

## Infestation levels of *Varroa destructor* in local honey bees of Jordan

Yehya Alattal<sup>1</sup>, Peter Rosenkranz<sup>2</sup> & Claus P. W. Zebitz<sup>1</sup>

<sup>1</sup> University of Hohenheim, Institute of Phytomedicine.

<sup>2</sup> University of Hohenheim, State Institute of Apiculture

**Abstract:** To determine *Varroa* mite infestation levels in Jordan, a survey covering 180 colonies of two bee types (*Apis m. syriaca* and *Apis m. syriaca* hybrids) from six locations of 4 climatic zones was conducted during August, 8 month after the last treatment. Sampled colonies had 8-10 frames covered with bees and 3-4 brood frames. Levels of infestation were determined on both adult worker bees and in sealed worker brood cells. Two-way ANOVA showed no significant differences due to bee type with average adult bee infestation of 10.9 % and 13.1 % on hybrid and local bee types, respectively. Average infestation levels in sealed brood worker cells were 37.6 % and 32.5 % in hybrid and local bee types, respectively. Differences in infestation levels on adult bees were significant due to location and ranged between 6.9 - 18.6 % in Daba'a (Desert climate) and Jerash (Dry Mediterranean), respectively. In sealed worker brood cells infestation levels ranged between 15.7 - 84.7 % in Baqa (Dry Mediterranean) and Jerash, respectively. This indicates clearly that the usual scheduled *Varroa* control practice by a single chemical treatment in autumn could be insufficient. Therefore, to prevent damages or even losses of colonies, including diagnosis of infestation rates as part of integrated *Varroa* management is highly recommended.

**Key works:** *Varroa destructor*, mite fertility, *Apis mellifera syriaca*, post-capping period, Jordan

Yehya Alattal, Claus P. W. Zebitz, University of Hohenheim, Institute of Phytomedicine, Department of Applied Entomology, D-70593 Stuttgart. Email: Alattal1@uni-hohenheim.de; zebitz@uni-hohenheim.de

Peter Rosenkranz, University of Hohenheim, State Institute of Apiculture, D-70593 Stuttgart. email: bienero@uni-hohenheim.de

The *Varroa* mite, *Varroa destructor* (ANDERSON & TRUEMAN 2000) is a cosmopolitan destructive ectoparasite of honey bee colonies (*Apis mellifera* L.). The natural host of the mite is the Eastern honey bee *Apis cerana* F. in Southeast Asia. On the new host *A. mellifera*, *V. destructor* has become a threat to beekeeping worldwide (BOOT et al. 1996; RATH 1999). In temperate climate, colonies without annual treatment against *Varroa* mite will shortly collapse whereas in most tropical regions the infestation levels remain below the damage threshold. It is not clear whether tropical climate or the specific bee types of the tropics contribute more to the observed stable host-parasite-relationships (ROSENKRANZ 1999).

In Jordan the mite was discovered first in 1985, one year later *Varroa* mite caused heavy damage led to the loss of about 50 % of the commercial colonies and most traditional colonies (MINISTRY OF AGRICULTURE 1986). Since then, beekeepers have used many acaricides to address the problem, such as bromopropylate, coumaphos, fluvalinate, amitraz, formic acid and oxalic acid (AL-ABADI & NAZER 2003). On the long run, continuous use of acaricides will increase resistance (LODESANI et al. 1995; HILLESHEIM et al. 1996; ELZEN et al. 1999). Beekeepers of Jordan recently treat their colonies yearly in autumn to prevent damage. Yet, after about 20 years of its first incidence in Jordan, *V. destructor* is still the most destructive pest of honey bee colonies.

Jordan provides the facility to compare *Varroa* infestation levels at different climates and in colonies of different bee type at the same time. Therefore, this study has two aims, 1) comparison of the infestation rates in different climatic zones, which may influence the mite population growth differently and subsequently the degree of damage 2) assessment of infestation rates shortly before the annual treatment to demonstrate whether the performed routine treatment is sufficient to prevent economic damages of the colonies. 3) To

compare between two different Honey bee types in the degree of infestation which may indicate the bee type genetic influence on the colony infestation.

### Materials and Methods

In August 2003, samples were taken from 180 bee colonies spread over six locations of four climatic zones in Jordan (Tab. 1). In each location two bee types were collected, colonies of the local bee type *Apis mellifera syriaca* (local bee type) and colonies of its hybrids with the most imported Italian honey bee (hybrid bee type). The hybrid colonies were selected from a group of colonies, which were headed with imported *Apis mellifera ligustica* from Kona Queen (USA) one year before the treatment. These colonies were then left for natural hybridization with the local race. Fifteen colonies of each bee type in each location were collected. Sampled colonies had 8-10 frames covered with bees and 3-4 frames of sealed brood. Colonies were not treated during the season and the last treatment was about 8 month before sampling.

Tab. 1: Name, latitude, climatic zone of sampled sites and number of colonies per location.

Location name	Latitude	Climatic zone	No. of colonies
Jordan Valley	31.95N-35.58E	Tropical	30
Yadodah	31.85N-35.93E	Mediterranean	30
Suweileh	32.30N-35.84E	Mediterranean	30
Daba'a	31.55N-36.05E	Desert	30
Jerash	32.20N-35.90E	Dry Mediterranean	30
Baqa	32.06N-35.85E	Dry Mediterranean	30

Relative infestation of adult bee was estimated by brushing about 200 adult worker bees from brood combs into small plastic jars (500 cc). Jars were filled with water and detergent and vibrated for a while then adult bees were separated from *Varroa* mite using double layer filter honey sieve under water stream. Relative infestation was calculated as (number of *Varroa*/ number of bees). Worker brood relative infestation was estimated by cutting a piece of about 200 cells from sealed brood combs, then brood samples were cut from the middle and washed with water over double layer filter sieve to separate *Varroa* individuals which were then counted to determine relative infestation. Infestation ratio (infestation in brood cells/infestation on adult bees) was also calculated (WOYKE 1987; ROSENKRANZ 1999).

Statistical analysis of different parameters was performed using statistical analysis system (SAS) on transformed data with arc sin function. Significance of main factors and interaction was determined based on ANOVA using LSM. The values presented are actual non-transformed data, given as mean  $\pm$  standard deviation (ARECHA VALETA-VELASCO & GUZMAN-NOVA, 2001).

### Results

None of the sampled colonies were free from *Varroa* mite with high differences in infestation levels among colonies and between locations. Over-all average infestation levels were 12 % and 35 % on adult bee worker and in sealed brood cells, respectively. Brood sealed cells had 4 times more *Varroa* than on adult bees (Tab. 2)

Tab. 2: Relative adult bee infestation, worker brood cells infestation, infestation ratio and over all average infestation of *Varroa* mite in both bee types (local and hybrid). (SD = std. dev.)

Bee type	No of colonies	Adult average infestation $\pm$ SD [%]	Worker brood average infestation $\pm$ SD [%]	Infestation ratio $\pm$ SD
Local bee type	90	13.13 $\pm$ 10.02	32.46 $\pm$ 17.79	3.49 $\pm$ 3.05
Hybrid bee type	90	10.88 $\pm$ 12.42	37.63 $\pm$ 29.34	4.76 $\pm$ 3.52
Over all average	180	12.01 $\pm$ 11.22	35.05 $\pm$ 23.56	4.12 $\pm$ 3.29

Both bee types were not significantly different in the average relative *Varroa* infestation on both adult bees and in sealed worker brood cells. The ratio of the infestation of brood cells and adult bees was also not significantly different due to bee type.

Average adult infestation rates were 10.9 % and 13.1 % on hybrid and local bee types, respectively, while infestation rates of sealed worker brood cells were 37.6 % and 32.5 % in hybrid and local bee type, respectively. Hybrid colonies had about 1.2 more *Varroa* in brood cells than local types (Tab. 2).

Results show that location has significant influence on infestation rates of both adult bee and sealed worker brood cells with ( $F = 2.41$ ;  $DF = 5$ ;  $P > F = 0.038$ ) and ( $F = 11.69$ ;  $DF = 5$ ;  $P > F = 0.0001$ ) respectively, but no significant influence on infestation rates. The highest infestation rates were in Jerash with 18.6 % and 84.7 % on both adult bees and in sealed brood cells (Tab. 3).

Differences in the infestation rates of adult bees between both bee types at different locations (location x bee type interaction) were not significant, while these differed highly significantly between relative infestation levels of sealed worker brood cells ( $F = 4.96$ ;  $DF = 5$ ;  $P > F = 0.0003$ ). This means that the differences between brood infestation levels and therefore differences in the ration of brood/bee infestation depend on the location (Tab. 4).

Tab. 3: Average relative *Varroa* mite infestation on adult bee, sealed brood and the infestation ratio in the different experimental sites.

Location	Mean infestation on adult bee [%]* $\pm$ SD	Mean infestation in sealed brood cells [%]* $\pm$ SD	Infestation ratio* $\pm$ SD
JordanValley.	11.16 $\pm$ 6.29 a	30.14 $\pm$ 10.40 b	4.03 $\pm$ 3.71 a
Yadodah	16.30 $\pm$ 22.57 a	30.28 $\pm$ 21.35 bc	3.33 $\pm$ 2.90 a
Swieleh	11.77 $\pm$ 18.17 a	24.07 $\pm$ 16.68 bc	4.69 $\pm$ 5.24 a
Daba'a	6.93 $\pm$ 4.11 a	25.45 $\pm$ 16.72 bc	4.43 $\pm$ 2.72 a
Jerash	18.64 $\pm$ 9.90 a	84.67 $\pm$ 63.65 a	5.27 $\pm$ 2.96 a
Baqa	7.24 $\pm$ 6.24 a	15.66 $\pm$ 12.57 c	2.99 $\pm$ 2.19 a

\*Means sharing same letters in the same column are not significantly different, Tukey's test,  $DF = 168$   $P < 0.05$ .

Tab. 4: The relative infestation level of *Varroa* in sealed brood cells for both bee types at each location (Location x bee type).

Location	Bee type	Average infestation of adult bees [%]* $\pm$ SD	Average infestation of sealed brood cells [%]* $\pm$ SD
Jordan Valley.	Hybrid	6.39 $\pm$ 3.37 b	21.98 $\pm$ 11.67 c
	Local	15.93 $\pm$ 9.21 ab	38.30 $\pm$ 9.12 b
Yadodah	Hybrid	18.46 $\pm$ 23.62 a	41.60 $\pm$ 29.85 b
	Local	14.14 $\pm$ 21.52 ab	18.95 $\pm$ 12.86 c
Sweileh.	Hybrid	11.82 $\pm$ 24.72 ab	15.05 $\pm$ 9.88 c
	Local	11.72 $\pm$ 11.62 ab	33.09 $\pm$ 23.48 c
Dabaa	Hybrid	7.20 $\pm$ 5.27 b	24.55 $\pm$ 18.88 c
	Local	6.66 $\pm$ 2.97 b	26.36 $\pm$ 14.58 c
Jerash	Hybrid	14.12 $\pm$ 9.24 ab	105.33 $\pm$ 90.83 a
	Local	23.07 $\pm$ 10.57 a	64.01 $\pm$ 36.47 a
Baqa	Hybrid	7.19 $\pm$ 8.27 b	17.29 $\pm$ 14.97 c
	Local	7.28 $\pm$ 4.21 b	13.02 $\pm$ 10.16 c

\*Means sharing same letters in the same column are not significantly different. Mean separation based on ANOVA and LSM,  $DF = 168$ ;  $P < 0.05$ .

## Discussion

The average relative infestation levels show that *Varroa* mites may reach high infestation levels within short times, after about 8 months of the last treatment with formic acid at the end of the season.

However these levels were still below 15 %, which is considered to be an economic threshold on adult worker bees in temperate zones, while the relative infestation levels in sealed worker brood cells exceeds the economic threshold (30 %) even shortly after the end of the season in August (TOSCANO 1996).

In both bee types, adjacent colonies in the same apiary may have very different infestation levels. This could be due to differences in individual colony development which influence the reproductive rates of the *Varroa* populations. Furthermore, specific robbing activities of individual colonies lead to an unequal distribution of mites between colonies kept in groups (GREATTI et al. 1992). Although damage and colony losses were not noticeable at the time of the study, a two years study on population dynamics in Jordan confirmed that most of the untreated bee colonies die during winter (Alattal, unpublished data). Therefore, neither the Jordan climate nor the local bee type provide the basis for a stable host-parasite-relationship at Jordan.

No significant influence of bee type on relative infestation levels indicates that *Varroa* mite may reach damage levels independently of the local bee genotypes. The defensive Syrian honey bee and its hybrid with the Italian honey bee obviously lack any specific *Varroa* resistant trait (ROSENKRANZ & ENGELS 1994; FRIES et al. 1994).

Results indicate that locations could not be classified according to their climatic zones or climatic parameters in expecting high or low infestation levels. Although the highest infestation level on both adult and brood cells were in a dry Mediterranean zone, significant differences in the infestation levels between two locations within one climatic zone exist. On the other hand, the infestation levels in Jordan valley with a tropical climate were not as low as expected from investigations in tropical Africa and South America (ROSENKRANZ 1999). This indicates that relation between location and relative infestation is rather a product of interaction between several factors including ecological factors, bee type and genotype of *Varroa* mite, which is currently under investigation. Additionally, the beekeeper practices and the effectiveness of the *Varroa* treatment in the year before could influence the infestation levels at certain apiaries. However, there exist general recommendations for the beekeeping technique and *Varroa* treatment at Jordan and, therefore, it is not likely that there are huge individual differences among the apiaries tested in this study.

The ratio between sealed brood and adult infestation in different locations was not significantly different. This value is important because it reflects the relative amount of reproducing mites in the bee colony. The highest ratio was in Jerash, which indicates that relative increase in *Varroa* mite population is higher than in other locations at the time of the study which means most of the *Varroa* population will be in the phoretic state. In such cases colony losses due to high adult worker bee infestation levels later after the brood area is reduced is higher than in other places.

From our results it is obvious that bee colonies in different locations are different in their relative infestation at a specific time of the year due to interaction between ecological factors, bee type and *Varroa* mite. We also demonstrated clearly that the usual scheduled *Varroa* control practices by a single chemical treatment in autumn could be insufficient. Therefore, to prevent damages or even losses of colonies, inclusion of diagnosis of infestation rates as part of integrated *Varroa* management is highly recommended

## Acknowledgment

Praise and thanks to DAAD, Bonn for Ph.D. fellowship support and Eiselen Foundation, Ulm for financing this study (project no. 18/2004).

## References

- AL-ABADI, A. & NAZER, I.K. (2003): Control of *Varroa* mite (*Varroa destructor*) on honey bees by aromatic oils and plant materials. – Agriculture and marine science 8 (1): 15-20.
- BOOT, W.J., CALIS, J.N.M., BEETSMA, J., HAI, D.M., LAN, N.K., TOAN, T.V., TRUNG, L.Q. & MINH, N.H. (1996): The phenomenon of the non-reproduction in worker cells as a *Varroa*-tolerance factors involves natural selection of the mites. – Apidologie 27 (4): 283.

- ELZEN, P.J., EISCHEN, F.A., BAXTER, J.R., ELZEN, G.W. & WILSON, W.T. (1999): Detection of resistance in US *Varroa jacobsoni* Oud. (Mesostigmata:Varroidae) to the acaricide fluvalinate. – *Apidologie* 30 (1): 13-17.
- FRIES, I., CAMAZINE, S. & SNEYD, J. (1994): Population dynamics of *Varroa jacobsoni*: a model and review. – *Bee World* 75 (1): 5-28.
- HILLESHEIM, E., RITTER, W. & BASSAND, D. (1996): First data on resistance mechanisms of *Varroa jacobsoni* (Oud.) against tau-fluvalinate. – *Exp. Appl. Acarol.* 20: 283-296.
- LODESANI, M., COLOMBO, M. & SPREAFICO, M. (1995): Ineffectiveness of Apistan® treatment against the mite *Varroa jacobsoni* (Oud.) in several districts of Lombardy (Italy). – *Apidologie* 26 (1): 67-72.
- ARECHAVALETA-VELASCO, M.E. & GUZMAN-NOVA, E. (2001): Relative effect of four characteristics that restrain the population growth of the mite *Varroa destructor* in honey bee (*Apis mellifera*) colonies. – *Apidologie* 32 (2): 157-174.
- MINISTRY OF AGRICULTURE OF JORDAN (1986): Department of Agricultural statistics. Beekeeping statistical book.
- RATH, W. (1999): Co-adaptation of *Apis cerana* Fabr. and *Varroa jacobsoni* Oud. – *Apidologie* 30 (2): 97-110.
- ROSENKRANZ, P. (1999): Honey bee *Apis mellifera* L. tolerance to *Varroa jacobsoni* Oud. in south America. – *Apidologie* 30 (2): 159-172.
- ROSENKRANZ, P & ENGELS, W. (1994): Infertility of *Varroa jacobsoni* females after invasion into *Apis mellifera* worker brood as a tolerance factor against Varroaosis. – *Apidologie* 25 (4): 402-411.
- SAS INSTITUTE INC. (1989): SAS/SAT Users's Guide, Version 6,4<sup>th</sup> ed., Vol. 1 & Vol. 2. – SAS Institute Inc.,Cary, NC: 1028 pp.
- TOSCANO, H. (1996): Estado actual de algunas enfermedades apícolas en Uruguay, Periodo 1985-1995. – Report of the Direccion de Laboratorios Veterinarios 'Dr Mc Rubino' (Di.La.Ve): 6-8.
- WOYKE, J. (1987): Comparative Population Dynamics of *Tropilaelaps clareae* and *Varroa jacobsoni* mites on honeybees. – *J. of Apicultural Research* 26 (3): 196-202.

