

REVIEW ARTICLE



Maintenance and application of multiple queen colonies in commercial beekeeping

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Received 10 December 2008, accepted subject to revision 7 July 2009, accepted for publication 5 August 2009.

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Summary

We have previously reported the artificial creation of stable multiple queen honey bee colonies in China. Here we describe, based on our experience and primary research, the techniques necessary to successfully maintain multiple queen colonies and how they can be used commercially. Special care including the provision of sufficient food, avoiding robbing and drifting, destroying newly built queen cells, and abandoning foragers before migration, are necessary for keeping sustainable multiple queen colonies. In practice, these colonies are used as supporting units for the faster build-up of productive colonies and for the production of royal jelly.

Mantenimiento y aplicación de colonias con múltiples reinas en la apicultura comercial

Resumen

Previamente hemos descrito la creación artificial de colonias de abejas con múltiples reinas de forma estable en China. Aquí describimos, basado en nuestra experiencia e investigación primaria, las técnicas necesarias para mantener satisfactoriamente colonias con múltiples reinas y cómo pueden ser usadas comercialmente. Hay que tener especial cuidado con la provisión de suficiente comida, para evitar el robo y la deriva, destruyendo las celdas reales nuevas, y abandonando pecoreadoras antes de la migración, para mantener colonias con reinas múltiples de forma sostenible. En la práctica, estas colonias se utilizan como unidades de apoyo para la acumulación más rápida de colonias productivas y para la producción de jalea real.

Keywords: multiple queen colonies, supporting colonies, brood production, colony build-up, royal jelly

Introduction

That a single mated queen reproduces in a colony is the central dogma in the biology of honey bees. Yet, for decades the economics of commercial beekeeping in the temperate zone have spurred many attempts to artificially produce multiple queen colonies of higher productivity (Kovtun 1949, 1950; Melnik, 1951; Spoja, 1953, Lensky and Darchen, 1963). Such colonies were not, however, widely accepted and used in beekeeping practice because of their

unsatisfactory sustainability. Honey bee colonies composed of several mated, egg-laying queens under freely moving conditions can be produced by removing part of the mandibles of queens to avoid queen rivalry and by using young workers to avoid aggressive behaviour toward the queens (Dietemann *et al.*, 2008; Zheng *et al.*, 2009; Fig. 1). We have also shown that such colonies are stable and can over-winter successfully (Zheng *et al.*, 2009). In this article we focus on the use and maintenance of such colonies for commercial beekeeping.

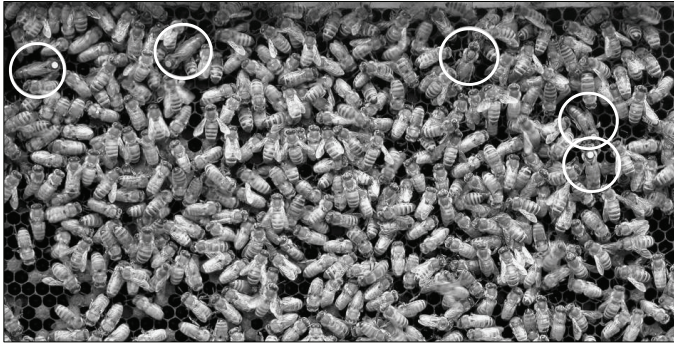


Fig. 1. Five queens on one side of a comb.

The basic research work on creating and managing multiple queen colonies, as well as publicity and promotion of the technique in China have been carried out over the last nine years. Due to their advantages for beekeeping practice, multiple queen colonies produced following this method have been warmly welcomed and are regularly maintained by beekeepers throughout several districts. This is especially the case in Zhejiang province, which is one of the prominent and most productive beekeeping provinces of China. The honey bees used to create multiple queen colonies were a strain of Italian honey bee (*Apis mellifera ligustica*), which was introduced into China in the 20th century. The productivity of this strain has been improved by breeding in the past decades. These colonies are mainly used for the production of honey and royal jelly, and minor products were beeswax and propolis, or some were used to supply drone larvae for human consumption. They are kept in static and migratory apiaries, but very few colonies are used for pollination as this market is not well developed in China.

Research conducted in 2004 revealed that the rates of egg production of 3-queen and 5-queen colonies are respectively 199% and 328% greater than that of a single queen colony (Hu and Zheng unpublished). It demonstrated that keeping multiple queen colonies is a viable way to improve the quantity of brood produced by a colony, despite the fact that the egg laying rate of the multiple queen colonies was less than directly proportional to number of queens per colony (Hu and Zheng, unpublished). This lays a solid foundation for the use of multiple queen colonies in beekeeping practice, since the production and maintenance of strong populations is a precondition for maximizing colony productivity. Close supervision and special care of these multiple queen colonies are, however, necessary since they are sensitive to internal and external factors such as social interactions among individuals and food supply. Here we report techniques to successfully maintain multiple queen colonies and to utilize their advantages in commercial beekeeping practice.

Maintaining multiple queen colonies

Management in the initial stages

To maximize the egg laying of queens and to build up more quickly in the early spring, hives are often requeened in the autumn with young

and vigorous queens. Chinese beekeepers often use the discarded queens to create multiple queen colonies. Queen coexistence is achieved after mandible ablation which prevents them from fighting to the death (Dietemann *et al.*, 2008; Zheng *et al.*, 2009). After being introduced into the host colonies, queens initially lay much less and may even stop laying eggs because of the disturbance resulting from the surgical treatment and the new social context of having multiple reproductives within the same colony. It may take about one week for the egg-laying rate to return to its original level. During this week, queens and workers are still in the process of accepting each other, a stage of fragile balance which requires special attention and support by the beekeeper.

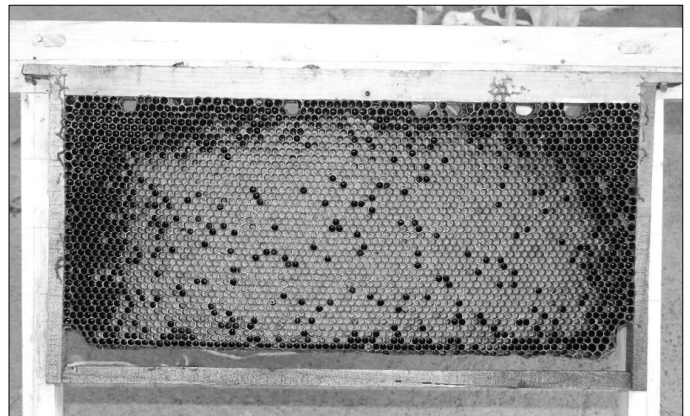


Fig. 2. A brood comb almost entirely filled with larvae by a five queen colony.

Firstly, because the colonies should be composed of young workers and foraging is thus reduced, a sufficient food supply, including high concentration sugar water and pollen, must be provided to the newly established multiple queen colonies. Secondly, newly established multiple queen colonies lack an efficient defence force and are easily robbed by stronger neighbouring colonies. The hives must therefore be placed away from others and the size of the entrance must be reduced to minimize robbing. If robbing occurs, the only solution is to move the hive to a new site. Thirdly, the multiple queen system needs more workers to attend the queens and the brood they produce, and thus a large population of nurse bees is needed. The colony should therefore be augmented with four to six combs covered with bees, and young bees or combs with emerging brood should be added to support the population if necessary, before the new workers emerge.

Daily management

Once the queens recover their egg laying ability, multiple queen colonies build up much more quickly than single queen colonies (Zheng, personal observation), as indicated by the increased number of eggs per day (Hu and Zheng unpublished). The strong egg laying capacity of a multiple queen colony results in most of the combs being occupied by brood (Fig. 2.), so that there is less space for food

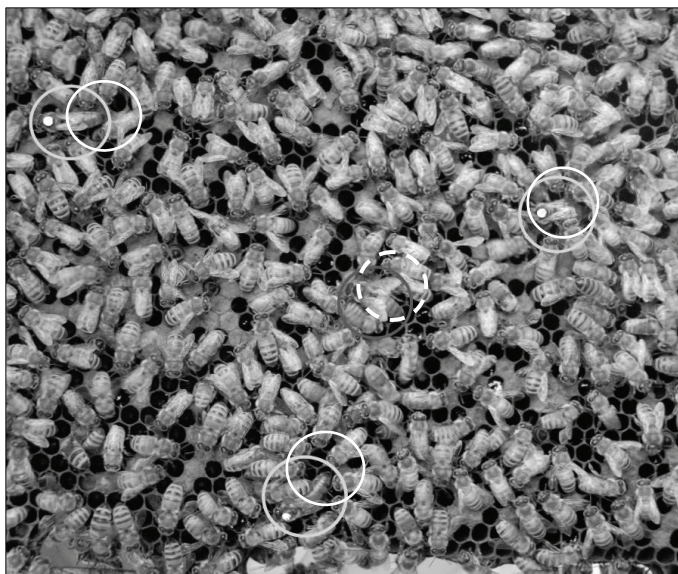


Fig. 3. Young mated daughter queen (in broken circle, unmarked) cohabiting with three old queens. Mating and reproductive status of the young queen was confirmed after being caged on one comb with no other queens.

storage, but more food is actually needed to rear the new brood. Consequently, these colonies must be fed more frequently when there is little nectar flow compared to single queen colonies, especially when no supers have been added.

Even at later stages, robbing, to which *A. m. ligustica* is prone (Darchen and Lensky, 1963), may lead to the collapse of multiple queens colonies and therefore must be avoided. This is especially true when there is a lack of a nectar and pollen flow. In addition, drifting, which commonly occurs in apiaries in which colonies are kept close together, may also cause problems if it occurs on a large scale (Pfeiffer and Crailsheim, 1998). These problems can be avoided by placing the hives away from others, and by monitoring the occurrence of robbing.

Close supervision of the colonies is necessary in routine management. These colonies may build queens cells despite the presence of multiple queens (as noted by Lensky and Darchen, 1963) even shortly after the queens are introduced. Queen cells must be destroyed otherwise one or more queens may be killed after the young queen(s) emerge. In some cases, however, new queens do integrate within the multiple queen organization and cohabit for several days with the old queens (two to five queens in seven cases) before being removed to avoid potential fights. In two cases in which young queens were left in the colonies, they mated and oviposited in the presence of the old queens for several months (Fig. 3). Further research will deepen our understanding of the decision making rules of queens and workers during the supersedure and swarming processes.

Over wintering

Few multiple queen colonies had been successfully over wintered (Kovtun, 1950) before the implementation of our methods. Multiple queen colonies produced by our methods are stable enough to survive the winter in the south of China, where the lowest temperature reaches -5°C and the cold season lasts three months. Over wintering makes these colonies readily available in the spring due to the greater and faster built up of the worker force. This is usually the crucial period for the colony to build up its worker population and to exploit the peak nectar flow. Winter is, however, a high risk period for queen loss, so multiple queen colonies must be well supplied with honey and pollen and have a strong bee population in the autumn. If this is not the case, combs with pollen and honey should be introduced into the colony before over wintering. The amount of food required depends on the forage still available, and on climatic as well as other colony conditions such as the egg laying status of queens.

Queens of colonies in temperate zones might be caged during the winter to reduce brood rearing activity. In multiple queen colonies, however, caging may lead to queen loss with only one queen surviving and should be therefore avoided, but if it is necessary, queens should be placed in close proximity to each other and should be located within the cluster to ensure optimal care.

Migrating

Migration plays an important role in commercial beekeeping nowadays. The stress resulting from the transport may, however, disrupt the artificially established multiple queen social organization and lead to queen elimination. Unrest of old bees in the colonies is usually the cause of such elimination (Lensky and Darchen, 1963). To reduce the possibility of queen losses during or after migration, these old workers must be removed. For this, the hive hosting the multiple queen colony should be moved during the day a short distance away from its original location two days before migration takes place. Young bees or brood combs should be introduced into these colonies to maintain a strong population upon arrival at the new apiary. A hive with one comb should be placed at the original location to collect the old forager bees which are flying back.

Multiple queen colonies and their applications

Multiple queen colonies build up rapidly and maintain a strong and genetically diverse worker population. This genetic diversity might be beneficial for the colony in terms of a better disease resistance (Baer and Schmid-Hempel, 1999; Palmer and Oldroyd, 2003; Hughes and Boomsma, 2004, 2006), a more efficient task allocation (Fuchs and Schade, 1994; Jones *et al.*, 2004) and higher productivity (Fuchs and Schade, 1994). We do not, however, recommend keeping populous multiple queen colonies for the production of hive products. Colonies with several free running queens are more sensitive to external environmental conditions and therefore require an increase in care,

but perform well when kept as supporting colonies. In this situation, the advantages of keeping multiple queen colonies outweigh the disadvantages involved in the additional amount of work needed for their maintenance. In China, according to our research and experience, they can contribute to commercial beekeeping by providing larvae for royal jelly production, capped brood for the rapid build-up of production colonies and workers for package bees.

Colony build-up

It appears that larger colonies tend to produce disproportionately more honey when compared to smaller colonies (Wenning, 2002). One of the best techniques to ensure and enhance the yield is, therefore, supplementing brood development prior to the onset of the main nectar flow to build up the bee population. In China, the first major nectar and pollen flows are provided by oilseed rape, *Brassica napus* L, which blooms early spring, in March and April. The time available for colonies to build up and be ready for the first flow usually lasts from 15 January to 15 March. It can often be a considerably shorter period, as rape may start blooming from mid-February in some areas such as Yunnan province, southwest China. As a result, preparing the colonies for the honey flow after wintering, which usually consists of increasing colony size from one or two combs to about twelve, can be a real challenge. Multiple queen colonies can therefore provide supplemental combs of brood or young workers to support other colonies during that buildup period, thus increasing worker population at an earlier date. Indeed, owing to the multiple queen egg laying system, multiple queen colonies can produce abundant and sufficient eggs during the spring buildup period. Since they are not expected to yield hive products, capped brood comb can

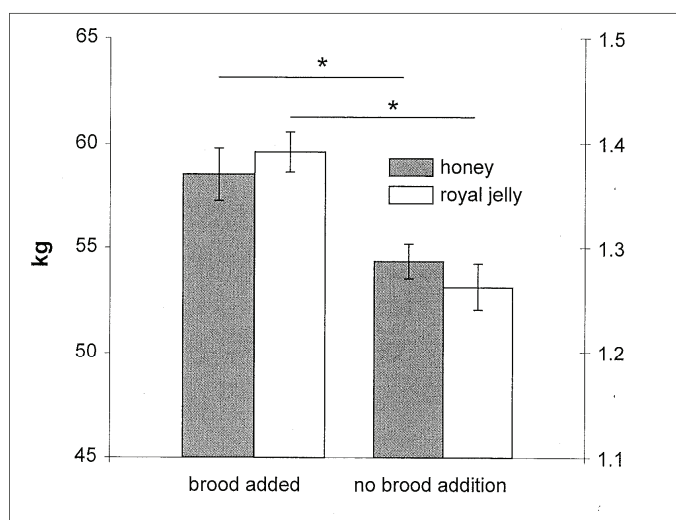


Fig. 4. Comparison of honey and royal jelly yield of the two differentially treated groups. One received a frame full of eggs before oilseed rape flowered in March. The yield of honey and royal jelly obtained from each colony after the flowering season, which lasted for three weeks, was recorded. * indicates statistically significant ($p < 0.02$, paired t-test) differences.



Fig. 5. An apiary keeping multiple queen colonies as supporting colonies for producing hive products in Pinghu, Zhejiang province, China.

be collected from these colonies to help building up others.

Adding brood to colonies can provide them with a long term population boost. In early spring every year between 2004 and 2006, twelve colonies with similar population size were selected and equally divided into two groups. All colonies were managed in the same way, except that one comb filled with eggs from a multiple queen colony was added into each colony of the test group one month before rape blooming in March. No brood was added in the control colonies. The yield of honey and royal jelly obtained from each colony after the blooming season, which lasted three weeks, was recorded. The results over this three year period showed that honey and royal jelly yield increased by an average of 12.2% and 6.5% per colony, respectively, after adding an extra comb of eggs one month prior to the nectar flow (paired t-test on normally distributed data, $t_{\text{honey}}=4.08$, $p_{\text{honey}}=0.001$; $t_{\text{royal jelly}}=2.66$, $p_{\text{royal jelly}}=0.02$; Fig. 4).

This method can also be used to consolidate and maintain populous colonies all year round when needed. The number of colonies that each multiple queen colony (e.g. a five-queen colony) can support varies from ten to thirty (Fig. 5), depending on the state of the colonies to be supported.

Supporting royal jelly producing colonies

Royal jelly is a hive product with a variety of biological functions for honey bees and humans (Krell, 1996; Nagai and Inoue, 2004; Gallmann and Bogdanov, 2008). It is one of the most important hive products for Chinese beekeepers who produce more than 2000 tons every year, accounting for 90% of the world's total harvest. It is a more important hive product than honey in the sense that it can be produced for almost seven months a year, while honey production is limited to nectar flow periods. This restriction affects non migratory apiaries to an even greater extent, and makes royal jelly production an attractive and lucrative activity in China.

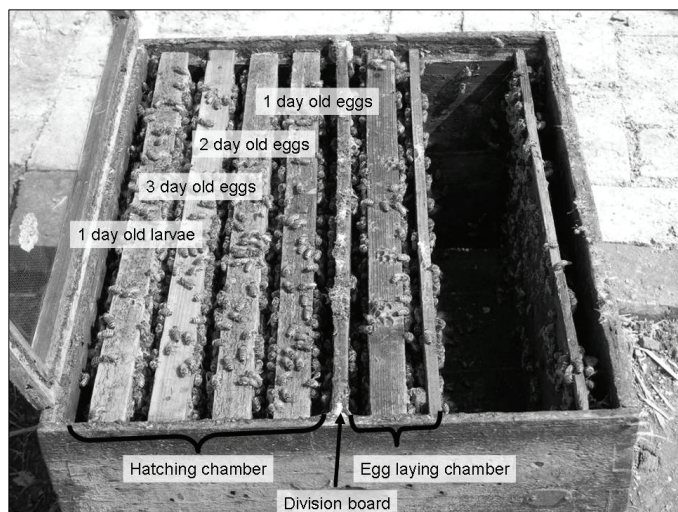


Fig. 6. A hive hosting a multiple queen colony for supplying one day old larvae is divided into two chambers. The egg laying chamber (on the right) contains one comb. The hatching chamber (on the left) consists of one comb with one day old larvae and three combs of one, two and three day old eggs. Every day one comb of newly hatched larvae is taken out for grafting, the comb in the egg laying chamber is transferred to the hatching chamber and an empty comb is inserted into the right hand part for egg laying.

When producing royal jelly, larvae not older than 24 h need to be transferred from worker cells into queen cups. Workers then provide these larvae with the royal jelly necessary for their development. Grafting is a skilful and labour intensive work because a large amount of small larvae need to be transferred. To obtain such a quantity of larvae, a supporting colony is usually set up with four or five combs of bees and a young queen (Chen *et al.*, 2002). Compared to single queen colonies, however, multiple queen colonies have greater egg laying rates (Hu and Zheng unpublished). For example, a five queen colony can supply a comb nearly full of eggs laid when confining the queens to one comb. Upon hatching, three days later, these combs full of larvae can be used for grafting (Fig. 2 shows such a comb containing older, more visible larvae).

A supporting colony for royal jelly production is divided into two parts: an egg laying chamber, where queens are confined to one comb, and a hatching chamber, where eggs are hatched (Fig. 6). Every day the brood comb on which queens have laid eggs for 24 h is transferred to the hatching chamber and a new empty comb is inserted into the egg laying chamber. Three days later, the hatched larvae can be used for grafting in the starter colonies. In this way, one comb full of one day old larvae from a multiple queen colony is available every day for grafting into a royal jelly producing colony. As a result, one multiple queen colony is as efficient as three single queen colonies to meet the needs for grafting. In addition, the combs almost full with larvae of similar age reduce the amount of labour required to find larvae of the appropriate age for grafting on many

combs from several colonies. To avoid depleting the worker force of such colonies, young workers should be added or some combs with larva should remain in the colony to maintain the optimum population and ensure sufficient brood care. For an apiary with 100 colonies used for the production of royal jelly, one third of them need young larvae every day, and two five queen colonies would be sufficient to supply the amount of larvae necessary.

Production of package bees

Multiple queen colonies can be economically beneficial and labour saving in providing young workers for producing package bees. This would be especially useful in countries in which the demand of package bees exceeds the supply due to colony losses such as those experienced with "Colony Collapse Disorder" due to the increasing demand for honey bees for pollination. Healthy and strong colonies are crucial to supply large numbers of workers for package bees. Multiple queen colonies are also ideal to serve as queen banks and therefore as a backup source of replacement queens in case of queen loss in other colonies (Spoja, 1953). According to Woyke (1988) queens in queen banks are often injured by worker bees in the colony. The proportion of queens injured in this way in multiple queen colonies is unknown.

Conclusions

Multiple queen colonies are sustainable enough for long term keeping, provided they are given close supervision and special care. Firstly sufficient food must be ensured, especially in places and during periods where nectar and pollen flow is lacking. Secondly, robbing and drifting must be avoided. Thirdly, colonies should be monitored for the presence of queen cells, and newly produced queen cells must be destroyed. Fourthly, queens should not be caged over the winter and lastly, before migration, foragers must be abandoned and young workers must be supplied to compensate for this loss of the work force.

Given proper management, the advantages of multiple queen colonies outweigh the amount of work needed for their maintenance. We recommend keeping multiple queen colonies as supporting colonies, contributing indirectly to apiary productivity. In China, they are used to build up populous colonies faster in the spring prior to major nectar flows, and to maintain the population all year round whenever needed. They are also helpful as supporting colonies to provide the larvae necessary for grafting for royal jelly production and as sources of replacement queens. Finally, they can contribute to package bee production by providing large numbers of workers.

Acknowledgements

We are grateful to Quan-Qing Jiang, Lin-Jie Fu from Pinghu, Zhejiang for the access to their colonies. This work was funded by the National Natural Science Foundation of China. HQZ was funded by the China Scholarship Council (CSC). CWWP was funded by a Claude Leon Fellowship.

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