Advanced management of bee health and beekeeping under Taiwan subtropical/ tropical climate

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Abstract

Taiwan with her dense population but because of the rich and diversified sub-tropical plant resources is suitable for the development of apiculture. The European bee (*Apis mellifera*) is the major housing bees, while Asian bee (*A. cerana*), the native local bee, is plentiful on the island as a wild bee. In addition to honey, royal jelly production is another famous bee product for the beekeeping industry in Taiwan. Being mild temperature in winter season, queen honey bees lay eggs all the year round in Taiwan -- that's a unique advantage and quite different from the temperate apiaries. This paper presents some management techniques focusing on controlling varroa mites and American foulbrood disease to give reference for the beekeeping industry with similar climates or condition in other part of the region. In addition, our report confirmed that the *Nosema ceranae* can severely infected European bee colonies has since drawn widely attentions of other part of world researchers. This paper also presents our recent studies on the new pests.

Keywords: Taiwan, honey bee, varroa mite, American foulbrood, *Nosema ceranae*.

Introduction

Taiwan is located in the subtropics/tropics and is rich in biodiversity. The warm weather and ever green scenery of the island endows with rich in nectar plant resources and thus suitable for the development of apiculture. As we have known, honey bees can produce nutritional honey, pollen, royal Jelly and propolis and other bee products, and also plays an important role of pollination on the crop and wild plants. As shown in Table 1, a total of 663 beekeepers keep around 70,000 bee colonies in Taiwan. The island is famous in its production of good quality longan honey and the royal Jelly which ranks second in the world and is an important feature of Taiwan beekeeping industry. Taiwan beekeeping has been 200 years of history, although the Asian bee (*A. cerana*) is a native species and is abundant in wild (An et al., 2004).

Being located in the subtropical/tropical region, there isn't the wintering seasons in Taiwan. The queen honey bees laying eggs all the year round, that's quite different from the temperate apiaries. In addition, being a long term of human domestication, it is important to establish an advanced management of bee health under Taiwan subtropical/ tropical climate. This paper presents some management strategies for controlling varroa mites and American foulbrood, two of the most important bee pests in Taiwan. It also presents some study results about the bee nosema disease in Taiwan.

Table 1. Annual reports* of Taiwan beekeeping industry from 2000 to 2009

Year	Beekeepers	Bee hives	Honey yield (tons)	Royal jelly (kg)
2000	709	94,970	5,839	299,002
2001	753	98,410	2,759	314,912
2002	749	99,630	3,230	328,779
2003	747	100,720	5,935	332,376
2004	702	101,530	4,987	345,202
2005	715	88,930	6,327	320,148
2006	669	80,750	4,978	319,760
2007	631	73,050	3,220	257,963
2008	663	71,200	7,219	215,400
2009	728	77,150	5,367	209,000

*Data from Council of Agriculture, Executive Yuan, Taiwan R.O.C. (http://www.coa.gov.tw/view.php?catid=21694).

Control Strategy of *Varroa destructor*

The control of *Varroa destructor* previously known as *Varroa jacobsoni* has been a big problem for worldwide (Anderson and Trueman, 2000). It is an ectoparasitic mite that feeds on the haemolymph of adult and immature honey bees (*A. mellifera*). Once honey bee colonies were infected by varroa mites and unattended by beekeepers for negligence of given proper control, the bee colonies gradually become weakened and destroyed within few years. Therefore, many beekeepers use Synthetic acaricides such as Apistan® (fluvalinate) and CheckMite+ (coumaphos) have been used by beekeepers and are highly effective to control *V. destructor*. The lone-term use of the synthetic acaricides by beekeepers may end up accumulation of these acaricides residues in wax and honey. Also, many reports showed that *V. destructor* has developed resistance to acaricides in mite population, such as reported by France (Colin et al., 1997), United States (Elzen et al., 1999), and Mediterranean (Floris et al., 2001).

Varroa mite has been found in Taiwan since 1970, it remains as the worst pest nowadays. Fluvalinate is the only registered chemical for the varroa control since 1995. Since it has been used for 15 years, varroa population had showed their tolerance to this chemical in Taiwan (Chen et al., 2002b). Therefore, we have to search new tools from the non-toxic and natural substances such as the organic acids (Chen and Chen, 2008) and essentials (Chen et al., 2009) for use against of *V. destructor*. In Europe, oxalic acid has been known to be effective control *V. destructor* since end of the 20th century (Imdorf et al., 1997). Oxalic is a natural substance of honey. It is allowed for use in the organic apiculture to control *V. destructor* (EU Council Regulation, No. 1804/1999). Numerous studies have been undertaking in using oxalic acid syrup to control *V. destructor* at the broodless period in honey bee colonies. The efficacy of control is up to 99.4% when the brood was not present, but when brood was present the efficacy of 39.2% was found after three OA treatments (Gregorc and Planinc, 2001). In Taiwan, it is brood-right all year round in the most honey bee colonies. Therefore, the application method of oxalic acid needs to be modified when using in Taiwan and other tropical regions.

Chen and Chen (2008) reported the multiple 3% oxalic syrups administration is needed to receive the well control effects. The field trials were conducted at fall season in Taiwan, administrated syrups containing 3% oxalic acid and 30% sucrose (w/w), twice a week and 5 successive applications totally, and the spraying dosage was 4 ml per comb frame. That was each treated bee colony receiving 40-50 ml oxalic syrup at each application; the mite mortality could reach to 82.4 ± 3.8% (mean ± s.d.) (table 2). In addition, no more brood death was found during the testing period (table 3). This reveals, by the repeated spraying with oxalic syrups, the varroa mites could be controlled effectively even when brood was present at the treated bee colonies. Excess oxalate concentration on honey bees may be a toxic; Ebert et al. (2007) feed adult worker bees with 1000 ppm oxalic syrup and 96% of them died after 8 days feeding. The 50% lethal time (LT_{50}) was only 4.8 days. Obviously, it is not suitable for feeding oxalic syrup into the honey bee colony; we also must avoid using oxalic acid sprayed into the cells of 1-3 day old larvae.

Table 2. Efficacy of honeybee colonies receiving five successive sprayings with 3% oxalic-sugar syrup (OA) at different doses (Chen and Chen, 2008)

Doses	Varroa drop-down (%)			
	Oct 15 (OA 1 + OA 2)	Oct 22 (OA 3 + OA 4)	Oct 29 (OA 5)	Cumulative mortality
0 mL / frame	1.4 ± 1.3a	4.7 ± 1.9a	8.1 ± 3.2a	14.1 ± 5.0a
2 mL / frame	25.6 ± 5.9b	27.5 ± 7.5b	19.5 ± 5.2b	72.6 ± 11.3b
4 mL / frame	22.9 ± 1.1b	39.2 ± 5.2c	20.3 ± 6.4b	82.4 ± 3.8c

*Means in the same column followed by a different letter are significantly different by Duncan's multiple range test ($p < 0.05$).

Table 3. Percentage of 1-day-old worker larvae developing into capped stage before and after spraying with 3% oxalic-sugar syrup (Chen and Chen, 2008)

Doses	Pre-treatment	3% Oxalic-sugar syrup			Fluvalinate + Coumaphos
	Oct 8*	Oct 15	Oct 22	Oct 29	Nov 5
0 mL / frame	90.3 ± 5.7 a**	87.8 ± 5.9 a	90.3 ± 9.2 a	87.3 ± 5.3 a	86.0 ± 3.2 a
2 mL / frame	85.7 ± 4.2 a	89.3 ± 8.7 a	84.5 ± 5.1 a	87.0 ± 6.0 a	87.5 ± 6.3 a
4 mL / frame	88.3 ± 8.0 a	91.8 ± 9.1 a	85.8 ± 6.0 a	88.8 ± 6.1 a	94.0 ± 4.3 a

Date of 1-day-old larvae labeling.

**Means in the same row followed by a different letter are significantly different by Duncan's multiple range test ($p < 0.05$).

Thymol is a natural ingredient isolated from thyme (*Thymus vulgaris*) plants. It has also been found to present a high control efficacy on varroa mites. At present, Europe has developed a variety of thymol commodities, for example: Apiguard® contains 25% thymol, 87% control efficacy was found after an administrated period of 5 weeks (Palmeri et al., 2007); Api Life VAR ® contains 74% thymol, 16% eucalyptus oil, 3.7% canphor and 3.7% menthol, the control efficacy was more than 74% after a 4 weeks of continuous use (Floris et al., 2004). Taiwan has not yet to import such a commercial thymol product, Chen et al. (2009) evaluated the efficacy of the pure thymol powder in Taiwan apiaries. Thymol is a slightly volatile material and the fumigated gradient depends on some factors such as temperature and exposing surface area. The application method needs some modifications to achieve a better control. Experimental results show that in summer, putting one dose of 20 g thymol powder upon the comb frames for 3 weeks received 93.2 ± 2.3% control effect (table 4); if the same dose changed to 10 g per week and used twice, the control rate was 89.1 ± 6.2% (table 5). But to Taiwan for use in winter time, one dose of 20 g thymol just received only 13.7 ± 4.6% control efficacy.

Using thymol as the alternative varroa mite control material, lethal effect could persist for 15 days or more except winter time, it is very convenient and you do not need to always open the hive boxes to administrate the infected bee colonies again and again. In winter, we suggest using the oxalic syrups for varroa control. It is worth noting, thymol with special smell, so it is recommended to stop the bee products during the pesticide application, avoid affecting honey products original flavor.

Table 4. Efficacy of honeybee colonies were administered with different doses of thymol in summer trial (Chen et al., 2009)

Dose	Varroa drop-down (%)			
	Week 1	Week 2	Week 3	Cumulative mortality
20 g	38.5 ± 9.3	45.8 ± 14.2	9.0 ± 4.9	93.2 ± 2.3 a*
10 g	40.0 ± 15.2	21.7 ± 11.2	9.4 ± 5.3	71.2 ± 14.2b
0 g	3.7 ± 1.9	4.1 ± 3.3	3.7 ± 2.1	11.5 ± 6.8c

*Means in the same column followed by a different letter are significantly different by Duncan's multiple range test ($p < 0.05$).

Table 5. Efficacy of honeybee colonies receiving two successive with 10 g thymol in summer trial*(Chen et al., 2009)

Treatment	Varroa drop-down (%)		
	Week 1	Week 2	Cumulative mortality
Thymol 10 g + 10 g	44.7 ± 23.5	44.4 ± 26.0	89.1 ± 6.2

*Week 1, 1st treatment; week 2, 2nd treatment with 10 g thymol

Control strategy of American Foulbrood

American foulbrood (AFB) is a severe bacterial disease affecting larvae of the honey bee *A. mellifera*. It is caused by spores of the *Paenibacillus larvae*. The infected colonies may die and pathogen spores will quickly spread to the other bee colonies, resulting in a great loss. The disease is present worldwide and cases have been reported in almost all beekeeping regions, the AFB were no exception in Taiwan. In order to monitor the epidemic of AFB in Taiwan, during 2005 and 2006, Chen et al. (2008) carried out a nationwide study to assess the presence and the amount of *P. larvae* spores in honey samples from Taiwan. A total of 838 honey samples collected from apiaries in Taiwan, including 173 samples were collected in local market but originally imported from Thailand. The result (table 6) showed that 208 samples were contaminated with *P. larvae* spores (24.8%). Since a very few samples contaminated with a higher spore count, there may be the bee AFB outbreak in this apiary; most of the honey samples are not detected or detected only a small number of spores, AFB in Taiwan's epidemic has eased.

Table 6. Culture results of American foulbrood spores in honey samples from Taiwan, 2005-2006 (Chen et al., 2008)

Honey sources	2005		2006		2005 + 2006	
	n	Positive	*n*	Positive	*n*	Positive
Taiwan, single hive	141	23 (16.3%)	103	13 (12.6%)	244	36 (14.8%)
Taiwan, apiary	147	31 (21.1%)	274	121 (44.2%)	421	152 (36.1%)*
Thailand, imported	143	14 (9.8%)	30	6 (20.0%)	173	20 (11.6%)
Total	431	68 (15.8%)	407	140 (34.4%)*	838	208 (24.8%)

*Significantly different by *Z-test* ($P < 0.05$)

Currently there is no approved drug for AFB control in Taiwan; the use of antibiotics is illegal and strictly prohibited. It means beekeepers must underlie all successful beekeeping management techniques designed to reduce and eliminate AFB. The strategies as follows:

1. Replacing Old Combs

Combs must be used no more than 2 years, to avoid the accumulation of the pathogens inside the hive.

2. Cleaning Hive Equipments

Clean and disinfect the hive boxes, gloves and tools once in each year at least.

3. Hygienic Apiary

Avoid using a dubious bee pollen cake to feed bee colonies; it may import pathogens into apiaries. Once finding the infected colonies must be immediately removed from its origin apiary, and some sterilizing agents such as 3% sodium hypochlorite is needed to disinfect the hive place.

4. Regular AFB Monitoring

It is difficult to find the AFB infection by the visual inspection; the disease usually has spread out once discovering a typical AFB sign in an apiary. For the early detection of infected colonies, Taiwan is now inspecting AFB spores in extracted honey from a particular apiary to achieve the goal of early detection of AFB. According to the 2004-2006 survey, the spore positive samples was about 15-33%, and these apiaries didn't present AFB outbreak, they were still at the early infected stage. After extracting their honey, bee colonies were no longer detected AFB spores (Chen et al., 2002). This contaminated honey should not feed the bee colonies, to avoid the spread of the disease.

6. Burning AFB Colonies

This is the most effective method when the typical AFB signs have appeared at bee colonies. Before burning the AFB hive, a hole should be dug. The hole will contain the fire, and will also ensure that any infected material not completely destroyed by burning will be buried so that foraging bees will not find it.

Researches on Nosema Disease

In the past years, the nosema disease of honeybee is not considered a serious problem of beekeeping in Taiwan, maybe because Taiwan extends from tropical into subtropical regions and the pathogen is heat sensitive. However, this disease spread widely throughout Taiwan after 1972 and was consistently detected each spring and autumn (An and Ho, 1980). The pathogen of microsporidiosis in Taiwanese apiaries was assumed to be *N. apis* based on optic microscopic observations. However, in 2005, Huang et al. (2005, 2007, 2008) reported the *A. mellifera* bee colonies in Taiwan infected *N. ceranae*, the other nosema pathogen found in *A. cerana* bee colonies of Beijing, China (Fries et al., 1996). This is a first documented finding that European bee colonies infected *N. ceranae* in the world. In past, China apiculture research scholars believed that *N. apis* infected *A. mellifera* bees and *N. ceranae* spores were found only in *A. cerana* bee colonies. This because of their similar shapes under a microscope examination, it is difficult to distinguish. After this, European researchers have found that *N. ceranae* also presented in *A. mellifera* colonies of the European regions, and in Spain it occupied a very high proportion of bee infection (Higes et al., 2006), the researchers speculated that *A. ceranae* may be the main cause of bee death in recent Spanish colonies. The pathogens of *N. ceranae* have now spread to the rest of the world (Klee et al, 2007; Huang et al, 2007). In some beekeeping area of the world, *N. ceranae* had been found to replace *N. apis* as a main caused agent of the nosema disease of *A. mellifera*.

Our recent research is to survey and confirm that *N. ceranae* is a major nosema pathogen in Taiwan honey bee population; to monitor the nosema spores continuously in each month and the variance in seasons. We also evaluated the influence of the temperature factor on the nosema epidemic and to select the drugs for preventing the nosema infection. The results showed that all seven monitored apiaries were parasitized by *N. ceranae*. Nosema went to the highest peak in winter and down to the lowest peak in summer. The average of nosema spores reached to $432.1 \pm 245.9\times10^4$ spores / bee in December and $32.8 \pm 28.0\times10^4$ spore / bee in September. It was found that the highest spore counts were I-lan and Taipei among the seven apiaries. Hsinchu and Hualien were the lowest spore density. The honey bee larvae were artificially infected with spores and incubated at 30□ and 34□ respectively, the results showed that the spores of infected larvae at 30□ were higher than that at 34□. For preventing nosema disease, fumagillin was found to effectively suppress nosema spores and the amount of spores decreased significantly after the drug administrating. Neither the probiotic bacteria nor propolis extract supplements could suppress the nosema infection.

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