

Original article

**Mortality of *Varroa destructor* in honey bee  
(*Apis mellifera*) colonies during winter**

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**Abstract** – The change in infestation levels of the mite *Varroa destructor* Anderson and Trueman on adult bees during periods with little or no brood rearing (late October/early November to early February) was investigated in 10 colonies for two consecutive years in a Swedish climate (N57°06' E18°16'). The results do not support the hypothesis that mites become concentrated on the remaining bees as bees die off from the winter cluster. When the number of all mites recovered from dead bees or from debris was used to calculate mites per dead bee, the level of infestation per bee was not significantly different between samples of live bee and dead bees. For modelling purposes, we presently find no reason to differentiate the mortality rates of bees and mites during periods when there is no or limited amounts of brood in the colonies, although the connection between bee mortality and mite mortality may not be as direct as previously assumed.

***Varroa destructor* / population dynamics / winter mortality**

## 1. INTRODUCTION

The biology of the mite *Varroa destructor* Anderson and Trueman, 2000 (formerly *V. jacobsoni* Oudemans) in colonies of the western honey bee (*Apis mellifera*) has been described in some detail (Boot et al., 1994; Donzé and Guerin, 1994; Martin, 1994; Martin, 1995; Donzé et al., 1996). Parallel to

growing information on mite biology, there also is good data on the intrinsic growth rate of the mite population during periods of brood rearing (Calatayud and Verdu, 1995; Kraus and Page, 1995; Marcangeli et al., 1995). However, mite mortality is poorly understood during periods of mite population growth, and even more so during periods with no brood rearing and mite population

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decline. For purposes of modelling, mite population dynamics, data on mite mortality is critical.

Based on assumptions and available data, there have been three attempts to develop mathematical models of the population dynamics of *V. destructor* mites (Calis et al., 1999; Fries et al., 1994; Martin, 1998). It has been assumed in two of these models that mites and bees have a similar death rate during winter (Calis et al., 1999; Fries et al., 1994), yielding a daily mite death rate of 0.004 per day during periods of no brood rearing. The third model used a winter mite mortality of only 50% of other models (0.002) (Martin, 1998). The latter death rate was based on the average of two citations in the literature in which the number of mites that died with bees outside the colonies was not considered (Korpela et al., 1993; Moosbeckhofer, 1991). The death rate of 0.004 was based on the assumption that approximately half of the population of bees may die in a climate similar to Scandinavia (Avitabile, 1978) and that mites are randomly distributed in the winter cluster (Ritter et al., 1989) and do not change host as the bees die (Müller, 1987). Recent data, however, indicate that mites may change their host during winter and that they actually may leave dying hosts before the bees drop from the cluster (Bowen-Walker et al., 1997). Bowen-Walker et al. (1997) studied the distribution of mites in one colony during a period when they assumed there was no brood rearing under British climate conditions and found an increase in infestation level of the adult bees from 12.9% in October–December to 36.8% in January and February. Bowen-Walker et al. (1997) interpreted this result as a differential mortality rate of mites and bees. Later, Bowen-Walker and Gunn (1998) used the data from the same colony and concluded: “As overwintering bees start to die of old age and disease, more and more mites, potentially carrying disease agents, will become concentrated on the remaining bees”.

We studied winter mite mortality in a cold climate. In particular, we investigated whether mites become concentrated on the remaining bees as bees die off during winter.

## 2. MATERIALS AND METHODS

To investigate the changes in infestation level of mites on adult bees during periods with little or no brood rearing, we equipped 10 colonies with net screen bottoms for collection of mites below the screen (30 mm between net and collection tray), and for collection of bees dying inside the colony above the screen (20 mm between net and lower frame bars), for two consecutive years. The colonies were also equipped with an external cage that allowed collection of bees leaving the hives to die outside the colonies. If mites dislodged from bees collected outside the hive, such mites were not collected. Before the experiment started each year, all colonies were examined and found free from sealed brood. At the onset of the experiment, a sample of approximately 1 dl of bees (~ 250) were taken from each colony and examined for mites. Dead bees to be examined for mites and mites in the debris were collected approximately every two weeks until the end of the experiment, when a sample of 1 dl of live bees again was taken from each colony and examined for mites.

The experimental site was located on Gotland (N57°06'; E18°16'). The first year, the experiment started on October 20 and was terminated on February 2 (mean daily temperature  $\pm$  standard deviation;  $1.94 \pm 3.31$  °C). The second year the experiment started on November 16 and was terminated on February 5 (mean daily temperature  $\pm$  standard deviation;  $2.04 \pm 2.45$  °C). The experiments were terminated in early February because honey bees may initiate some brood rearing at this time under similar climate conditions (Avitabile, 1978).

### 3. RESULTS

The numbers of mites recovered per live bee, in the debris, on dead bees, and on dead bees and debris combined are tabulated in Table I.

There was no significant change in the infestation levels in the colonies between the initiation and termination of the experiments in either of the two years (for each year;  $P > 0.05$ , paired  $t$ -test, 9 $df$ ). The same was true when the data from both years were combined ( $P > 0.05$ , paired  $t$ -test, 19 $df$ ). The infestation level of the dead bees was significantly lower than the level on the live bee samples during both years ( $P < 0.001$ , paired  $t$ -test, 9 $df$ ). However when the number of mites recovered in the debris was combined with the mites found on dead bees to give the recovered number of mites per dead bee, the difference was not significant for either of the years ( $P > 0.05$ , paired  $t$ -test, 9 $df$ ), or for the two years combined ( $P > 0.05$ , paired  $t$ -test, 19 $df$ ). To avoid damaging the colonies, they were not checked for brood at the end of the experiment in early February. We assume that brood rearing was very limited in early February under the prevailing weather conditions. Thus, it is plausible that minimal, if any, mite reproduction occurred during the experiment considering the climatic conditions and the fact that all colonies did not have sealed brood at the onset of the experiment each year.

In Figure 1 a scatter diagram of the change in infestation level in live bees at the start and at the end of the experiment (end level minus start level) vs. the number of dead bees is plotted. The correlation coefficient is low ( $r = -0.12$ ) and non-significant ( $P = 0.62$ ) indicating that there is no linear relationship between the two variables.

### 4. DISCUSSION

A problem with the presented data is the level of bee mortality. With low bee

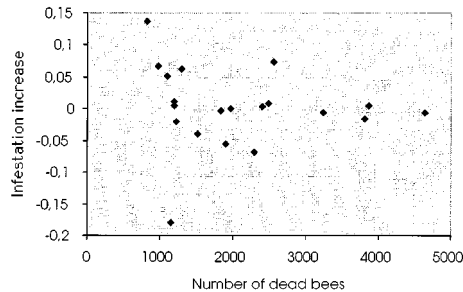
mortality, any change in infestation rate of live bees due to mites leaving dying hosts will be difficult to detect. Nevertheless, if mites do become concentrated on remaining bees as bees die off, we would expect a linear relationship between the variables in Figure 1. However, the correlation coefficient is low ( $r = -0.12$ ) and not significant ( $P = 0.62$ ) indicating that no linear relationship between the variables exists. In other words, an increase in bee mortality does not seem to be linked to an increase in live bee infestation levels.

From the presented data (Tab. I, Fig. 1), we find no support for the conclusion presented by Bowen-Walker and Gunn (1998) that mites become concentrated on the remaining bees as bees die from the winter cluster. What is obvious, however, is the large degree of variation between colonies with an apparent increase in infestation rate in some colonies and an apparent decrease of similar magnitude in others (Tab. I). Sampling errors may explain some of the observed variation, in particular during the first year with a low infestation level. In the second year the variation between colonies prevailed in spite of a higher infestation level, indicating that much variation may be due to individual differences between colonies. This variation remains unexplained.

In the present experiment, we can not determine if the mites found on dead bees fell from the cluster and remounted dead bees. Nor can we determine if mites in the debris fell from the cluster or from already dead bees. When all mites recovered were considered together, the number of mites per dead bee was not significantly different from number of mites per live bee over all colonies. There was a non-significant tendency that the number of mites recovered per dead bee was smaller than the number of mites recovered per live bee. We collected all dead bees both inside and outside the hive entrance, but if mites fell from bees on the outside, they were not retrieved (31%

**Table I.** Number of mites recovered per live bee before and after the experiments, number of mites recovered per dead bee, number of mites recovered in colony debris, and total number of recovered dead mites per dead bee.

Colony	Mites per live bee, before	Mites per live bee, after	Number of dead bees	Number of mites on dead bees	Number of mites in debris	Total recovered mites per dead bee
<b>Year 1</b>						
1	0.013	0.018	1 199	0	5	0.004
2	0.014	0.010	1 839	7	20	0.015
3	0.006	0.012	3 862	2	18	0.005
4	0.008	0.008	1 969	4	20	0.012
5	0.037	0.030	4 645	15	254	0.058
6	0.049	0.052	2 409	10	16	0.011
7	0.076	0.085	2 487	48	62	0.044
8	0.044	0.029	3 809	27	66	0.024
9	0.045	0.024	1 222	5	26	0.025
10	0.012	0.006	3 235	19	9	0.009
<b>Year 2</b>						
1	0.055	0.067	1 195	36	29	0.054
2	0.349	0.486	819	35	76	0.136
3	0.408	0.228	1 157	101	178	0.241
4	0.090	0.152	1 294	100	36	0.105
5	0.206	0.151	1 904	285	33	0.167
6	0.151	0.111	1 516	68	19	0.057
7	0.133	0.200	974	92	37	0.132
8	0.079	0.131	1 093	103	143	0.225
9	0.154	0.086	2 290	49	68	0.051



**Figure 1.** Scatter diagram of infestation increase of live bees between the start and the end of the experiment (end infestation rate minus start infestation rate) vs. number of dead bees recovered. The correlation coefficient ( $r = -0.12$ ) is not significant ( $P = 0.62$ ).

of the dead bees were collected outside the hives). Thus, the number of mites per dead bee may have been slightly underestimated, further weakening the concentration hypothesis. Factors which may add to the differences between this study and the results of Bowen-Walker et al. (1997) could be different opportunities for brood rearing during the studied period (climate) as well as different opportunities for mites to re-mount adult bees after falling from the cluster (due to differences in bottom board construction).

The variation found between the level of infestation among colonies emphasizes the importance of sample size when comparing the mortality rate of mites and bees. The conclusion that mites become concentrated on the remaining bees as bees die off is based on data from one colony only (Bowen-Walker et al., 1997). The data presented here, based on 20 colonies under conditions when brood rearing is unlikely or very limited, and in which mites that fell through the bottom screen probably did not re-enter the bee cluster, does not support this conclusion. For modelling purposes, we presently find no reason to differentiate the mortality rates of bees and mites during periods when there is no or limited amounts of brood in the colonies, although the connection between bee mortality and mite

mortality may not be as direct as previously assumed (Calis et al., 1999; Fries et al., 1994). Further studies of mite mortality, both inside and outside of bee colonies, during periods with and without brood are warranted to further refine the predictive value of existing models of the mite population dynamics.

### Résumé – Mortalité de *Varroa destructor* dans les colonies d'abeilles domestiques (*Apis mellifera*) au cours de l'hiver.

Bowen-Walker et al. (1997) ont étudié la répartition de l'acarien *Varroa destructor* Anderson et Trueman dans une colonie d'abeilles au cours d'une période supposée sans production de couvain dans les conditions climatiques britanniques. Ils ont trouvé une augmentation de l'infestation des ouvrières passant de 12,5 % en octobre-décembre à 36,8 % en janvier-février. Ils ont interprété ce résultat comme étant dû à une différence du taux de mortalité des abeilles et des acariens. Plus tard, sur la base des données de cette même colonie, Bowen-Walker et Gunn (1998) ont conclu : « Quand, durant l'hivernage, les abeilles se mettent à mourir de vieillesse ou de maladie, de plus en plus d'acariens, vecteurs potentiels d'agents pathogènes, se concentrent sur les abeilles restantes ».

Nous avons étudié la mortalité hivernale dans une région à climat froid (N57°06' ; E18°16') et recherché particulièrement si les acariens se concentraient sur les abeilles survivantes au fur et à mesure que la mortalité des abeilles augmentait au cours de l'hiver. Durant deux années consécutives nous avons comparé sur 10 colonies dépourvues de couvain le taux d'infestation des abeilles en début d'expérience (de fin octobre ou mi-novembre) et en fin d'expérience (début février). Les abeilles mortes et les débris de la colonie ont été récoltés environ chaque semaine durant toute l'expérience pour y rechercher les acariens morts. Le tableau I donne le nombre d'acariens par abeille vivante, le nombre d'acariens trouvés

dans les débris et sur les abeilles mortes et leur somme, qui donne le nombre d'acariens par abeille morte. On n'a trouvé de modification significative ( $P > 0,05$ , test- $t$  par paires, 9df) pour aucune des deux années, ni pour les deux années réunies ( $P > 0,05$ , test- $t$  par paires, 19df). Pour chacune des deux années le taux d'infestation des abeilles mortes était significativement plus faible que celui des abeilles vivantes ( $P > 0,001$ , test- $t$  par paires, 9df). Si l'on regroupait les acariens trouvés dans les débris et ceux sur les abeilles mortes, pour obtenir le nombre d'acariens trouvés par abeille morte, la différence n'était significative pour aucune des deux années ( $P > 0,05$ , test- $t$  par paires, 9df), ni pour les deux années prises ensemble ( $P > 0,05$ , test- $t$  par paires, 19df). Les différences trouvées entre colonies soulignent l'importance de la taille de l'échantillon lorsque sont comparés les taux de mortalité des acariens et des abeilles. La conclusion, selon laquelle les acariens se concentrent sur les abeilles survivantes, n'a été tirée qu'à partir de l'étude d'une seule colonie (Bowen-Walker et al., 1997). Les données présentées ici, obtenues à partir de 20 colonies dans des conditions où la production de couvain était improbable ou très réduite, ne confirment pas cette conclusion. Actuellement il n'y a donc aucune raison, lorsqu'on construit des modèles de population, de considérer séparément la mortalité des abeilles et des acariens au cours des périodes où la production de couvain est nulle ou réduite. Pourtant la relation entre la mortalité des abeilles et la mortalité des acariens n'est probablement pas aussi directe qu'on le supposait jusqu'à présent (Calis et al., 1999 ; Fries et al., 1994).

#### ***Varroa destructor* / dynamique populations / mortalité hivernale**

**Zusammenfassung – Sterblichkeit von Varroamilben (*Varroa destructor*) in Honigbienenvölkern (*Apis mellifera*) während des Winters.** Bowen-Walker

et al. (1997) untersuchten die Verteilung von Milben (*Varroa destructor* Anderson und Trueman) in einem Bienenvolk innerhalb eines Zeitraums, für den sie annahmen, dass unter britischen Klimabedingungen keine Bruterzeugung vorhanden war. Hierbei fanden sie eine Zunahme des Befalls der Bienenarbeiterinnen von 12,5 % im Oktober und Dezember auf 36,8 % in Januar und Februar. Sie führten dieses Ergebnis auf eine unterschiedliche Sterberate von Bienen und Milben zurück. Später schlossen Bowen-Walker und Gunn (1998) auf Grundlage dieser Daten, dass "Wenn überwinternde Bienen durch Alterung oder Krankheit sterben, mehr und mehr Milben als potenzielle Träger von Krankheitserregern auf den übrigen Arbeiterinnen konzentriert werden".

Wir untersuchten die Wintersterblichkeit in einem kalten Klimabereich (N57°06' ; E18°16') und untersuchten besonders ob die Milben sich mit fortschreitender Bienensterblichkeit auf den überlebenden Bienen konzentrieren. In zwei aufeinander folgenden Jahren untersuchten wir in 10 brutfreien Völkern im späten Oktober/November die Befallsrate der lebenden Bienen und verglichen sie mit der Befallsrate im frühen Februar. Tote Bienen und das Gemüll wurden etwa wöchentlich bis zum Versuchsende gesammelt und auf tote Milben untersucht.

Die Anzahl von pro lebender Bienen gefundener Milben, von Milben im Gemüll, auf toten Bienen und die Summe von Milben im Gemüll und auf toten Bienen sind in Tabelle I aufgeführt. In keinem der beiden Jahre gab es eine signifikante Änderung ( $P > 0,05$ , gepaarter  $t$ -Test, 9df), auch nicht wenn beide Jahre zusammengefasst wurden ( $P > 0,05$ , gepaarter  $t$ -Test, 19df). In beiden Jahren war die Befallsrate der toten Bienen signifikant niedriger als die der lebenden Arbeiterinnen ( $P < 0,001$ , gepaarter  $t$ -Test, 9df). Wenn die Milben im Gemüll mit den Milben auf den toten Bienen zusammengefasst wurden, um die Anzahl gefundener Milben pro toter Biene zu ermitteln, ist dieser Unterschied allerdings weder in einem der beiden Jahre signifikant ( $P > 0,05$ ,

gepaarter *t*-Test, 9df) noch in beiden Jahren zusammengenommen ( $P > 0,05$ , gepaarter *t*-Test, 19df).

Die zwischen den Völkern gefundenen Unterschiede belegen die Wichtigkeit einer ausreichenden Probengröße bei der Untersuchung der Sterbensraten von Milben und Bienen. Die Schlussfolgerung, dass die Milben sich auf den übrigbleibenden Bienen konzentrieren wenn die Bienen sterben wurde auf Grund der Untersuchung nur eines Volkes gezogen (Bowen-Walker et al., 1997). Die hier dargestellten Daten, erstellt auf Grundlage von 20 Völkern unter Bedingungen, in denen Bruterzeugung unwahrscheinlich oder sehr begrenzt ist, unterstützen diese Schlussfolgerung nicht. Zur Zeit gibt es daher bei der Erstellung von Populationsmodellen keinen Grund, die Bienen – und Milbensterblichkeit zu Zeiten geringer oder keiner Brutstätigkeit getrennt zu betrachten. Dennoch ist die Beziehung zwischen Bienensterblichkeit und Milbensterblichkeit vermutlich nicht so direkt wie bislang angenommen wurde (Calis et al., 1999; Fries et al., 1994).

### **Varroa destructor / Populationsdynamik / Wintersterblichkeit**

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