

**CHALLENGES ASSOCIATED WITH THE HONEY BEE (*APIS MELLIFERA ADANSONII*) COLONIES ESTABLISHMENT IN SOUTH WESTERN NIGERIA**

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## ABSTRACT

The southwestern part of Nigeria is a tropical rainforest region having many local beekeepers. These beekeepers have experienced decline in colony establishment in the recent past. A study carried out in Lagos, Ogun and Osun states between December 2009 and September 2011 examined 14 randomly selected commercial bee farms for problems associated with decline in colony establishment. Sampling and treatments were split equally between each apiary and three colonies were selected in each. All the colonies were housed in Tanzania/local top bar hives. There were 58.34, 44.84 and 40.61 average percentage declines in colony establishment in Lagos, Ogun and Osun States, respectively. Presence of pests and diseases, pesticide poisoning, poor hive and seasonal management, ecological problem and lack of queen rearing were potential problems identified by the beekeepers. All the apiaries had pests like Crickets, Ants (*Companotus pennsylvanicus*), Small Hive beetle (*Aethina tumida*), Termites (*Macrotermes spp*) and Spider (*Lactrodectus mactan*). *Varroa* mite infestations were found in 33 (78.57%) of apiaries sampled. There was no significant difference between the levels of *Varroa* infestation in all the colonies during the dry and wet seasons at confidence interval of 95 percent ( $t = 1.542$ ,  $df = 13$ ,  $p = 0.147$  ( $p > 0.05$ )). *Nosema* spores were found in 27 (64.29%) colonies examined. The number of spores range from  $16 \times 10^3$  to  $30.4 \times 10^3$ . There were no significant differences in the infection from colony to colony, apiary to apiary and between dry and wet seasons ( $t = -0.094$   $df = 11$ ,  $P = 0.927$  ( $P > 0.05$ )). Diseases like American and European foulbrood were absent while chalkbrood disease was prevalent. Environmental factors of high temperature, high rainfall and high relative and hive humidity enhanced the spread of pests and disease pathogens. Many insecticides were constantly applied by the farmers on the crops in the surrounding farmland. Two of the commonly used insecticides DDVP (Dichlorvos) and Cyperforce (Cypermethrin) were discovered to have increased mortality on worker honey bees with progressively larger doses. It was obvious that insecticide toxicity had a significant effect on the colony. Hence, ecological and management problems had contributed to the decline in colony establishment in this area.

**Key words:** colony, decline, varroasis, infestation, toxicity

## INTRODUCTION

In recent times, many countries have experienced increase in reported cases of colony losses from Europe, USA, South America, Australia, Middle East and Japan [1]. Africa is not excluded; Oyerinde and Ande [2] reported the impacts of bee pests on colony establishment in Kwara State in Nigeria which resulted in 15% decline in honey bee colony establishment in some of the Local Government areas.

In Nigeria, beekeeping has been part of the normal traditional agricultural enterprise of the Oyos in Okeogun and Tivs in Benue but honey production has always been on the decline and never satisfies local demand [3]. The behaviour of these bees such as regular absconding and aggressiveness has contributed to low colony establishment. Many people, according to Fletcher [4] found it difficult to work with the bees, study them or rear queens to improve them because they attack readily.

For unexplained reasons, honey bee colonies are dying in record numbers throughout the world [5]. United States experienced its second worst recorded loss in the winter of 2009/2010 when a total of 33.8% of all managed hives perished between October and April. Several opinions show that modern beekeeping methods could weaken bee colonies to the point where they succumb to diseases that would otherwise have only minor impacts on colony health [5, 6, 7]. Similarly, inability to find solutions to problems that plague the beekeeping industry, particularly those associated with the decline in agricultural areas [8], habitat loss and decline in plant diversity [9] have also been associated with the plummeting populations of both feral and managed colonies. Some scientists believe it is a result of the synergistic effects of pesticides, while others believe it is caused by sublethal doses of the agricultural insecticides such as imidacloprid or clothianidan, that can cause bees to become disoriented and lose their way home [10]. Many attribute honey bees loss of vitality and disease resistance to the accumulation of chemicals in the hive [11, 12].

In Nigeria, there is no detailed study of the effects of parasites, diseases and pesticides on beneficial arthropods like honey bees. Current literature on insecticide toxicity does not provide information on vulnerability and resistance shown by the local honey bees and the consequences on the beekeeping activities in Nigeria. The honey bee is infected by a wide range of parasites and disease pathogens. The introduction and spread of parasitic arthropods of honey bees, *Acarapis woodi*, *Varroa destructor* and *Aethina nutida* have been tagged the major cause of decline in the population of both feral and managed honey bee colonies [13, 14, 15, 16].

According to May [17], of the natural enemies that enter the bee nest itself, the wax moth *Galleria mellonella* L. and *Achroia grisella* F. are considered serious beekeeping pests. Such pests may not affect a strong colony but a weak one that cannot protect its comb may be seriously affected leading to colony absconding. Also, ants are common terrestrial pests of honey bees [18]. Dawid [19] who opined that few *Apis* diseases are of importance in Africa listed *Nosema* and European foul brood

disease while American foul brood and diseases caused by mites were unknown. This researcher was of the opinion that the absconding habit and rapid destruction of abandoned combs by wax moth probably assists materially in the natural control of diseases.

Diseases, pests and parasites are biotic factors while environmental factors and pesticide poisoning are abiotic factors that pose a great threat to beekeeping. Hence, this study investigated the ecological and management problems associated with honey bee colony establishment in southwest Nigeria.

## MATERIALS AND METHODS

### Study site

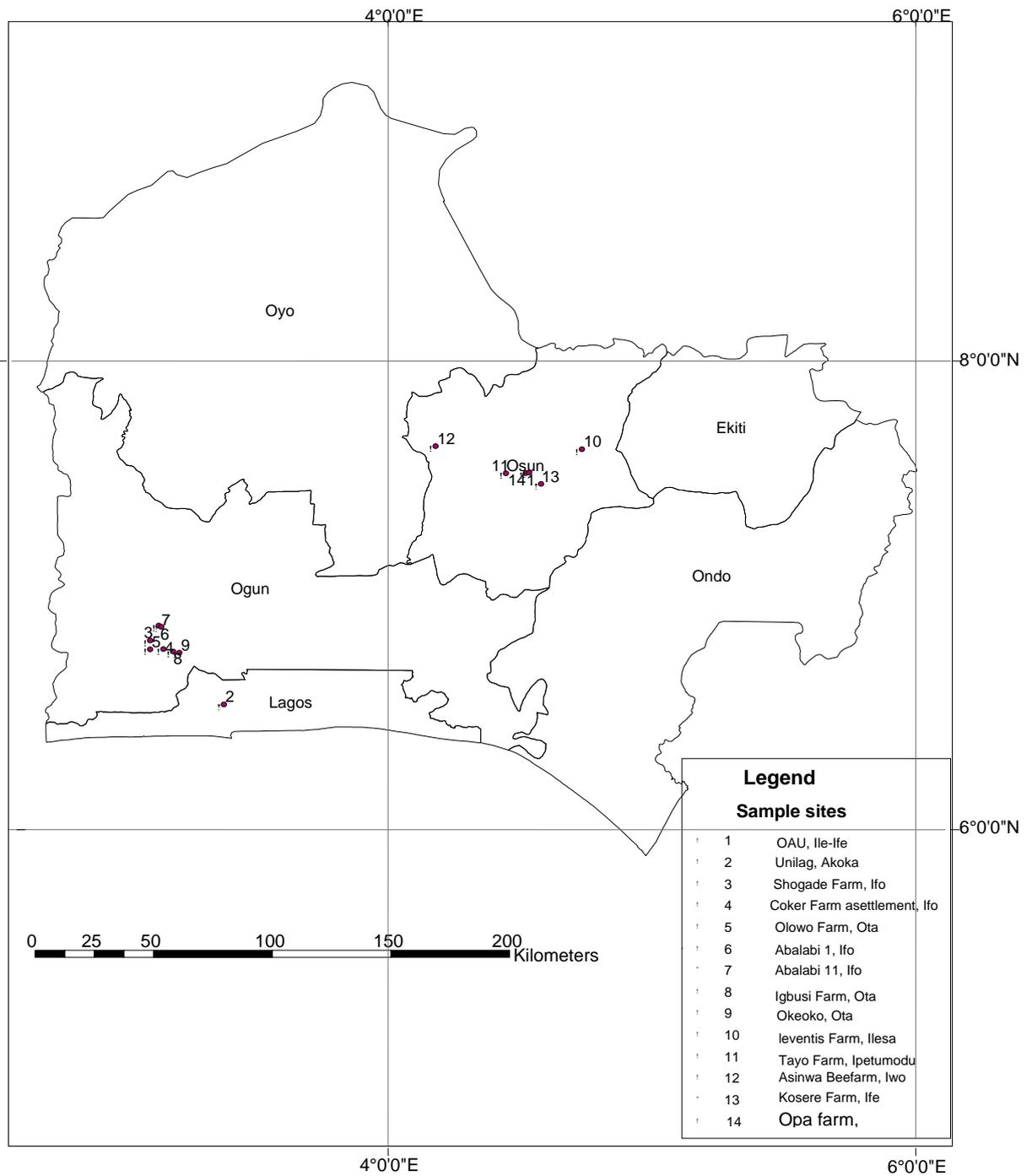
One control and thirteen experimental apiaries were used for these studies conducted between December 2009 and September 2011. The control apiary was located in the Teaching and Research Farm of Obafemi Awolowo University, Ile – Ife and experimental apiaries were spread in three states, Lagos, Ogun and Osun. All the experimental apiaries were located within 20 to 200km radius of the control site (Figure 1). Sampling and treatments were split equally between each apiary and three colonies selected in each. All the colonies were housed in Tanzania top bar hives.

### Methodology

Questionnaires were administered and survey pathology and toxicological tests carried out to confirm the beekeeper's reports. The questionnaire administered sought comprehensive information on:

- i. Number, age and types of hives and management practices adopted by the beekeepers such as seasonal management, pest and disease management, application of medicated sugar syrup and food supplements to the hives.
- ii. Hive conditions such as colony strength, brood rearing, hive storage, comb building and nest hygiene.

The colonies were monitored for the entire study period for their health and survival in connection with all factors that potentially affect bee health, insecticide toxicity and quality of honey. Colonies were sampled for pests and parasites and diseases according to OIE methods [20] and disease diagnosis was carried out according to Shimanuki and Knox [21] on the colonies and broods to verify the levels of infestation with nose mosis, varroasis, acariosis, chalkbrood and foulbrood diseases. Information was sourced on the pesticides frequently applied in the surrounding farmland and toxicity tests were carried out according to Robertson *et al.* [22]. Lethal and sublethal doses of the formulated grade Dichlorvos (DDVP 1000EC) and Cyperforce (Cypermethrin 10% EC) were administered to worker bees by contact (topically) to identify if the products have low non-target activity. Analysis of variance (ANOVA), probit analysis and independent-samples *T*-tests were used to analyze the data collected.



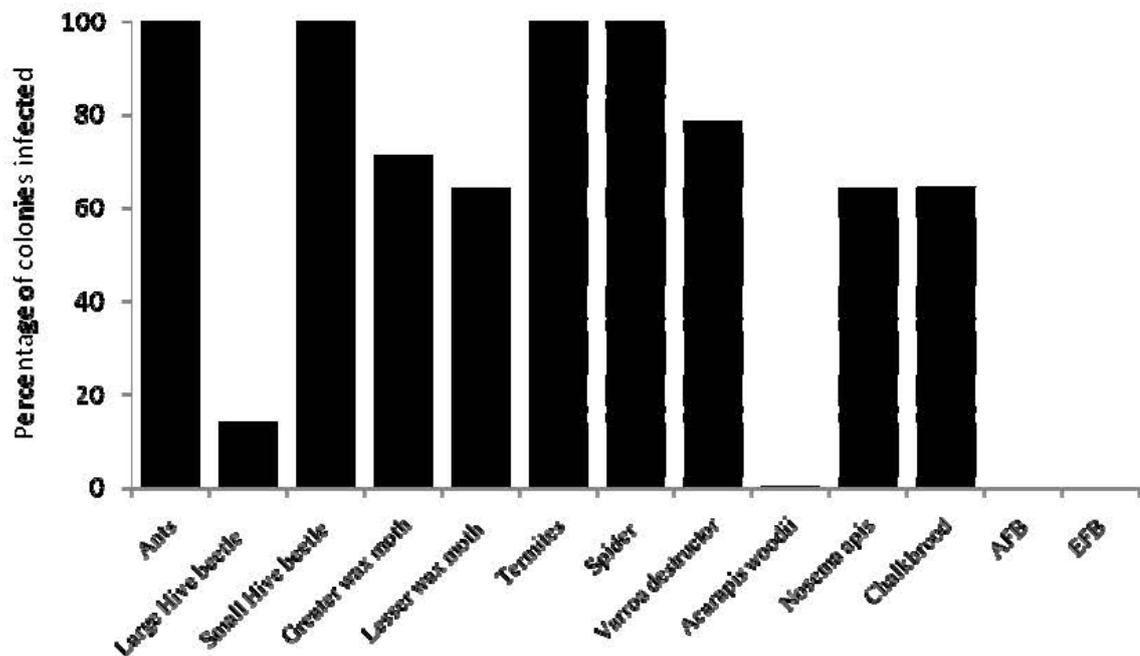
**Figure 1: Distribution of Apiaries in Southwestern Nigeria where colonies were sampled from December 2009 to December 2011 for decline, pathology and toxicity tests**

## RESULT

All the apiaries in southwestern Nigeria have experienced a decline in the number of their honey bee colonies. There were annual declines of 3.43%, 5.46% and 4.93%, in colony establishment in Lagos, Ogun and Osun States, respectively (Table 1). The declines represent the number of colonized hives lost either through the bees absconding, die-off of colonies or colony collapse annually and from the start of the apiary till the time of the study. Most of the beekeepers do not manage their colonies properly because of the widely held opinion that local top bar hives do not require constant inspection and also due to the aggressiveness of the bees. However, observed presence of pests and diseases, poor hive and seasonal management, ecological problems and lack of queen rearing were potential problems raised by the beekeepers.

The apiarists also admitted intentional use of some agrochemicals on the crops in the surrounding farmland. The insecticides and percentages of beekeepers that reported their applications were endosulfan (21.4%), methyl parathion (21.4%), carbofuran (28.6%), deltamethrin (14.3%), stampade (7.1%), malathion (35.2%), dimethoate (28.6%) and DDVP, Darksh (Dichlorvos) (50%) and Cyperforce (Cypermethrin)

A total of 42 (100%) colonies examined were infested with Crickets, Ants (*Companotus pennsylvanicus*), Small Hive beetle (*Aethina tumida*), Termites (*Macrotermes spp*) and Spider (*Lactrodectus mactan*) (Figure 2). The weeds around the hives and dropped leaves covering the hive top provide the conducive environment for the pests. Again, 6 (14.29%) colonies were infested with Large hive beetle (*Hoplostomus fuligenius*), 30 (71.43%) colonies with Greater wax moth (*Galleria mellonella*) and 27 (64.29%) colonies with Lesser wax moth (*Achroia grisella*) (Figure 2). The presence of all these pests remained throughout the period of study.



**Figure 2: Pests, parasites, diseases and percentage of colonies infested**

*Varroa* mite infestations were found in 11 (78.57%) out of 14 apiaries sampled (Table 2 and 3). There was no significant difference between the levels of infestation of the colonies during the dry and wet seasons at confidence interval of 95% ( $t = 1.542$ ,  $df = 13$ ,  $p = 0.147$  ( $p > 0.05$ )) (Table 4). Similarly, there was no significant difference at 95% confidence interval between the average mite load per adult bee during dry and wet season in all the apiaries grouped together in Osun, Ogun and Lagos states ( $t = -1.115$ ,  $df = 2$ ,  $p = 0.381$  ( $p > 0.05$ )). The mite infestation cannot be established whether due to poor hive management or climatic factors or related to the geographical location of the apiaries. The mite specimen sent to CSIRO Laboratory Canberra, Australia for confirmation and banking was identified as *Varroa destructor* and haplotype confirmation revealed that they were the Korean haplotype “K” type commonly referred to as the virulent type.

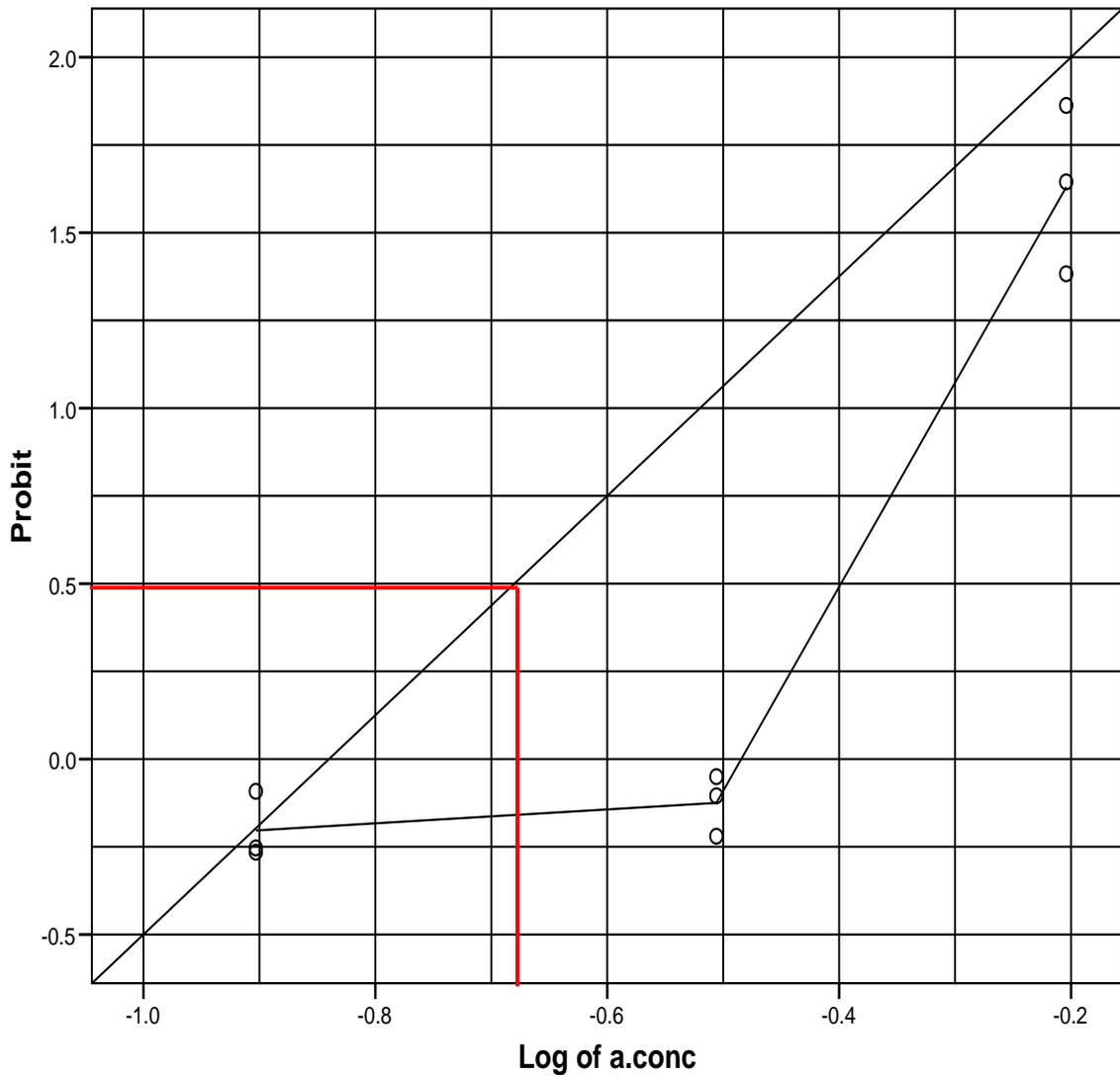
The bee guts dissected and examined, confirmed the absence of tracheal mite (*Acarapis woodi*). The tracheal tubes of the bees appeared healthy, with outer surfaces pale, white, smooth and clear of debris. This observation was contrary to the pathogenesis in infected bees which usually have the trachea appearing reddish brown, full of mites and very brittle. In the case of Nosemosis, *Nosema* spores were found in seven apiaries with a total of 27 (64.29%) colonies tested (Table 5). In the dry and wet seasons, the infected colonies remained the same. The numbers of *Nosema* spores in the infected colonies during the dry season range from  $16 \times 10^3$  to  $30.4 \times 10^3$  and in the wet season range from  $16.3 \times 10^3$  to  $28.9 \times 10^3$  (Table 5). On the average, the mean number of spores in infected colonies is  $16861.54 \pm 8026.88$  (Mean

+ S.D) during the dry season and  $16846.23 \pm 7881.72$  (Mean + S.D) during the wet season (Table 5). There were no significant differences in the infection from colony to colony, apiary to apiary, and across the forestry or geographical areas and between dry and wet seasons ( $t = -0.094$   $df = 11$ ,  $P = 0.927$  ( $P > 0.05$ ) (Table 6).

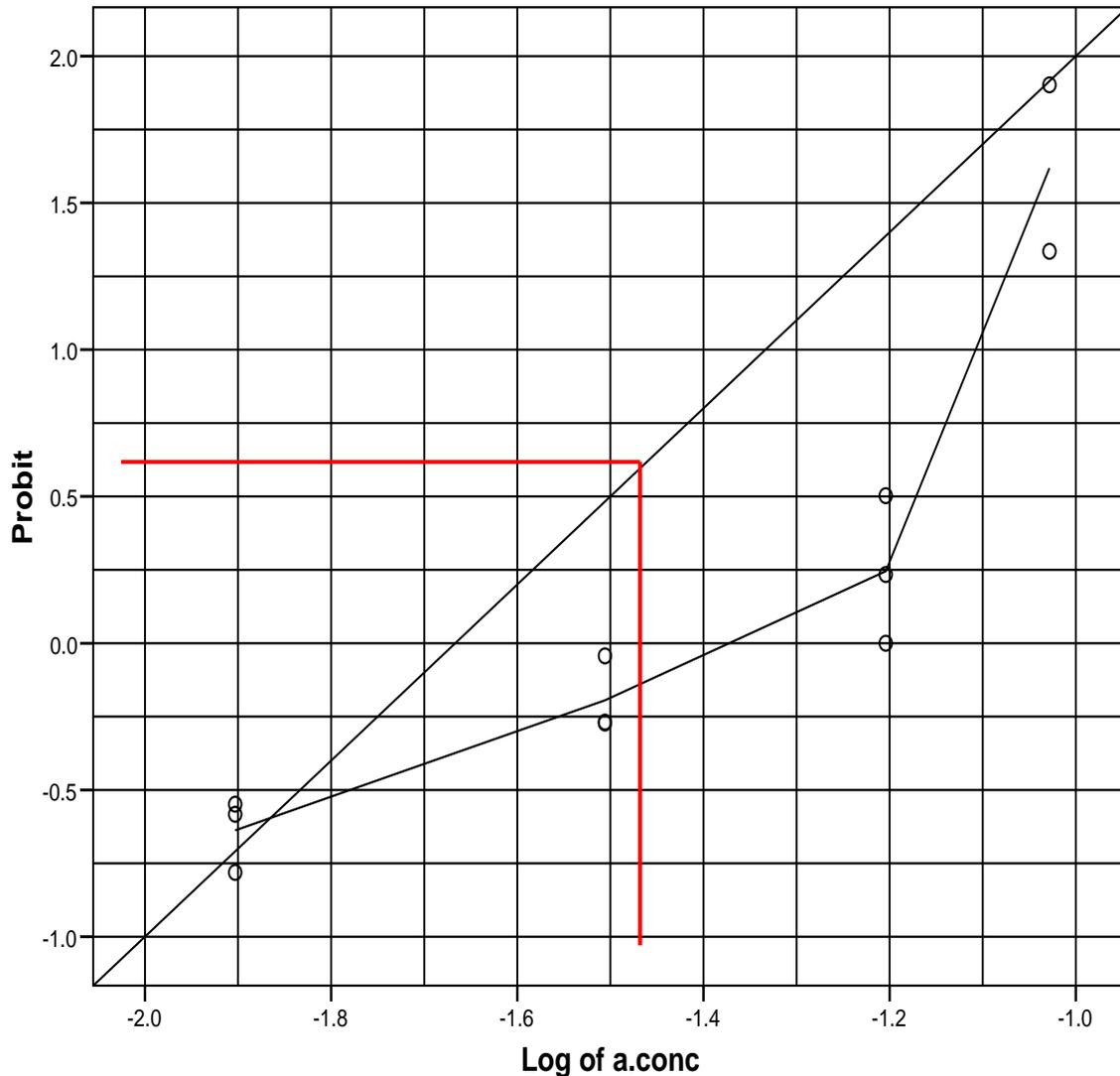
American and European foulbrood diseases were not found in all the colonies subjected to the pathology tests. However, Chalkbrood mummies found in the hive entrance and few in the brood cells of 27 (64.28%) colonies confirmed chalkbrood infection. In addition, these colonies had spotty brood patterns, evidence that the prepupals had formed white or black chalky substances or mummies that occupied the brood cells. These mummies were removed due to the bee hygienic behaviour.

Two of the insecticides used often in the surroundings of the apiaries, DDVP (Dichlorvos) and Cyperforce (Cypermethrin) showed increased mortality on worker honey bees which increased consistently with progressively larger doses (Figures 3 and 4). There were higher significant differences between the mortality of the bees in the control (taken its mortality 26.63% at 48hr as test value) compared to Dichlorvos ( $t = 4.75$ ,  $df = 5$ ,  $p = 0.005$ ) and Cypermethrin ( $t = 3.09$ ,  $df = 5$ ,  $p = 0.027$ ). Similarly, there was a high significant difference between the toxicities of Dichlorvos and Cypermethrin ( $t = 1.67$ ,  $df = 5$ ,  $p = 0.154$ ). The intensity of dose – toxicity effect was particularly weaker with Dichlorvos and stronger with Cypermethrin.

LD<sub>50</sub> of Dichlorvos was  $0.205 \mu\text{g}/\mu\text{l}$  (Slope = 3.125) where regression equation =  $3.125x + 2.625$  (mX + C) (Fig. 3) and Cypermethrin was  $0.039 \mu\text{g}/\mu\text{l}$  (Slope = 3.00) where regression equation =  $3.00x + 5$  (mX + C) (Fig.4) measured as active ingredients in the formulated products. Average weight of bees in DDVP bioassay was 0.38g and in Cyperforce bioassay it was 0.367g. The LD<sub>50</sub> of DDVP in  $\mu\text{g}/\text{g}$  bee ( $(0.205 \times 1.00/0.38)$ )  $0.540 \mu\text{g}/\text{g}$  bee and for Cyperforce it was ( $(0.039 \times 1.00/0.367)$ )  $0.106 \mu\text{g}/\text{g}$  bee (Figures 3 and 4)



**Figure 3: Probit of mortality versus log of concentration (doses) after 48hours for worker honey bees treated with DDVP**



**Figure 4: Probit of mortality versus log of concentration (doses) after 48hours for worker honey bees treated with Cyperforce**

**DISCUSSION**

The results from the study confirmed the general reports from beekeepers about regular colony absconding, poor colony strength and high percentage of colony decline as peculiar behaviour of *Apis mellifera adansonii* species. According to Fletcher [4] *A. m. adansonii* absconds regularly compared to other species of bees, because the species has a high rate of reproduction leading to overcrowding in the nest and rapid consumption of hoarded honey. Fletcher [4] claimed absconding helps

to regulate the hive population. The problem of regular absconding and swarming can be solved and poor colony strength improved through queen honey bee rearing [23]

The presence of a myriad of pests on the colonies confirmed that pest infestation is a problem in beekeeping in the tropics [2, 24, 25, 26]. The pests are responsible for the destruction of the colony and decline in its establishment [2]. Again, the presence of *Varroa destructor*, a parasitic mite could also cause the decline in colony establishment [13, 14, 15, 16]. For acariosis and noseiosis, the absence of Tracheal mites in all the 42 colonies examined could be linked to the claim that acariosis is more associated with the Western honey bees [27, 28, 29], while the presence of *Nosema* spores confirmed noseiosis infection in some apiaries [30]. The severity of the infection symptoms cannot be established. Similarly, it cannot be established whether the infection was due to poor hive management or climatic factors or related to the geographical location. *Nosema* infection can lead to reduction in honey production, colony dwindling, queenlessness and queen supersedure which could eventually cause a colony to collapse [30].

All the colonies testing negative to American foulbrood are in agreement with a study conducted in South Africa which reported that the pest is not yet prevalent among African bees [31]. These researchers pointed out that honeybee pathologists in Africa have not reported cases of outbreak of European foulbrood on the continent. However, Johannsmeier [32] speculated that severe outbreaks of the European foul brood disease might have occurred in the 1940s and 1990s. The presence of chalkbrood disease and percentage of colonies infested may possibly be due to relatively high temperature, rainfall and relative humidity and colony humidity all year round. These conditions are suitable for the growth of the causative fungus [33, 34]. The presence of spotty brood patterns in all the brood colonies confirmed Chalkbrood. The mummified larvae formed as a result of Chalkbrood infection were removed by the worker honey bees as hygienic behaviour and a natural mechanism of resistance [35, 35, 37, 38]. This resulted in low brood population and the spotty brood pattern observed [37, 38].

## CONCLUSION

Insect pests, diseases and pesticide poisoning could be the major factors responsible for the annual decline in honey bee colony establishment. Further research in this field needs to be encouraged, as this will help to sustain the number of honey bee (*Apis mellifera adansonii*) colonies already established in the commercial bee farms in the region.

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**Table 1: Status of the apiary sampled from Ogun, Osun and Lagos States**

Apiary location	Year of establishment	Initial No. of colonies	Present no. of colonies	Percentage decline	Annual Percentage decline
1. OAU, Ife Osun State	7 years	16	3	0.00	0.00
2. Unilag, Lagos State	17 years	12	5	58.34	3.43
3. Shogade , Ifo Ogun State	12 years	13	7	46.2	3.85
4. Coker farm, Ifo, Ogun State	10 years	11	6	45.5	4.55
5 Olowo farm, Ota, Ogun State	8 years	11	7	36.4	4.55
6. Abalabi 1, Ifo, Ogun State	7 years	13	8	38.5	5.50
7. Abalabi 1I, Ifo, Ogun State	6 years	10	6	40.0	6.67
8. Igbusi farm, Ogun State	6 years	8	4	50.0	8.33
9. Okeoko, Ogun State	7 years	9	6	33.3	4.76
10. Lev Farm, Ilesa, Osun State	15 years	20	13	35.0	2.33
11. Ipetumodu Osun State	9 years	25	12	52.0	5.78
12. Iwo, Osun State	7 years	17	10	41.2	5.89
13. Kosere, Ife, Osun State	10 years	13	6	54.0	5.40
14. Opa farm, Ife, Osun State	8 years	12	7	42.0	5.25

**Table 2: *Varroa* mite population levels (mites per bees) in 42 colonies sampled from 14 apiaries monitored during the wet season from May 2010 to October 2010**

Apiary	Colony Samples			Av. Level of infestation
	Colony 1	Colony 2	Colony 3	Mite per bee
1. OAU, Ife Osun State	250/0 mites	100/0 mites	300/0 mites	0.000
2. Unilag, Lagos State	317/10 mite	293/15 mite	312/17 mite	0.046
3. Shogade , Ifo Ogun St.	312/7 mites	304/6 mites	284/10 mites	0.026
4. Coker farm, Ifo, Ogun St.	217/0 mite	243/0 mite	240/0 mite	0.000
5 Olowo farm, Ota, Ogun St.	312/4 mites	210/3 mites	168/2 mites	0.013
6. Abalabi 1, Ifo, Ogun St.	294/16 mites	286/13 mites	250/13 mites	0.051
7. Abalabi 1I, Ifo, Ogun St.	300/60 mites	300/57 mites	152/25 mites	0.184
8. Igbusi farm, Ogun State	300/39 mites	300/27 mites	412/37 mites	0.102
9. Okeoko, Ogun State	100/0 mites	156/0 mite	107/0 mite	0.000
10. Lev. Farm, Ilesa, Osun St.	300/34 mites	279/20 mites	250/16 mites	0.083
11. Ipetumodu Osun State	290/11 mite	210/10 mites	300/20 mites	0.047
12. Iwo, Osun State	306/29 mites	279/20 mites	257/16 mites	0.076
13. Kosere, Ife, Osun State	298/19 mites	345/23 mites	179/19 mites	0.079
14. Opa farm, Ife, Osun State	324/6 mites	235/5 mites	315/2 mites	0.015

**Table 3: *Varroa* mite population levels (mite per bees) in 42 colonies sampled from 14 apiaries monitored during the dry season from November 2009 to April 2010**

Apiary	Colony Samples			Av. Level of infestation
	Colony 1	Colony 2	Colony 3	Mite per bee
1. OAU farm, Ife, Osun St.	290/0 mites	300/0 mites	317/0 mites	0.000
2. Unilag garden, Lagos St.	298/3 mites	321/9 mites	301/6 mites	0.019
3. Shogade Farm, Ifo, Ogun St.	297/6 mites	294/6 mites	304/10 mites	0.024
4. Coker farm setlmt, Ifo	207/0 mite	233/0 mite	340/0 mite	0.000
5 Olowo farm, Ota, Ogun St.	322/4 mites	280/6 mites	268/9 mites	0.022
6. Abalabi 1 farm, Ifo, Ogun St.	299/14 mites	316/13 mites	325/17 mites	0.047
7. Abalabi II farm, Ifo, Ogun St.	310/59 mites	297/56 mites	252/35 mites	0.172
8. Igbusi farm, Ifo, Ogun St.	310/37 mites	297/28 mites	312/32 mites	0.105
9. Okeoko farm, Ota, Ogun St.	300/0 mites	256/0 mite	217/0 mite	0.000
10. Lev Farm, Ilesa, Osun	298/30 mites	299/19 mites	312/21 mites	0.077
11. Tayo Farm, Ipetumodu	230/0 mite	300/18 mites	307/19 mites	0.041
12. Asinwa farm, Iwo, Osun St	312/33 mites	279/19 mites	301/18 mites	0.078
13. Kosere Farm, Ife, Osun St.	256/17 mites	307/18 mites	231/14 mites	0.062
14. Opa farm, Ife, Osun State	199/2 mites	249/7 mites	309/7 mites	0.022

**Table 4: Student *t*- test for significant differences between the levels of infestation of *Varroa* mites samples in the dry and wet seasons**

	Mean	N	Std. Deviation	Std. Error Mean	<i>t</i>	<i>df</i>	Sig. (2 tailed)
wet season	.154857	14	.153411	.041000	-	-	-
dry season	.143143	14	.144941	.038737	-	-	-
Pair1 wet season & dry season	.011714	14	0.02842	0.00759	1.542	13	0.147

**Table 5: Number and differences in *Nosema* spores counted per infested worker bee during the dry and wet seasons: (number of spores = spore counted x (4 x 100) in haemocytometer slide)**

Apiary	Average no. of spores (per bee)		Difference in spore counted
	Dry season	Wet season	
1. OAU, Ife	0.0000	0.0000	0
2. Unilag, Akoka	16,000	16,667	667
3. Shogade Farm, Ifo	21,600	21,600	0
4. Coker farms, Ifo	0.0000	0.0000	0
5. Abalabi 11, Ifo	30,400	28,933	-1,467
6. Igbusi farm, Ifo	29,467	28,800	-667
7. Okeoko, farm, Ota	0.000	0.000	0
8. Lev Farm, Ilesa	24,400	25,067	667
9. Tayo Farm, Ipetu	20,800	21,467	333
10. Asinwa farm, Iwo	21,467	21,067	-400
11. Kosere Farm, Ife	17,333	17,467	134
12. Opa farm, Ife	16,133	16,333	200
Mean	16861.54	16846.23	-15.31

**Table 6: Paired sample test between the means of spore counts for *Nosema* disease during the dry and wet seasons in each three colonies sampled in 11 apiaries**

Pair 1	Paired Differences					df	Sig. (2-tailed)	
	Mean	Std.Dev.	Std. Error	95% Confidence Interval of the Difference				
Nosema spores wet and dry season	-16.583	613.951	177.232	-406.66	373.50	-0.094	11	0.927

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