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CONTROLLING OF INSECT-PARASITES OF ALFALFA LEAFCUTTING BEESTOCK (*MEGACHILE ROTUNDATA* F., HYMENOPTERA, MEGACHILIDAE)

Jenö FARKAS and László SZALAY
with the technical collaboration of Zoltán TISZAI

Research Centre for Animal Breeding and Nutrition
H-2100 Gödölló, P.O.B. 57, Hungary

SUMMARY

Many millions of *Megachile rotundata* (Fabricius) prepupae were imported from the USA to Hungary between 1972 and 1978, but the general introduction of alfalfa leafcutting bee to Hungary planned for 1974 and 1975 could not be realized because of the heavy infestation by parasites.

In 1981 we determined the *Melittobia acasta* Walker infestation of cocoons left behind by emerged alfalfa leafcutting bees. In 1982 we counted all the parasites emerged during the incubation period.

In 1981 after the completion of incubation one male leafcutting bee and 4 913 Chalcidoidea emerged from the 1 200 leafcutting bee cocoons during 50 days ; they died without food within 10-14 days.

Melittobia acasta Walker chosen randomly run towards day-light with a velocity of 9-14 m/h.

In 1982 the number of emerged *Megachile* and *Coelioxys* increased by keeping the cocoons at lower temperatures and by treatment with Sevin, while the number of emerged *Melittobia* decreased.

Besides the usual measurements new technological and management methods are needed in order to control the parasites.

INTRODUCTION

The alfalfa leafcutting bee, *Megachile rotundata* (Fabricius) has become one of the world's most important pollinators of alfalfa. Most alfalfa seed growers who use this bee have substantially higher seed yields than those who rely on native pollination. Several bee species, including bumble bees and native leafcutting bees are efficient pollinators, but their populations fluctuate from year to year or even

decrease as nesting habitats are reduced or destroyed by modern agricultural practices.

According to our knowledge today the mass use of alfalfa leafcutting bees is the only way to obtain large-scale alfalfa seed production (BOHART, 1962 ; HOBBS, 1968 ; HOLM, 1982 ; KRUNIC and HINKS, 1972 ; MORGNER, 1973 ; RANK *et al.*, 1981 ; RICHARDS, 1982 ; STEPHEN, 1961).

There are several testing centers in the United States and Canada, where samples submitted by beekeepers are controlled by the method of STEPHEN and UNDURRAGA (1976). This control method takes first of all the quality and parasitism of bees into consideration.

The native population of *Megachile rotundata* is small in Hungary (MÓCZÁR and BÖJTÖS, 1957).

Between 1972 and 1978 several millions of prepupae were imported from the United States and Canada to Hungary. Nevertheless, the general use of alfalfa leafcutting bees for alfalfa seed growing (MANNINGER, 1970) planned for 1974-1975 could not be realized. The main reason for the failure was the heavy parasitism of the stock.

This situation induced us to investigate the possibilities of artificial propagation and methods for controlling parasites of the alfalfa leafcutting bee.

MATERIALS AND METHODS

Our Research Centre was given 1 400 prepupae in 1981 and 1 620 in 1982 from Kompolt (North-Hungary) from the imported stock for experimental purposes.

The number of parasites is followed in the relevant literature only until the emergence of bees (HOBBS, 1968). The principal objective of our tests in 1981 was to determine the infestation rate of cocoons of emerged bees by *Melittobia acasta* Walker. In 1982 our aim was to count the total number of parasites during the emergence period.

The prepupae were held in Kompolt at 0-1 °C for one month in an attempt to kill 96 per cent of the parasites. Before incubation the prepupae were treated with a solution of Carbaryl (0.2 %), aiming at 98 per cent mortality of the parasites.

In 1981 the 1 400 emerging bees were placed in alfalfa fields (variety Tápiószelei-I) in Dömsöd (Middle-Hungary) after being treated with Carbaryl and incubated.

1 200 cocoons left by hosts were carried from the field to the laboratory and kept in polyethylene bags at 21 °C and 70 per cent R.H. for 50 days. The number of newly emerged parasites was recorded every day.

The lifespan of adult *Melittobia* was examined in groups of 3 kept loosely and in groups of 96 and 106 kept jam tight in test-tubes, under the same environmental conditions, without food.

The mobility of *Melittobia* was examined between 23 and 30 July 1981 under similar microclimatic conditions, by randomly choosing 1.4 and 100 females per group, respectively. The movement of the adults towards day-light was followed by drawing it on a piece of 1 m wide cardboard and recording the time.

In 1982 we were given 1 620 overwintered cocoons. One part was treated with chemicals, the other part was used as an untreated control.

The groups were incubated in four different places at 28 °C, according to a rotational plan (Fig. 1).

We succeeded in making good quality radiographs of the prepupae with the MAMMOMAT (GDR) TUR D 240 automatic radiographic apparatus.

Instead of the usual glass vials, paper and polyethylene bags or paper-polyethylene and metal boxes, we used bags of finer texture silk-sieve, used in the milling industry, to separate the several groups.

RESULTS

1.1. Contrary to the well known and applied control methods, the emergence rate of alfalfa leafcutting bees was very poor in 1981 (26.3 per cent). This high mortality rate was caused by *M. acasta* contamination of 48.6 per cent ; 24.8 per cent of the bees perished as larvae, prepupae or adults and 0.3 per cent were infested with chalkbrood. Under the described conditions, i.e. during 50 days after the completion of emergence, one male leafcutting bee and 4 913 *Melittobia* emerged from the 1 200 alfalfa leafcutting bee cocoons.

1.2. Under crowded conditions the *Melittobia* groups kept without food perished in 14 (14-28 September) and 10 (18-28 September) days, respectively. In another treatment 33 per cent of the loosely kept and 56 and 62 per cent of the crowded groups, respectively, perished in 13 days.

1.3. The first *Melittobia* chosen randomly covered a distance of 28.55 m during the three days between 23 and 25 July (it was made to run for an hour every day). It made a total of 130 small jumps, 93 the first, 19 the second and 18 the third day.

The four females made to run on the next occasion exceeded the first female by a velocity of 9.3-14.4 m/hour. The directions of motion and traces of these four females and those of individuals from the third group of 100 were so similar that they could hardly be distinguished form one another.

2.1. In 1982 we examined 57 per cent of the 1 620 incubated preppuae, i.e. 930 individuals by x-rays once, 263 prepuae were examined twice and 66 three times (Fig. 1). The first group was radiographed on the 12th and 19th ; the second group on the 7th, 14th and 21st ; the third group on the 0th, 7th and

14th ; and the fourth group on the 3rd and 10th day of emergence. The content of the unopened cocoons could be examined by this method. We found that 9 per cent of the overwintered prepupae contained small larvae, 39 per cent large larvae and 27 per cent 37 per cent of the prepupae were infested with parasites.

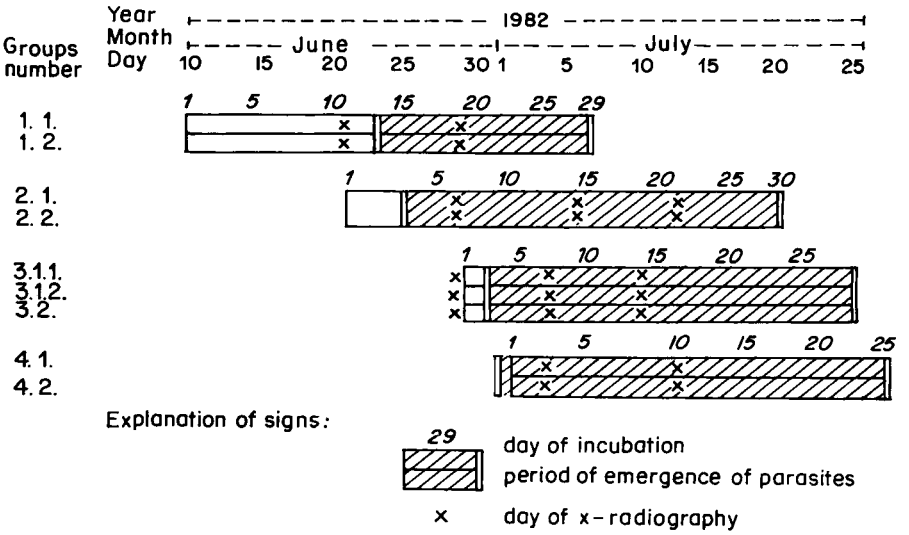

FIG. 1. — *Incubation of* Megachile rotundata *and x-radiography of prepupae (groups 1-4)*

TABL. 1. — *Number and percentage of alfalfa leafcutting bees and parasites emerged from one leafcutting bee stock at different times* (GÖDÖLLÖ, 1982)

Nr. of treatments	Nr. of prepupae	*Megachile*	*Coelioxys*	*Melittobia*	Parasites total
1.1.	55	33	6	192	198
2.1.	250	31	14	430	444
3.1.1.	250	44	32	833	865
4.1.	250	12	15	194	209
Treated total	805	120	67	1 649	1 716
1.2.	65	11	5	316	321
2.2.	250	4	2	1 935	1 937
3.2.	125	3	1	640	641
4.2.	250	5	13	1 674	1 687
Control total	690	23	21	4 565	4 586
Control + treated total	1 495	143	88	6 214	6 302
3.1.2. treated	125	8	4	649	653
Total	1 620	151	92	6 863	6 955

2.2. In the four groups of stock, 7 106 insects emerged from the total of 1 620 cocoons (Table 1) during four consecutive periods. Besides the 151 (9.3 per cent) alfalfa leafcutting bees, two dangerous hymenopteran parasites also emerged : 92 (5.7 per cent) *Coelioxys rufocaudata* Smith (Megachilidae) and 6 863 *Melittobia acasta* (Eulophidae).

2.3. According to the evaluation of data collected, the treatment with Sevin increased not only the emergence rate of leafcutting bees, but also that of *Coelioxys rufocaudata*. Comparing the emergence rate of the treated groups to the control, five times more (120) *Megachile* and three times more (67) *Coelioxys* emerged from the cocoons ; while the 1 649 *Melittobia* were one third of the individuals of the control group.

The highest number of *Melittobia* emerged between the 6th and 10th days of incubation in the treated groups and between the 16th and 20th days in the control ones, respectively (Table 2).

TABL. 2. — *Number of alfalfa leafcutting bees and parasites emerged from the 1.2-4.1 treated and 1.2-4.2 untreated groups* (GÖDÖLLÖ, 1982)

Date of emergence	*Megachile*		*Coelioxys*		*Melittobia*		*Megachile*		*Coelioxys*		*Melittobia*	
	emerged from											
	805-270 containing pollen = 506 treated cocoons						690-180 containing pollen = 506 untreated cocoons					
	Nr	%	Nr	%	Nr	%	Nr	%	Nr	%	Nr	%
1 - 5	—	—	—	—	48	3	—	—	—	—	113	2
6 - 10	8	7	1	1	583	35	3	13	4	19	305	7
11 - 15	13	11	—	—	289	18	2	9	—	—	788	17
16 - 20	37	31	29	43	190	12	2	9	8	38	2 591	57
21 - 25	47	39	30	45	345	21	1	5	2	9	589	13
26 - 30	15	12	2	3	153	9	4	18	2	9	160	3
31 - 40			5	8	41	2	11	46	5	25	19	1
Total	120	100	67	100	1 649	100	23	100	21	100	4 565	100

2.4. 1 620 prepupae were investigated by x-radiography. 31 per cent of them contained only pollen, or pollen, nectar and dead eggs. This result is similar to that of New Zealand leafcutting bees (READ and DONOVAN, 1980).

DISCUSSION AND CONCLUSION

1. The motion of all *Melittobia acasta* insects tends towards day-light. Those leaving the strip of paper sideways took a longer time to make the shorter distance than the ones moving towards light. Owing to its lifespan and mobility, this species is able to travel further than previously supposed.

2. It will be possible in the future using x-radiography, to determine the stage of development of prepupae before storage in a refrigerator for the winter period, in order to choose the right time of cooling.

3. The radiographs taken are of very good quality, even the species of parasites can be determined, e.g. *Coelioxys spp.* can be distinguished from *Megachile* and the propagation of Chalcidoidea can also be followed.

TABL. 3. — *Quality of treated and untreated groups at the beginning and the end of incubation* (GÖDÖLLÖ, 1982)

Condition of	treated 1.1 - 4.1		control 1.2 - 4.2		Total		Without cells containing only pollen	
	Nr	%	Nr	%	Nr	%	Nr	%
Groups 1.-4. at the first x-radiography control on the 5th day of incubation								
Cont. pollen	129	33	79	32	208	33	—	—
Small larvae	38	10	13	5	51	8	51	12
Large larvae	98	27	88	34	186	32	186	43
Pupae	73	23	13	5	86	13	86	21
Parasites	27	7	62	24	89	14	89	24
Total	365	100	255	100	620	100	412	100
Groups 1.-4. at the last x-radiography control on the 16th day of incubation								
Cont. pollen	67	29	25	20	92	26	—	—
Small larvae	9	4	4	3	13	4	13	5
Large larvae	23	10	21	17	44	12	44	17
Pupae	81	36	5	4	86	25	86	33
Parasites	47	21	67	56	114	33	114	45
Total	227	100	122	100	349	100	257	100
Difference in percentage								
Cont. pollen		— 4		— 12		— 7		—
Small larvae		— 6		— 2		— 4		— 7
Large larvae		— 17		— 17		— 20		— 26
Puppae		+ 13		— 1		+ 12		+ 12
Parasites		+ 14		+ 22		+ 19		+ 21

Parasitism could be determined the day before (0) emergence by x-radiography. In the nine groups of the four stocks investigated — without the cocoons containing only pollen — 24 per cent of the prepupae were infested by parasites at the time of the first radiography on the 5th day and 45 per cent of them were infested at the time of the last x-radiography on the 16th day (Table 3).

It was possible to compare the rate of development of the host and its two most important parasites during the emergence period.

3.1. *Melittobia acasta* is a polyvoltine species. Its development is rapid and seems to be independent of the number of emergence days. Some *Melittobia* adults appeared even on the second day of emergence from the leafcutting bee cocoons stored at lower temperatures. These adults can reinfest the same *Megachile* stock. It can be seen on the x-radiographs taken at different stages of emergence (Table 3), that the development of small and large larvae and prepupae is irregular.

This is due to the parasitism caused by a new larval generation. The degree of infestation is dependent upon the applied treatment method.

This second *M. acasta* generation emerges after the emergence of alfalfa leafcutting bees, and its emergence can take up to 50 days. This generation starts from the cocoons left behind at the nesting place and infests the newer, young larvae of the host.

3.2. *Coelioxys rufocaudata* is a univoltine species. According to our records taken from 23 June to 23 July 1982 the *Coelioxys* emerges after the emergence of male leafcutting bees, together with *Megachile* females. Its emergence culminated after the swarming period of the host. Sporadic appearence of some *Coelioxys* females was also observed after 26 July. This can be explained by the fact that *Coelioxys* needs the open leafcutting bee cells instead of new larvae for starting its new generation.

4. The damages caused by the two parasites has rendered the stocks unsuitable for maintenance. New stocks should be imported and new technological management tools must be introduced, in addition to currently available methods, to achieve reduction in infestations.

5. It is advisable to make test incubations every year in order to determine the hygienic state of the stocks propagated at several places. The determination of sex ratio is also of great importance.

6. The quality of the stock can be determined by x-radiography.

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RÉSUMÉ

LUTTE CONTRE LES INSECTES PARASITES DE LA SOUCHE DE MÉGACHILES, *MEGACHILE ROTUNDATA* FABR.

Des millions de prénymphes de *Megachile rotundata* ont été importées des Etats-Unis en Hongrie entre 1972 et 1978, mais l'introduction généralisée de cette abeille en Hongrie, prévue pour 1974 et 1975, n'a pu être réalisée en raison du parasitisme élevé. La situation nous a conduit à rechercher les possibilités d'une propagation artificielle et des méthodes pour lutter contre les parasites. Notre Centre de recherches a reçu pour ses expériences 1 400 prénymphes en 1981 et 1 620 en 1982 du stock importé.

En 1981 nous avons déterminé l'infestation des cocons par *Melittobia acasta* Walker après émergence de *M. rotundata*. En 1982 nous avons dénombré les parasites qui ont éclos durant la période d'incubation de l'hôte. En 1981 et 1982 les prénymphes ont été conservées à Kompolt durant l'hiver pendant un mois à 0-1 °C et traitées avant la période d'incubation au carbaryl à 0,2 %. En 1981 on a installé dans des champs de luzerne (variété Tapiószelei-l) à Dömsöd (Hongrie centrale) 1 400 mégachiles naissantes après traitement au carbaryl et incubation. 1 200 cocons laissés par les hôtes ont été transportés des champs au laboratoire et conservés dans des sacs de polyéthylène à 21 °C et 70 % HR pendant 50 jours. Des ces 1 200 cocons de mégachiles ont éclos un mégachile mâle et 4 913 *Melittobia*. Les *Melittobia* adultes se sont déplacés vers la lumière du jour à la vitesse de 9-14 m/h ; ils sont morts dans les 14 jours suivants par absence de nourriture.

En 1982 on a reçu 4 groupes de prénymphes ayant hiverné. Une partie des cocons fut traitée avec des produits chimiques. On a traité un groupe avec du Sevin à 2 % en lavant les nymphes, au lieu de les laisser quelques jours dans la solution, selon la méthode habituelle. On a examiné 57 % des 1 620 prénymphes incubées : 930 individus ont été radiographiés une fois, 263 deux fois et 66 trois fois. Sur l'ensemble des 1 620 cocons des 4 groupes, 7 106 insectes ont éclos, se répartissant en 151 (9,3 %) mégachiles et 2 dangereux hyménoptères parasites : 92 (5,7 %) *Coelioxys rufocaudata* Smith (Megachilidae) et 6 863 (*Melittobia acasta* Walker (Eulophidae). Selon l'évaluation des données récoltées, le traitement au Sevin a accru non seulement le taux d'émergence des mégachiles, mais aussi celui de *Coelioxys rufocaudata*.

En comparant le taux d'émergence des groupes traités et du groupe témoin on s'aperçoit que 5 fois plus de mégachiles (120) et 3 fois plus de *Coelioxys* (67) ont éclos des cocons, tandis que les 1 649 *Melittobia* représentent 1/3 des individus du groupe témoin. Les populations de mégachiles importées des Etats-Unis ont été conservées à des températures inférieures et traitées avec des insecticides à base de carbaryl (par exemple Sevin) durant l'incubation réalisée à grande échelle. Des pièges lumineux ont également été utilisés pour tuer les parasites mais ils ne pouvaient que capturer des adultes en vol. Malgré ces méthodes de lutte, des parasites ont survécu et infesté la descendance. Le déplacement des cocons à 4 m du site de nidification — selon les recommandations de NÉMETH, 1981 — n'offre pas de protection fiable contre de tels parasites, puisqu'ils sont capables de ramper sur une distance de 100 m, comme l'ont montré nos tests.

Melittobia peut rester dangereux malgré les opérations de lutte menées jusqu'à présent. Son éclosion protégée, sa durée de vie relativement longue et sa grande mobilité peuvent aboutir non seulement à une réinfestation mais causer aussi des dégâts à d'autres hyménoptères pollinisateurs de la luzerne.

Coelioxys était considéré comme un parasite rare en Hongrie (*Fauna Hungariae*, 1958, vol. 12, n° 12), mais il s'est maintenant propagé et étendu à de nouvelles régions parallèlement à l'introduction de la mégachile sur une grande échelle.

Les dégâts causés par les deux parasites ont rendu les souches impropres à leur maintien. Il faudrait en importer de nouvelles et introduire de nouvelles techniques d'élevage, en plus des méthodes déjà disponibles, pour parvenir à réduire les infestations.

ZUSAMMENFASSUNG

BEKÄMPFUNG PARASITISCHER INSEKTEN DER LUZERNE-BLATTSCHNEIDEBIENE, *MEGACHILE ROTUNDATA* FABR.

Zwischen 1972 und 1978 wurden Millionen von Vorpuppen aus den USA nach Ungarn importiert, aber der für 1974 und 1975 geplante allgemeine Import der Luzerne-Blattschneidebiene *Megachile rotundate* Fabricius konnte wegen schwerer Parasitierung nicht durchgeführt werden. Diese Situation veranlaßte uns, die Möglichkeiten einer künstlichen Vermehrung und Methoden einer Kontrolle der Parasiten der Blattschneidebienen zu untersuchen. Unsere Forschungsstation hat im Jahre 1981 etwa 1 400 Puppen und 1982 1 620 Puppen aus dem importierten Bestand erhalten.

1981 bestimmten wir aus leeren Kokons der geschlüpften Blattschneidebienen den Befall mit *Mellittobia acasta* Walker. Im Jahre 1982 zählten wir alle Parasiten, die während der Inkubationszeit ausschlüpften. 1981 und 1982 wurden die Vorpuppen im Winter für einen Monat in Kompolt bei einer Temperatur von 0-1 °C gehalten. Vor der Inkubation wurden sie mit 0,2 % Carbaryl behandelt. 1981 wurden 1 400 schlüpfende Bienen in Luzernefeldern (Varietät Tápiószelei-I) in Dömsöd, Mittelungarn nach Behandlung mit Carbaryl und Inkubation ausgesetzt.

Diese Wirtstiere lieferten 1 200 Kokons, die in das Laboratorium gebracht und in Plastikbeuteln bei einer Temperatur von 21 °C und 70 % r.F. für 50 Tage gehalten wurden. Aus diesen 1 200 Kokons der Luzerne-Blattschneidebiene schlüpften eine männliche Blattschneidebiene und 4 913 Individuen von *Melittobia*. Die *Melittobia*-Tiere bewegten sich mit einer Geschwindigkeit von 9-14 m/h gegen das Tageslicht ; sie gingen ohne Futter innerhalb von 14 Tagen ein.

1982 erhielten wir vier verschiedene Gruppen überwinterter Vorpuppen. Ein Teil der Kokons wurde mit Chemikalien behandelt. Eine Gruppe wurde von uns mit 0,2 % Sevin behandelt, aber nicht wie üblich durch Eintauchen in die Lösung für mehrere Minuten, sondern durch Waschen. 57 % der 1 620 inkubierten Vorpuppen untersuchten wir 1-3 mal mit Röntgenstrahlen. Aus der Gesamtzahl dieser 1 620 Kokons der vier Gruppen schlüpften 7 106 Insekten : 151 (= 9,3 %) waren Blattschneidebeinen, 92 (= 5,7 %) *Coelioxys rufocaudata* Smith (Megachilidae) und 6 863 *Melittobia acasta* Walker (Eulophidae), beides gefährliche parasitische Hymenopteren.

Nach den vorliegenden Daten erhöhte die Behandlung mit Sevin nicht nur die Schlüpfrate der Blattschneidebiene, sondern auch die von *Coelioxys rufocaudata*. Es schlüpften fünf mal mehr *Megachile* (120) und drei mal mehr *Coelioxys* (67), während die Zahl der *Melittobia* mit 1 649 nur ein Drittel der Tiere der Kontrollgruppe betrug.

Aus den USA nach Ungarn importierten Populationen der Luzerne-Blattschneidebiene wurden während der Überwinterung bei niedrigeren Temperaturen gehalten und während der im großen Maßstab durchgeführten Inkubation mit carbarylhältigen Insektiziden (z.B. Sevin) behandelt. Es wurde auch versucht, Parasiten mit Hilfe von Lichtfallen zu töten, aber damit konnten natürlich nur adulte Tiere erreicht werden. Trotz dieser Bekämpfungsmaßnahmen sind Parasiten übriggeblieben und es kam zu einem Befall der Nachkommenschaft. Die Verbringung der Kokons bis in eine Entfernung von 4 m vom Nistplatz, entsprechend den technischen Empfehlungen von NEMETH, 1981, bietet gegen diese Parasiten auch keinen Schutz, da sie nach unseren Ergebnissen bis 100 m weit kriechen können.

Melittobia könnte trotz aller bisher durchgeführten Bekämpfungsmaßnahmen gefährlich bleiben. Der geschützte Ausschlupf, die relativ lange Lebensdauer und die große Beweglichkeit des Parasiten können nicht nur zur Reinfektion, sondern auch zu Schäden bei anderen Hymenopteren unter den Bestäubern der Luzerne führen.

Coelioxys war bisher für Ungarn als relativ seltener Parasit bekannt (Fauna Hungarica, 1958, Vol. XII. Nr. 12), aber gegenwärtig nimmt er zu und breitet er sich in neue Gebiete zusammen mit der im großen betriebenen Einführung von Blattschneidebienen der Lunzerne aus.

Die durch die beiden Parasiten verursachten Schäden haben die Weiterführung der Stämme zwecklos gemacht. Neue Stämme sollten importiert werden, zusammen mit neuen Technologien ihrer Zucht, zusätzlich zu den schon vorhandenen Methoden, um eine Verringerung des Befalls zu erreichen.

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