4,142,000 kilos. Japan has become an increasingly important market for Canada, and beekeepers are exploring export opportunities in the U.K., the EU and the Middle East.

A decade ago, 80% of Canada's honey exports went to the U.S. Japan now represents Canada's largest market, and prices to Japan are much higher. There is much less need to compete with adulterated honey. Although there are signs of high varroa counts for some beekeepers, Canada is gearing up to increase honey production. The bee numbers have recovered in Alberta, with increases of over 32,000 colonies in autumn 2021. With higher prices to all markets, beekeepers are optimistic. Rod Scarlett of the Canadian Honey Council reports that the Council is planning to promote Canada's high-quality honey at international trade shows in New Delhi, Shanghai, Paris, Jakarta and Ufa (Russia).

Mexico

In 2021 there was a ten percent increase in honey production over 2020. The 22,000 metric ton crop of 2020 was the lowest in 40 years. Mexico is anticipating favorable weather conditions for the 2022 crop.

At present, the conditions of bees are generally strong throughout Mexico. The beekeepers attribute this to an increase in prices of 80% in the past year which increase has re-incentivized beekeepers for diligent

care for the beehives and new investments. As Mexican friends have said, "Beekeepers have returned to care for the hives." The attitude is very positive and hopeful. In the 4th quarter of 2021 demand was frozen because the industry was waiting for the decision on antidumping. After the decision, prices increased about 10%. Since the preliminary decision may change in respect to both rates and the imposition of critical circumstances, significant uncertainty remains.

Mexican Honey Exports Jan-September 2021						
Destination	US Dollars					
World	29,266,686	\$107,714,102				
Germany	12,581,408	\$ 46,898,380				
UK	2,656,651	\$ 9,317,111				
USA	2,611,592	\$ 9,249,789				

Mexico has been a leader in the fight against economically motivated adulteration. They are actively participating in crucial scientific research projects on honey authenticity.

The macro environment

The global macro environment has been deeply influenced by both the pandemic and the climate crisis. The flow of goods and the flow of labor for the beekeeping industries have been impeded. Lack of visas hurt many beekeeping operations.

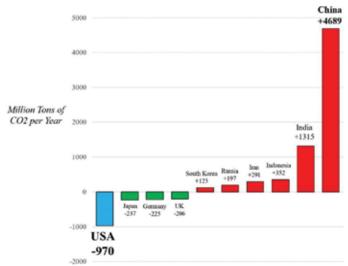
Unfortunately the pernicious pandemic has not disappeared. There are new epicenters, new surges and mutations. All of this has consequences in respect to human health, emotions, economics and education.

International freight rates have soared, increasing from \$3,000 to \$25,000 for one container. Long delays for vessels to enter ports, difficulty or inability to book export shipments, lack of dock workers, and costs for ocean freight which have gone up 500% in 2021, have all contributed to the supply chain crisis.

The pandemic has also influenced availability of labor and permission to enter seaports. The global supply crisis, mainly due to the transportation bottlenecks and astronomical freight prices, has influenced the honey industry and the packing industry.

2005 to 2020 Change in CO2 Emissions

Annual Million Metric tons of CO2 Difference



Source: Our World in Data

While China tries to skirt any responsibility for the Covid pandemic, and to pose as a champion of protecting the global climate, an interesting chart shows the disharmony between words and deeds. A recent study of CO2 emissions, by country, revealed that the U.S. emissions declined dramatically, while China's increased exponentially over the last 15 years:

China's emissions are 150% greater than those of the U.S. and Europe combined. Only China had an increase in emissions during the pandemic years.

Honey testing

The overall picture remains very dynamic. As mandated under a Congressional authorization bill, Nuclear Magnetic Resonance technology is expected to be utilized for testing both country of origin and adulteration by Customs and other agencies. With antidumping duties in place, the temptation for circumvention rears its ugly head. Both circumvention of new antidumping duties and economically motivated adulteration of honey face more stringent and powerful testing capacity in an expanded and more sophisticated toolbox.

Apimondia identified 6 modes of honey adulteration in 2019:

- · Immature honey
- Addition of sugar syrups
- Use of resin technology
- Improper use of antibiotics, fungicides, pesticides
- Excessive bee feeding at time of foraging
- Adding extraneous pollen

As honey from major producers such as Canada, the U.S., and Argentina enter the eastern markets, the contrast between authentic honey and adulterated products will become clearer. The demand for and appreciation of authentic honey will be felt internationally, including from those countries which have practiced illicit modes of production.

The judicial arena

A class action suit is working its way through the federal court system. The complaint is in the public records and makes serious allegations regarding depressing markets for authentic honey by compelling U.S. beekeepers to compete with honey adulterated in any of the several modes cited by the U.S. Pharmacopeia and Apimondia. The filing contends that unfair competition arising from food fraud is a result of collusion and violation of antitrust law.

In the U.K., legal action is being considered to address violations of country-of-origin designations for honey.

Discovery in the U.S. case was expected to begin as early as January 2022. The UN FAO has a memorandum on Food Fraud nearing authorization.

Given the global pandemic, the international transportation crisis, the supply chain crisis, and environmental pressures, there are multiple factors that are leading to price escalation for food and other essentials. In this context, we may see an end to price gouging in the honey and a reincentivization of beekeepers throughout the world.

The U.S. and international markets have entered a stage in which market manipulation has become more difficult and the need to reward beekeepers for high-quality authentic honey has never been greater.

There is a profound shift in momentum and direction in the international honey market.

Given the major deliberations in the U.S. Department of Commerce, filings in the judicial system against antitrust activities and food fraud, and continuing research on honey authenticity, we can remember, as we say in baseball, the game's not over until the fat lady sings. ■

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EurBeST – a pilot study testing varroa resistant bees under commercial beekeeping conditions

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ccording to official reports, there are almost 16 million honey bee colonies in the EU, managed by about 600 thousand beekeepers, with an annual honey production of almost 2 billion Euros. On top of the value of the direct production of beehive products, honey bees contribute to pollination services of agricultural crops and of wild flora, ensuring maintenance of biodiversity and of healthy agroecosystems. Honey bees are, however, under huge stress due to intensification of agricultural practices as well as climatic changes and globalisation, which bring new diseases to bees. Amongst them is the parasitic mite Varroa destructor, which can lead to the death of most infested colonies within one year, if no treatment is applied by beekeepers.

Varroa mites feed on the adult bees and bee pupae and, during this process, can transmit deadly viruses. Since its arrival in Europe in the late 70s, varroa now infests most colonies, and represents the most impacting pathogen threat for honey bees and the beekeeping industry worldwide. Beekeepers only have limited solutions to control the mite without incurring risk of residues in hive products, secondary effects on honey bees and inducement of treatment-resistant mites. A promising and sustainable solution emerges from numerous reports worldwide that some honey bee populations are able to survive mite infestation in the absence of treatments. These survivor bees develop defences to maintain the parasite population under control. As this ability can be transmitted to the next generation, it opens up the possibility for beekeepers to specifically select and breed for varroa resistant bees.

Goals and methodology of the EurBeST study

In 2017, the European Commission contracted an international bee research consortium (European Bee Selection Team = EurBeST) under the lead of the Bee Institute in Kirchhain, Germany to answer the following questions:

- What is the status and extent of the honey bee breeding and reproduction market in the EU?
- What is known about varroa resistance? Do varroa resistant bees exist in the EU? Are they available for beekeepers to use?

- Are beekeepers interested in using varroa resistant honey bees? What do they expect when they buy honey bee queens?
- What methods are available for selecting varroa resistant bees? Do they work?
- What are the efforts and costs to obtain varroa resistant honey bee stock?

The EurBeST consortium involved experts in beekeeping, bee biology, breeding, economics and statistics. They analysed the EU market for honey bee reproductive material (Fig. 1) and ran a literature review and expert interviews on the state of play in varroa resistance. Selection programs on varroa resistance were reported in 20 EU countries and naturally resistant populations in six. However, commercially available resistant stock was found to be present in only four countries.

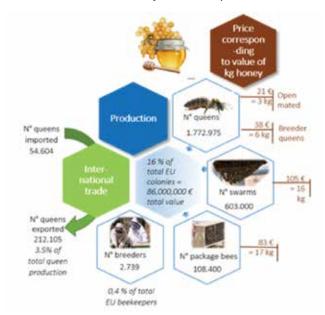


Fig.1: EU market for honey bee reproductive material

Customer survey on breeding stock

A customer survey on the current queen market revealed high expectations, but moderate satisfaction. European beekeepers want to buy high quality queens which, most importantly, express disease resistance and a good productivity. However, they are least satisfied with the disease resistance compared to the other traits (Fig. 2). Almost 50 % of the customers trust in selection as an important, or the only, tool to achieve treatment-free bee keeping. Interestingly, the approval rate was higher in countries with a long tradition of selective breeding.



Fig. 2: Results of an online survey on expectations and satisfaction about marketed queens by 396 beekeepers from different European countries

The largest ever study on honey bee selection

As the core part of the EurBeST project, five large-scale case studies including seven EU countries and 130 participating beekeepers were carried out (Fig. 3). The EurBeST team identified and selected 23 lines, belonging to six subspecies and also of mixed origin, from surviving populations or from varroa-resistance selection programs. They were tested for their general beekeeping and resistance traits on two different levels: on the one hand by performance testers who extensively compared several lines within the same apiary and, on the other hand, by commercial beekeepers who compared one or several test lines with their own stock under normal field conditions. With more than 3,500 colonies tested for one whole season, this constitutes the largest investigation on honey bee selection ever conducted in Europe.



Fig. 3: EurBeST case study countries (in yellow) with dots marking the position of the 130 test apiaries involved. The German case study included testing sites in Austria and Croatia, and the Italian one included a smaller separate case study in Sicily.

Higher resistance of selected stock

The EurBeST selected lines showed similar survival rates to the beekeepers' own stock. While there was on average not much difference for the general traits (honey production, defensive behaviour and swarming tendency), the EurBeST lines clearly outperformed the commercial beekeepers' own stock with regard to mite infestation (Fig. 4).

In the performance test apiaries, which refrained from any mite treatment during the one-year duration of the investigation, some EurBeST lines kept infestation with varroa below the 3% infestation threshold for required mite treatment until the end of the season (Fig. 5). Some of the selected lines demonstrated high productivity, combined with low varroa infestation.

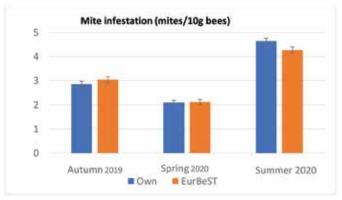


Fig. 4: While starting with higher mite infestation levels in autumn 2019, the EurBeST lines were on average less infested compared to the commercial beekeepers' own stock by the end of the test season in summer 2020

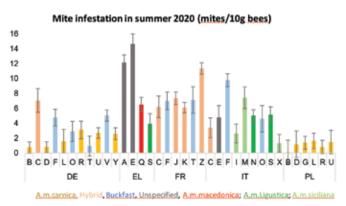


Fig. 5: After a full season without any treatment against varroa, the infestation of several lines clearly remained below the 3% infestation threshold for required mite treatment, showing promising avenues for a treatment-free beekeeping (letter codes present different EurBeST lines, and two-letter codes present case study countries, columns show mean values with standard errors).

Varroa resistance traits

When measuring specific varroa resistance traits, we observed that infestation levels closely correlate with the colonies' hygienic behaviour: on average, higher removal of damaged brood (measured by pin-test) corresponded to lower colony infestation by varroa. Lines with a selection history for this trait displayed higher levels of hygienic behaviour (Fig. 6). Varroa Sensitive Hygiene (VSH) also seemed to affect varroa infestation, which was lower in colonies with a higher VSH. The Recapping (REC) trait (that indicates inspection of brood cells on behalf of worker bees) was found to be correlated with VSH, meaning that it was higher in colonies displaying a high VSH. However, the connection of this trait with varroa infestation was not clear, and a similar situation was also found for the Suppression of Mite Reproduction (SMR) trait.

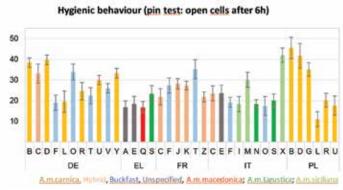


Fig. 6: Different expression of hygienic behaviour (measured by pin-test) among the EurBeST lines (for colour and letter coding see Fig. 5)

Local adaptation is important

The case study results also showed strong interactions between genetic and environmental factors in regulating honey bee colony general performance as well as varroa-resistance potential. Practically, the same line of bees used in two different locations may perform very differently, highlighting the need for local selection strategies (Fig 7). Commercial beekeepers depend on well-adapted stock to reduce disease burdens and to achieve sustainable economic success.

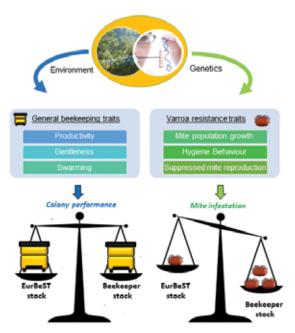


Fig. 7: The selected lines outranged beekeepers' usual stock in varroa resistance, depending on their adaptation to local environmental conditions.

Selection is expensive

As a part of the study, participating queen producers, performance testers and commercial beekeepers were interviewed on their production costs and selling prices. Testing a colony costs 193 € on average, ranging from 273 € in Germany to 85 € in Greece. The main costs of colony evaluation derive from testing for varroa resistance. Monitoring varroa infestation and testing for hygienic behaviour together reach almost 20% of the total costs, while the highest share of the colony evaluation costs, with more than 60% of the total, results from assessing specific varroa resistance traits (SMR, REC and VSH) (Fig. 8).

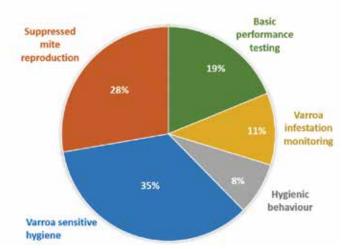


Fig. 8: Contribution of the different testing activities to the overall costs of selection.

Queen price does not cover the costs of selection

The average cost for queen production across the study amounted to $22.58 \in \text{per}$ queen, but with huge variation (from $8.22 \in \text{in}$ Poland to $37.30 \in \text{in}$ France). The main share originates from labour costs, which significantly vary between countries. The average selling price per queen of $23.32 \in \text{sometimes}$ does not cover even the pure production costs. More significantly, it does not in any way compensate the efforts of a serious and continuous selection program, including testing, breeding value estimation and maintenance of the mating stations.

Conclusions and recommendations from the study

Selective breeding of honey bees is an efficient way to increase productivity, to reduce colony losses, and to improve bee health. The use of well-selected stock is a major factor of economic success in commercial beekeeping.

Regional breeding structures are needed to select locally adapted bees. These include cooperation among breeders, queen producers and commercial beekeepers, with scientific support.

Selection for resistance works, but it is costly. Mite infestation development and hygiene behaviour are useful criteria to select varroa resistant stock. However, the costs of testing for the breeders are high and need to be compensated.

The market for queens must be improved. There is a high demand from commercial beekeepers for queens selected for varroa resistance. However, the usual market prices for queens do not cover extra costs for selection. Subsidising the production of high quality queens could help.

Honey bee breeding needs support. The success of breeding programs depends on their scale and consistent development over several years. Considering the high costs for specific selection methods towards improved varroa resistance, public funding of the beekeeping breeding sector is recommended and beekeeper associations should lobby for this.

Reference of the complete EurBeSt published report:

European Commission, Directorate-General for Agriculture and Rural Development: EurBeST Pilot Project: Restructuring of the Honey Bee Chain and Varroa Resistance Breeding & Selection Programme, Final Study Report AGRI-2017-0346. Brussels, 2021, DOI: 10.2762/470707

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Identification of genetic determinants of antimicrobial resistance and virulence in Canadian isolates of Melissococcus plutonius

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uropean foulbrood (EFB), a bacterial disease affecting honey bee larvae, is a re-emerging threat for the Canadian beekeeping industry, with EFB outbreaks anecdotally reported to be increasing in severity and incidence (1). The bacterium responsible for this disease, Melissococcus plutonius, was detected in 37% of honey bee colonies in Alberta by molecular methods (1), with an annual estimated economic impact to the Alberta beekeeping industry ranging from \$158,504 to up to \$35.37 million in an outbreak scenario(1). In combination with integrated pest management (IPM) techniques, the antibiotic oxytetracycline is widely used by Canadian beekeepers to treat EFB in their colonies. Considering that antimicrobial resistance has been reported in Paenibacillus larvae (2), the bacterium responsible for American foulbrood, there is concern that antimicrobial resistance in M. plutonius may explain the reported increase in EFB disease in Canadian apiaries. To investigate this hypothesis, we performed whole genome sequencing of 37 Canadian isolates of M. plutonius, as well as 8 American isolates of M. plutonius, to investigate for the presence of antimicrobial resistance (AMR) genes, in combination with antimicrobial susceptibility testing. Additionally, using pangenome analysis, we characterized the genetic diversity of M. plutonius isolates from Canada and the USA, and investigated these isolates for the presence of putative virulence genes. Specifically, we screened M. plutonius isolates for the plasmid-encoded melissotoxin A (mtxA) gene, which is hypothesized to encode a toxin that may increase the pathogenicity of M. plutonius to honey bee larvae (3,4).

Materials and Methods

Whole genome sequencing of 45 isolates of M. plutonius was performed using Illumina MiSeq PE250, NovaSeq6000 PE150 (Genome Quebec) and PacBio (Maryland Genomics). The genome sizes ranged from 2.03 megabase pairs (Mbp) to 2.18 Mbp and each genome contained between 1799-2025 coding DNA sequences. Genomes were analyzed for the presence of antimicrobial resistance (AMR) genes using the Comprehensive Antibiotic Resistance Database (https://card.mcmaster.ca) and investigated for the presence of the melissotoxin A gene (mtxA) using BLAST+ 2.11.0 (https://blast.ncbi.nlm.nih.gov/Blast. cgi). Multi-locus sequence typing (MLST) of the isolates is in progress using the established MLST database (http://pubmlst.org/mplutonius/) (5). Phylogenetic analysis of the 45 M. plutonius genomes from this study, in addition to 18 M. plutonius genomes in the public domain, was performed based on comparison of 1483 core genes using Bacterial Pan Genome Analysis (BPGA) (6). Antimicrobial susceptibility to the three antibiotics licensed for use in beekeeping (oxytetracycline, tylosin, lincomycin) was performed for 22 M. plutonius isolates by establishing the Minimum Inhibitory Concentration (MIC) in µg/ml for these antibiotics using the broth microdilution method (7).

Results and Discussion

We examined the genomes of 37 Canadian isolates of *M. plutonius* (15 from British Columbia, 2 from Alberta, 4 from Saskatchewan, 16 from Quebec) and 8 American isolates (2 from each of Michigan, Oregon, Texas, and Utah) collected from 2007-2021 (Table 1). Preliminary phylogenetic analysis of these North American isolates (Figure 2) revealed the isolates clustered in two groups which were distinct from *M. plutonius* isolates sequenced previously from Japan, Switzerland, Norway, England, and the USA. Multi-locus sequence typing of the isolates is in progress; however, one of the isolates from Saskatchewan (2020SK1)

▶ pag. 22

was found to represent a novel genetic sequence type (8), supporting the hypothesis that Canadian apiaries may harbor new genetic variants of this pathogen. 49% of *M. plutonius* isolates in this study were found to carry the mtxA gene, while 51% did not (Table 1), suggesting that spread of *M. plutonius* strains carrying mtxA may not explain anecdotal reports of increased clinical severity of EFB in Canada, although additional field data is necessary to investigate further.

To date, no canonical/well-established AMR genes have been identified in the 45 M. plutonius genomes analyzed. In contrast, oxytetracycline (OTC)-resistance was observed in 21/22 isolates examined for antimicrobial susceptibility. OTC-resistant isolates had MICs ranging from 8-64 µg/ml OTC (Table 1), which were in excess of the susceptibility breakpoint of 2.5 µg/ml OTC established by the Clinical and Laboratory Standards Institute (CLSI). Only one Canadian M. plutonius isolate (2017QU1) was considered susceptible to OTC with an MIC of 4 µg/ml, which is less than two-fold greater than the CLSI breakpoint value. Although CLSI breakpoint values are not established for tylosin (TYL) and lincomycin (LMC) for M. plutonius, all isolates examined had MIC values of 4 µg/ml or less for these antimicrobials, suggesting that the M. plutonius isolates were susceptible to TYL and LMC (Table 1). Importantly, TYL and LMC are not licensed for treatment of EFB in North America.

Conclusions

The population of M. plutonius isolates in Canadian honey bee colonies is genetically distinct from other regions of the world, and contains at least one previously undescribed genetic variant. Based on laboratory testing, OTC-resistance is common among Canadian isolates of M. plutonius; however, to date, whole genome sequence analysis of these isolates did not identify any AMR genes to explain this resistance phenotype. Despite the observed OTC-resistance of M. plutonius in vitro, OTC may still be effective at treating colonies with EFB in the field in combination with the inherent social immune defenses of honey bee colonies, as well as other IPM strategies recommended for EFB. OTC is the only licensed antimicrobial for treatment of EFB in North America and our results do not support the use of other antimicrobials for treatment of EFB at this time.

Future directions

Future studies will explore the most effective antimicrobial dosing strategies for treatment of EFB in both the laboratory and the field. Additionally, we will continue to investigate the genetic diversity of Canadian *M. plutonius* isolates, as well as characterize the pathogenicity of these isolates to honey bee larvae. Taken together, our current and future research will enhance antimicrobial stewardship and management of EFB within the Canadian beekeeping industry and, in turn, improve the productivity and profitability Canadian honey bee colonies.

Isolate Name	Year of isolation	Province / State	mtxA	OTC MIC	TYL MIC	LMC MIC
2020AB1	2020	Alberta, Canada	no	32	2	0.5
2020AB2	2020	Alberta, Canada	no	16	1	0.5
2020BC1	2020	BC, Canada	yes	16	1	0.5
2020BC2	2020	BC, Canada	no	32	1	0.5
2020BC3	2020	BC, Canada	no			
2019BC1	2019	BC, Canada	yes	32	2	0.5
2020BC4	2020	BC, Canada	yes	16	2	0.5
2021BC1	2021	BC, Canada	no			
2021BC2	2021	BC, Canada	yes			
2021BC3	2021	BC, Canada	yes			
2021BC4	2021	BC, Canada	no			
2021BC5	2021	BC, Canada	yes			
2021BC6	2021	BC, Canada	yes			
2021BC7	2021	BC, Canada	yes			
2021BC8	2021	BC, Canada	yes			
2021BC9	2021	BC, Canada	yes			
2021BC10	2021	BC, Canada	yes			
2020MI1	2020	Michigan, USA	yes	64	2	0.5
2020MI2	2020	Michigan, USA	no	16	2	1
2020OR1	2020	Oregon, USA	no	32	2	1
2020OR2	2020	Oregon, USA	no	8	1	0.5
2007QU1	2007	Quebec, Canada	no			
2008QU1	2008	Quebec, Canada	no			
2015QU1	2015	Quebec, Canada	yes			
2016QU1	2016	Quebec, Canada	no	32	2	1
2016QU2	2016	Quebec, Canada	yes			
2017QU1	2017	Quebec, Canada	yes	4	0.5	<0.25
2017QU2	2017	Quebec, Canada	yes			
2018QU1	2018	Quebec, Canada	no	16	1	0.5
2018QU2	2018	Quebec, Canada	no	16	1	1
2018QU3	2018	Quebec, Canada	yes			
2020QU3	2020	Quebec, Canada	no			
2008QU2	2008	Quebec, Canada	no			
2019QU1	2019	Quebec, Canada	no			
2019QU2	2019	Quebec, Canada	no			
2020QU1	2020	Quebec, Canada	no			
2020QU2	2020	Quebec, Canada	yes			
2020SK1	2020	Saskatchewan, Canada	no	8	1	<0.25
2021SK1	2021	Saskatchewan, Canada	yes	16	2	1
2021SK2	2021	Saskatchewan, Canada	yes	16	0.5	<0.25
2021SK3	2021	Saskatchewan, Canada	yes	32	2	0.5
2020TX1	2020	Texas, USA	no	16	4	1
2020TX2	2020	Texas, USA	no	16	4	0.5
2020UT1	2020	Utah, USA	yes	64	1	0.5
2020UT2	2020	Utah, USA	no	64	2	0.5

Table 1. Origin of 45 isolates of Melissococcus plutonius submitted for whole genome sequencing and evaluated for the presence of the melissotoxin A gene (mtxA). Minimal Inhibitory Concentrations (MICs) in μg/ml of the antimicrobials oxytetracycline (OTC), tylosin (TYL) and lincomycin (LMC) were determined for 22 isolates.

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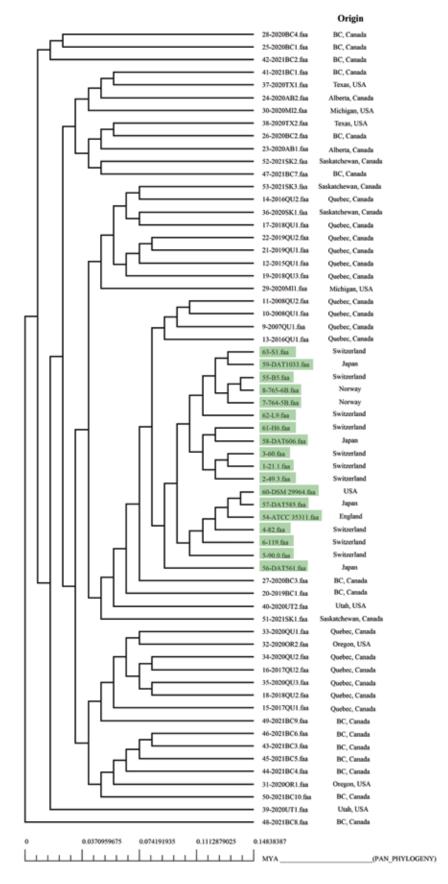
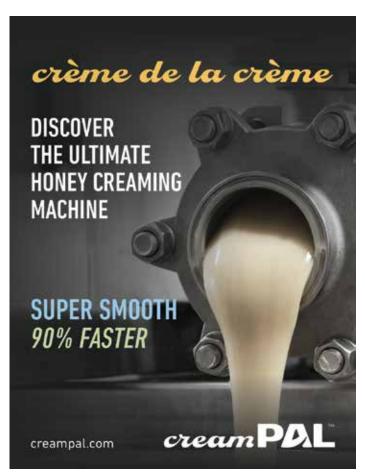
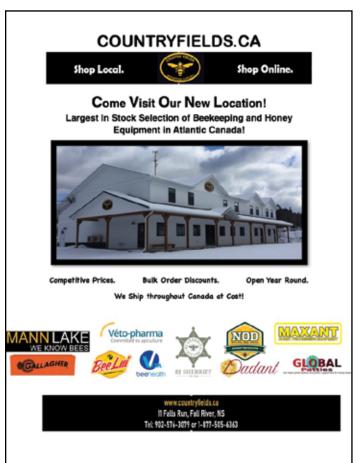


Figure 2. Phylogenetic analysis of 45 isolates of Melissococcus plutonius from this study, in addition to 18 isolates of M. plutonius whose genomes are in the public domain (highlighted in green), based on comparison of 1483 core genes using Bacterial Pan Genome Analysis (BPGA).









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Project Title: Field Trials of a New Acaricidal **Compound Against Varroa** destructor in Honey Bee Colonies

Project Leads: Dr. Steve Pernal - Agriculture and Agri-Food Canada Steve.Pernal@agr.gc.ca (PI) Dr. Erika Plettner - Simon Fraser University plettner@sfu.ca (Co-Pl) Project Duration: June 2021 - March 2024

Project Description:

We have discovered a new acaricide, 1-allyloxy-4-propoxybenzene (code: 3c{3,6}), with strong acaricidal activity against Varroa destructor, having no appreciable toxicity towards bees or vertebrate animals. This compound has been tested extensively in the laboratory and once previously in field trials in British Columbia and Alberta, during the fall of 2019. The mite mortality caused by compound 3c{3,6} in these trials was comparable to that of other commercial acaricides in use.

The current project is designed to generate additional data towards product registration under field-realistic conditions and further finetune release devices and application methods. These trials will also allow the collection of wax and honey samples to determine residual levels of compound 3c{3,6} after applications in the fall and spring.

Progress Towards Goals:

Field work to evaluate compound 3c{3,6} started this fall using two identical field experiments, one in the in the lower mainland of BC and the other in Beaverlodge, AB.

In preparation for the work in Alberta, Mr. Robert Lu was recruited as a new Masters' student, in the Department of Biological Sciences at the University of Alberta, co-supervised by Dr. Steve Pernal and Dr. Olav Rueppell. Robert was resident in Beaverlodge this fall to conduct the experiment at that location with assistance from AAFC technicians Abdullah Ibrahim and Rassol Bahreini. Field work in the lower mainland of BC was led by Jorge Enrique MacIas-Samano of the Plettner lab with the assistance of Carolyn Essaunce.

The treatments for these experiments consisted of ten replicate colonies of the following:

A. Compound 3c{3,6} - 4 g with 1.26 g glycerol, administered via a single porous wooden release device, per brood box. These were placed on top of the frames, above the broodnest.

B: Thymovar - one 15 g strip per colony, with only one Thymovar device used for the 28 day treatment period. One strip was cut into two halves and placed in diagonally-opposing corners of the broodnest, on top of the frames.

C: Empty Release Device - wooden release device (treated with solvent only and 1.26 g glycerol). Placed on top of frames, above the broodnest.

All 30 colonies per experiment were located within the same apiary, and were equalized as much as possible for brood, pollen and honey stores, as well as varroa levels, over the preceding summer. Colonies at both locations were derived from New Zealand package bees in May 2021, having new-year Olivarez queens installed. Bees were maintained in single brood chambers and situated on screened bottom boards with their entrances reduced. Hives were formally assessed for adult bee and brood populations prior to experimental Day 0 (Sept 3) and at Day 28.

Porapak devices were also installed in all colonies on Day 0 and replaced on Day 14. These remained in place until Day 28, when they were replaced again with new devices for post-treatment monitoring. Poropak absorbs organic volatiles and will allow us to determine airborne concentrations of 3c{3,6} and thymol.

On experimental Day 28 (Oct 1) all treatments were removed. Apivar strips were inserted and remained in place until Day 70 (Nov 12), to determine post-treatment varroa levels in colonies and thus enable the calculation of treatment efficacies. Mite mortality was evaluated by counting mite "fall" on sticky boards at prescribed intervals, and alcohol washes of adult bees were taken on Days 0, 28 and 70 to determine phoretic mite densities. The number and reproductive status of mites in 100 sealed brood cells per colony was also evaluated prior to the start of the experiment and again on Day 28. Finally, cluster sizes were scored on Day 70, before wintering.

The experiment in Beaverloge started with considerable mite pressure, with colonies having an average phoretic mite density of 9.2 \pm 0.8%, while levels in BC were 2.0 \pm 0.2% prior to the application of treatments.

Full data will be presented in our March 2022 report. Data collected to date suggest that compound 3c{3,6} performed similarly to Thymovar and slightly less effectively than during our fall 2019 experiment. For field trials in upcoming seasons, we will re-evaluate the amount of 3c{3,6} impregnated per release device (i.e. this experiment compared 4 g 3c{3,6} against 15 g of thymol), and we will also extend the treatment period over a full 6 weeks. Lastly, we will be analyzing wax samples collected from colonies in the fall of 2021, and samples of honey from the same colonies during the summer of 2022, to determine incurred residues of $3c\{3,6\}$.

Funders:

Project Apis m., Alberta Beekeepers Commission, Canadian Bee Research Fund, Bee Maid Honey





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Research bulletin: Tracking temperatures of nuc shipments from Vancouver to Whitehorse in June, 2021

Alison McAfee, Jonathan Jakes, Marlene Jennings

ransporting honey bees can be a risky business. On top of hazards like leaking bees or shipping delays, extreme temperatures can damage the queen's fertility and reduce the productivity of her colony. With rising frequencies of extreme weather events combined with growing interest in domestic sustainability of bee supplies, it is time to re-evaluate how and when we ship bees.

Hot and cold temperatures can cause queen infertility by killing the sperm stored within the spermatheca,¹⁻³ and my previous research has shown that failing queens have more dead sperm than healthy queens.^{3,4} So far, most studies have focused on temperature fluctuations in shipments of caged queens,¹⁻³ where the bees have limited thermoregulatory capacity and are vulnerable to the ambient temperatures.

Temperature fluctuations within nucs, however, have seldom been investigated, despite the fact that nucs are also shipped within and between provinces and territories. One study documented temperature fluctuations experienced by box-style packages shipped on a flat-deck truck in the U.S., and identified a link between temperature fluctuations and subsequent colony outcomes.⁵ Enclosed nucs with frames of bees and brood may experience different temperatures, though, since nuc shipping boxes are generally not as well ventilated as packages.

Methods:

In June 2021, we investigated temperature fluctuations in three shipments of nucs that were delivered from Vancouver to Whitehorse via Air North Ltd., a small airline with a limited schedule and cargo capacity. Months ahead of time, a plan was agreed upon by the airline and supplier to ship three batches of 16 nucs each. The nucs were produced locally in BC's lower mainland and made up of four frames of bees, brood, food, and drawn comb contained in Ontario-style corrugated plastic shipping boxes. The boxes were engineered to have extra vent holes (approximately 180 cm² total meshed ventilation, or 60 cm² each), such that each box had one vent on the top and two on opposite sides. Shipment 1 was scheduled for June 1st, Shipment 2 on June 2nd, and Shipment 3 on June 7th).

The nuc boxes were strapped to a shipping pallet and spaced to avoid obstructing the vent holes. We included temperature loggers taped under the lid of a subset of boxes (Figure 1) in each shipment, with the loggers sealed in stamped envelopes for the recipient to mail back to the UBC laboratory. Each nuc was also enveloped in a light mesh transport bag to prevent bee leaks. Upon arrival in Whitehorse, Air North accommodated an immediate pick-up for the bees from the airport, and the boxes were kept outside in the shade until the recipients arrived to claim their nucs.

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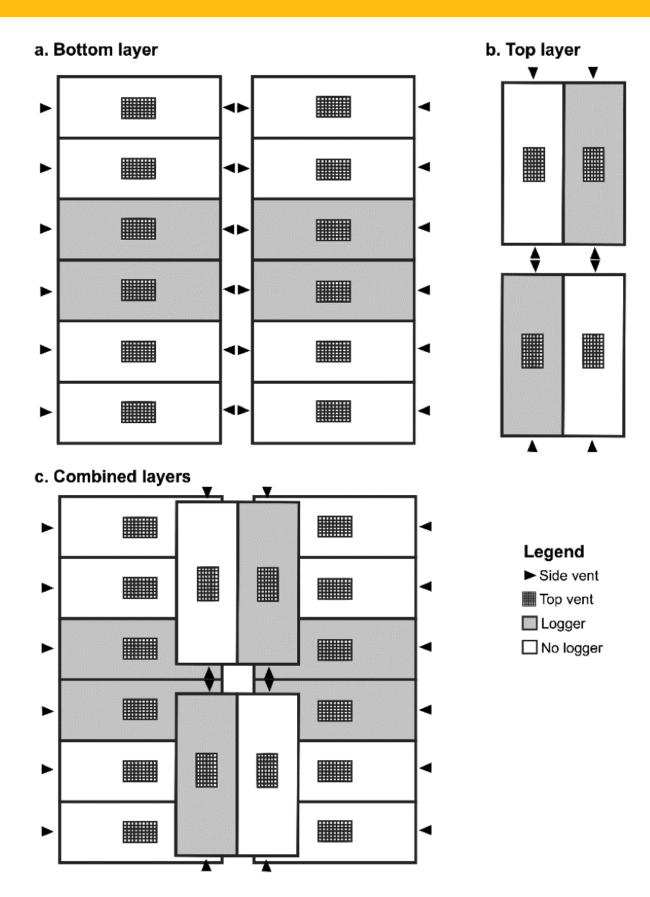


Figure 1. Nuc shipment orientation. Combined layers show the final nuc arrangement on the shipment pallet.

Results:

The daily highs at the Vancouver departure location for the three respective shipment dates were 24.1°C, 28.2°C, and 15.6°C (airport data obtained from Environment Canada). In Whitehorse, the corresponding daily highs were 17.8°C, 17.2°C, and 19.4°C. We obtained temperature data for four, six, and two nucs in shipments 1, 2, and 3, respectively (designated as nucs 1 - 12; not all temperature loggers were returned). We found that under the lid, temperatures fluctuated from 20°C up to 48°C in one instance (nuc 5) – well beyond the "safe zone" for queens, which is defined as 15 - 38°C based on the best available data.3

Worker honey bees have been previously shown to die at 48°C,6 and indeed, the nuc in Shipment 2 experiencing extreme heat unfortunately perished during transit. Other nucs (nucs 6, 8, and 9) in Shipment 2 experienced temperatures between 39°C and 41°C, a range which can lead to partial loss of queen fertility but which does not cause worker mortality.

constructed of a more breathable material (e.g. un-waxed cardboard or framed wire mesh), more like package bee containers, that can still allow for evaporative cooling. The nuc boxes used in these shipments are designed for shipping, and even had extra vent holes than the standard design. There was ample ventilation for oxygen flow but this was apparently not enough to enable sufficient cooling. Different materials should be tested to find the best balance between structural integrity and ventilation capacity, so that shipped bees can have the best outcomes possible.

Acknowledgements:

Thank you to all the Yukon beekeepers who returned the temperature loggers! We appreciate your support for this study. ■

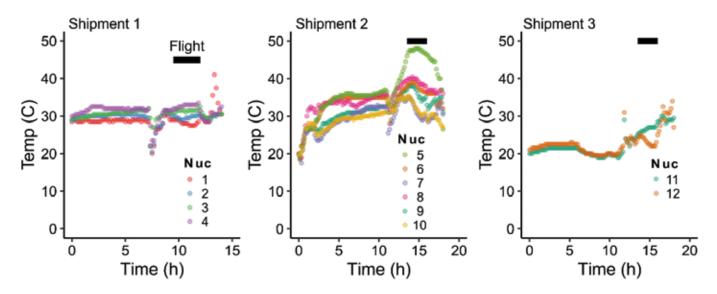


Figure 2. Temperature data for three shipments from Vancouver to Whitehorse. Each color corresponds to a different nuc. The black bar indicates the time during which the nuc was in flight. Time = 0 h corresponds to midnight prior to departure.

Recommendations:

Several factors could influence the outcomes for shipped nucs, such as ambient temperature at the departure and arrival locations, ambient temperatures during transit, bee density within the nucs, construction material of the nuc boxes, and degree of ventilation. Ambient temperatures were the hottest on the day of Shipment 2, which was also the only shipment with nuc mortality. The temperature of the Shipment 2 nucs was stable in the early morning but began climbing ahead of the actual flight, which indicates that the ambient temperature on the ground in Vancouver probably contributed to loss of life. Future shipments should aim to depart at night or early in the morning, if possible, to avoid high temperatures during the late morning and afternoon. If airline delays push departure times into the late morning or afternoon, the shipment should be cancelled to ensure animal welfare.

Honey bee density within the nuc may have also been a contributing factor. Strong (populous) nucs are desirable in order to grow into robust colonies once received by the beekeeper, but having too many bees in a confined space could elevate risk of temperature stress during transit. One way to help avoid this problem may be to use nuc boxes

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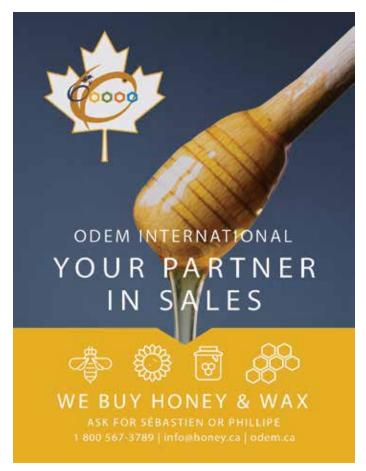
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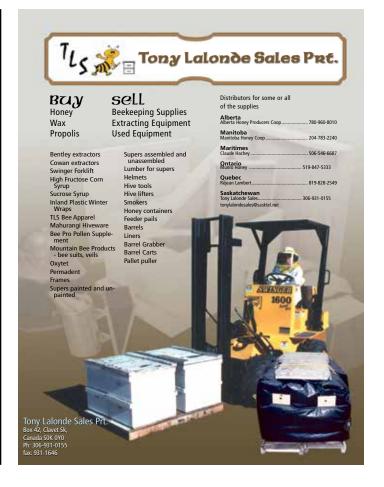






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Project title: Requeening with queen cells in canola pollination

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Introduction

Canadian beekeepers cite poor quality queens as one of the most common causes of colony losses (CAPA 2021) and report needing to replace half of their queens each season (Amiri et al., 2017). There are many ways to introduce a new queen but being able to requeen a colony without removing the old queen would save beekeepers a lot of time and manual labor. Unfortunately, experiments from the 70's and 80's showed requeening queenright colonies with queen cells or virgins is not typically successful (Jay, 1981; Boch and Avitabile, 1979; Forster, 1972). For example, Szabo (1982) found that of 474 introductions in Beaverlodge in 1978-79, queens introduced as a cell only successfully headed 12.7% of the colonies, making queen introductions to queenless colonies the recommended practice.

It has recently come to our attention that some commercial bee-keepers in Alberta have been performing queenright queen cell introductions with enough success to continue the practice. While their successes are anecdotal, it does beg the question, would the results from these 40-year-old experiments be supported today? Much has changed in beekeeping and the environment in the last 40 years. There have been great leaps in technology and improvements to apiculture practices. But most interestingly, and perhaps more importantly is, a honey bee's world is vastly different today than it was 40 or more years ago, and it is not clear if repeating these experiments would deliver similar results.

The environmental and physiological stressors plaguing honey bees and apiculture industries today are not only changing the way beekeepers manage their honey bees but may also be changing honey bee biology and the way honey bees interact with one another and their environment. For example, breeding queens and selecting queen breeders for their desired traits (e.g., hygienic behaviours, parasite and pathogen resistance, gentleness, and productivity) may be inadvertently selecting honey bee worker behaviors that play a role in new queen acceptance. In addition, queen issues are reported by beekeepers to be a frequent reason for colony loss, indicating that queen quality (or worker accep-

tance) may be poorer now than when previous studies were conducted. Thus, this project set out to test whether new queens can successfully be introduced to queenright colonies to determine whether 40 years of beekeeping, environmental change, and honey bee biology have changed the best management practices for introducing queen cells.

Methods

Previous experiments on queenright queen introductions involved marking the old queen before introducing an unmarked new queen into a colony (Szabo, 1982). After enough time passed, where the new queen should have superseded the old queen, experimenters would simply look for the marked old queen. If the marked old queen was found, requeening was unsuccessful, if an unmarked queen was found, requeening was successful. This practice however is labour intensive and can miss situations where multiple laying queens are present. Instead, we can use modern molecular tools to sample honey bees from their colonies and determine their matriline (mothers) from their genetics.

Thus, working with a commercial beekeeper in the summer of 2020, we requeened 100 queenright honey bee colonies located in canola pollination yards in Alberta. Prior to introducing a new queen to each colony, we sampled drone pupae (where possible) and adult workers (where drone pupae were not available) and sent them to the National Bee Diagnostic Centre for maternal source genetic testing. These preintroduction samples would provide the genetic profile of the original queen. Drone pupae are preferred for matriline analysis because they are haploid and therefore carry only maternal genes. Where possible, we sampled drone pupae to ensure we were not sampling drones that may have drifted from another colony.

We grafted 200 queen cells into the research colonies at Alberta Agriculture and Forestry Lethbridge. From these grafts, we selected 100 queen cells (half-sister progeny) to introduce to the 100 colonies. The queen cells were introduced to the highest honey super that was fully occupied by bees. After six weeks (i.e., a window of time long enough to permit new queens to supersede their old queens) we re-sampled drone pupae and adult workers from each colony and sent for maternal source genetic testing. As with the first samples, we preferentially sampled drone pupae when available, and adult workers when drones were not present in the colony. From this data we can distinguish between three possible requeening outcomes: 1) the original resident queen remained laying in the colony for at least 6 weeks, 2) the original queen was superseded by a daughter queen, and 3) the introduced queen cell was accepted and produced progeny.

Results

New queen cells were introduced to 100 queenright colonies. Of these queen cells 87 queens successfully exited their queen cells and 82 colonies were still queenright 6 weeks after queen cell introductions. Thus, the following data is based on 82 queenright colonies. The original queen was found to mother the sampled offspring in 67% of the queenright colonies (Figure 1). New queens were successful in superseding the original queen in only 5% of queenright colonies. We found daughter queen supersedures were more common (14%) than new queen supersedures, and in one colony, our results suggest two queens were present, the original queen and a daughter queen.

Beekeepers could observe a benefit from queenright queen cell introductions if the 5% of colonies that accepted a new queen were those that were in need of a new queen in the first place. There could also be a benefit if the 14% of colonies that reared their own daughter queens were stimulated by a queen cell introduction to rear their own daughter queens. However, the daughter queen supersedures (14% of colonies) could also have been the product of a swarming event (although we saw no evidence that this was so, and all colonies had adequate honey supers to provide room for large populations).

We are still working to resolve the maternity for the 11% of colonies that were inconclusive. Regardless, our results clearly agree with studies from the 70s and 80s (Szabo, 1982; Jay, 1981; Boch and Avitabile, 1979; Forster, 1972) and show that queen cell introductions in the presence of a queen are unlikely to succeed ().

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Maternal Source of Sampled Honey Bees

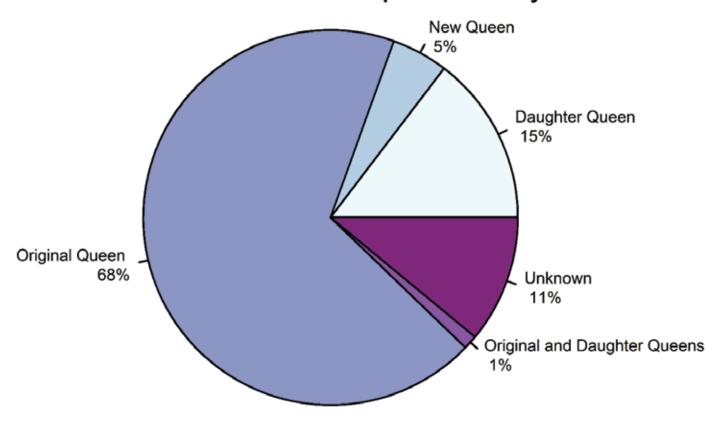


Figure 1. Maternity of sampled honey bee colonies located in canola pollination yards in Alberta, July, 2020. Queenright colonies (N = 82) were sample for drone pupae and worker bees (where necessary) 6-weeks after queen cell introductions to determine maternal source of offspring (e.g., original queen, new queen, or daughter of original queen). Results suggest requeening queenright colonies will more often result in the new queen failing to supersede the original queen ($\chi = 121.64, df = 4, p < 0.001$).



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