

## Reproduction of *Varroa destructor* in sealed worker bee brood cells of *Apis mellifera carnica* and *Apis mellifera syriaca* in 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:** The reproduction of the honey bee mite, *Varroa destructor* in sealed worker bee brood cells represents an important factor for the population development of this parasite in honey bee colonies. In this study, the relative infestation levels of worker brood cells, mite fertility (mites that lay at least one egg) and reproductive rate (number of viable adult daughters per mother mite) of *Varroa* mite in worker brood cells of *Apis m. carnica* and *Apis m. syriaca* were compared in fall 2003 and summer 2004 at two locations in Jordan. The relative infestation levels in sealed worker brood cells ranged from 23 – 32 % in fall and 19 – 28 % in summer. The average fertility of *Varroa* mite ranged between 90 - 98% in colonies of *A. m. carnica* and between 88 - 96 % in *A. m. syriaca* with minor differences between colonies and locations. The number of total progeny of fertile mites in worker brood cells was 4.0 in both bee races. The reproductive rate was high with 2.7 and 2.6 in both honey bee races. The post-capping period of the worker brood cells differs only slightly between both bee races and between locations (284.4 h on average, n = 4,000). Our data reveal surprisingly high mite fertility and reproductive rates in both honeybee races under Mediterranean conditions of Jordan. The possible physiological background of *Varroa* reproduction and the impact of mite fertility on the development of *Varroa* tolerance are discussed.

**Key words:** *Varroa destructor*, fertility, *Apis m. syriaca*, *Apis m. carnica*, 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

*Varroa* mite, *Varroa destructor* (ANDERSON & TRUEMAN 2000), a destructive ectoparasite of honey bees, reproduces in sealed brood cells of honey bee colonies *Apis mellifera* L. (MARTIN 1994). In its original host, *Apis cerana* FABR. the mite reproduces only in drone sealed brood cells, which among other factors lead to balanced host-parasite relationship (RATH 1999 & ROSENKRANZ et al. 1993). In *Apis mellifera* L. colonies, the mite's new host, reproduction occurs in both drone and worker sealed brood cells. This leads to rapid increase in *Varroa* population. Therefore, damages in untreated colonies occur within short time periods (ROSENKRANZ 1999). It is assumed that limited reproduction of *Varroa* mite females in worker brood cells is correlated with increased tolerance to varroasis, as it could be demonstrated with the Africanized honey bees of Brazil (ROSENKRANZ 1999; ROSENKRANZ & ENGELS 1994; MARTIN & KRYGER 2002.).

Two aspects of mite reproduction are of particular interest: mite fertility (percentage of egg laying female mites) and reproductive rate (number of viable female off-springs per mother mite). Typically, a female *Varroa* mite produces one male and three to four female progeny, from which then the male with one or two daughters will reach maturity (MARTIN 1994 & IFANTIDIS et al. 1999).

The developmental time of worker brood cells during the capping period limits the time of *Varroa* mite reproduction. Therefore, it influences the total number of progeny and the reproductive rate of mother mite.

In Jordan, little is known about *Varroa* mite fertility and reproduction in worker brood cells of the local race *Apis m. syriaca*.

The aim of the study is to investigate mite reproduction in colonies of *Apis m. syriaca* and *Apis m. carnica* under Mediterranean climate of Jordan, as well as studying worker brood post-capping period in both races.

### Materials and Methods

Colonies of the local race *Apis m. syriaca* and colonies headed with queens of *Apis m. carnica* reared in Germany were used (n = 20 in total). The evaluations were carried out at two locations (Baqa; 32.06N: 35.85E, Dry – Mediterranean, and Yadodeh; 31.85N:35.92E Mediterranean). Evaluation was done in fall, 2003 and summer, 2004 in Jordan. All colonies were not treated against *Varroa* mite for the last 10 months at the beginning of the study.

Relative infestation of sealed worker brood cells was determined by checking individual brood cells directly from seventeen colonies out of twenty included in the evaluation. The assigned colonies were in total 9 *Apis m. syriaca* and 8 *Apis m. carnica* divided between both locations (Tab. 1). The relative *Varroa* infestation was determined in both bee races in Baqa and Yadodah in fall, 2003 and summer 2004. Relative brood infestation was calculated as the percentage of infested cells divided by the total number of cells examined per colony. Mite fertility (mites that lay at least one egg) was measured by checking 28 - 35 infested sealed worker brood cells from each colony. Only cells infested with one mother mite and containing late stages of white-eyed pupae or elder were considered (REMBOLD & KREMER 1980; ROSENKRANZ & ENGELS, 1994).

The number of males, eggs, protonymphs, deutonymphs and adult daughters were counted in sealed worker brood cells (n = 263 infested brood cells, Tab. 1) of both *Apis m. carnica* and *Apis m. syriaca*. For this purpose cells with fully developed adult worker bees shortly before being emerged (more than 270 hours after cell capping) were considered as described by ROSENKRANZ & ENGELS, (1994). These data were analyzed independently of both, locations and seasons.

The post-capping period of worker brood was determined in twenty bee colonies, five colonies per race in each location. The evaluation was done in fall, 2003 and summer, 2004 using the same colonies in each time. From each colony 100 worker brood cells were marked shortly before capping and were monitored every 8 hours during capping and at time of emergence. The capping period was then calculated (n = 4,000).

Data of mite fertility, mite reproduction and post-capping period were subjected to transformation using Logarithmic function. Three-way analysis of variance of three factors (Race, location and season) using statistical analysis system SAS, was applied on data of mite fertility and post capping period. One-way ANOVA was performed on data of mite reproduction. Separation of means was done based on ANOVA and LSM. All numerical means and values are original not transformed values.

### Results

The relative infestation levels of sealed worker brood cells in individual colonies ranged from 23 – 32 % in fall and 19 – 28 % in summer (Tab. 1).

Average mite fertility was surprisingly high with over all average of 93 %. Differences in average mite fertility were minor in different locations or between both races and ranged between 88 – 98 % (Tab. 1). Both races showed relatively lower mite fertility at Yadodah with 88 % and 89 % for *Apis m. syriaca* in summer and fall, respectively, and 90 % and 93 % for *Apis m. carnica* in summer and fall, respectively (Tab. 1). Mite fertility in individual colonies ranged between 73 – 100 % in *Apis m. syriaca* and 74 – 100 % in *Apis m. carnica*.

Results showed no significant differences due to race or to season in mite fertility. On the other hand, mite fertility was significantly influenced by location (F = 19.9; DF = 1; P>F = 0.0001). Other interactions were not significant.

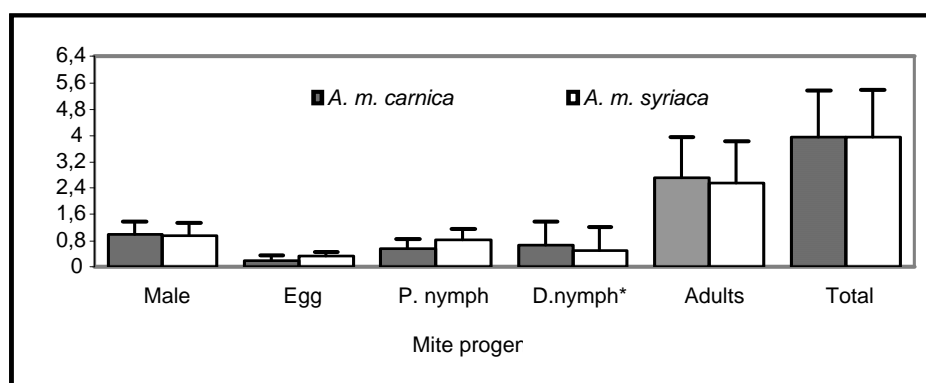
Our results reveal high average number of adult daughter mite produced with 2.7 and 2.6 in *Apis m. carnica* and *Apis m. syriaca*, respectively. The total number of mite progeny was 4.0 and 3.9 in both races respectively (Fig. 1).

Average number of viable daughters, total number of mother mite progeny and number of males were not significant due to bee race . Average number of deutonymphs was slightly different between both races (F = 4.3; DF = 1; P>F = 0,04).

Tab. 1: Average *Varroa* mite fertility in colonies of *Apis. m. carnica* and *Apis. m. syriaca* at two different locations in fall 2003 and summer 2004).

		No. of colonies	Total examined cells	No. infested cells	Infestation of brood [%]	Fertility [%] ±SD	
Fall	Baqa	<i>A. m. carnica</i>	4	468	121	26	98 ± 37.40 a*
		<i>A. m. syriaca</i>	5	531	161	30	96 ± 38.65 ab
	Yadodeh	<i>A. m. carnica</i>	4	535	122	23	93 ± 37.55 ab
		<i>A. m. syriaca</i>	4	371	120	32	89 ± 37.71 ab
Summer	Baqa	<i>A. m. carnica</i>	4	536	144	27	94 ± 37.46 ab
		<i>A. m. syriaca</i>	5	853	164	19	98 ± 38.42 a
	Yadodeh	<i>A. m. carnica</i>	4	485	126	26	90 ± 38.16 ab
		<i>A. m. syriaca</i>	4	468	131	28	88 ± 37.77 b

\* Different letters indicate differences between means, based on ANOVA and LSM tests.



\*Differences between races at that level are significant, TSRT; DF=260; P<0.05.

Fig. 1: Number of males, eggs, protonymphs, deutonymphs, adult daughters and total progeny per mother mite in sealed worker brood cells shortly before emergence (>270 hours after capping) in colonies of *A. m. carnica* and *A. m. syriaca*.

Tab. 2: Post capping period of both bee races *A. m. syriaca* and *A. m. carnica* at different locations.

Location	Honey bee race.	No. of colonies	Total number of cells observed.	Average post-capping period ± SD
Baqa	<i>A. m. carnica</i>	5	1000	285.3 ± 5.1 a*
	<i>A. m. syriaca</i>	5	1000	282.7 ± 5.2 c
Yadodah	<i>A. m. carnica</i>	5	1000	284.7 ± 5.3 a
	<i>A. m. syriaca</i>	5	1000	285.0 ± 5.3 a

\*Different letters indicate differences between means, TSRT; DF= 5; P<0.05.

The average post-capping period was 284.4 hours (n = 4,000, Tab. 2). The shortest average post-capping period was in colonies of *Apis m. syriaca* in Baqa with 282.7 h and the longest average post-capping period was for colonies of *Apis m. carnica* in Baqa with 285.3 h.

The average post-capping period of worker brood cells was not significantly different due to season or to location, but it was significant due to race (F = 4.15; DF = 1; P>F = 0.05) and due to location – race interaction (F = 5.7; DF = 1; P>F = 0.02).

### Discussion

Our data reveal surprisingly higher overall mite fertility (93 %) on both races than stated previously (GARRIDO 2004; ANDERSON & FUCHS 1998; ANDERSON & TRUEMAN 2000; MARTIN 1994; ROSENKRANZ & ENGELS 1994). In the Africanized honey bee, which is a clear example of a tolerant honey bee, mite fertility ranged between 50 % in 1985 to 82 % in 2001 (GARRIDO et al. 2001).

In our study average mite fertility ranged between 88 % and 98 % with no significant differences due to bee race or to season. This means that *Varroa* mite is reproducing in colonies of the Syrian honey bee as successful as in the Carniolian honey bee. On the other hand mite fertility in Carniolian honey bee colonies was higher in Jordan than in Europe (ROSENKRANZ 1999).

Average viable adult daughter was high in both races with about one more adult daughter per mother mite than described before (DEGRANDI-HOFFMAN et al 2002; MARTIN & KRYGER 2002; ROSENKRANZ et al, 1999). This high fertility of both races and high number of adult mite daughters could be related to the genetic composition of *Varroa* mite in Jordan or to other ecological factors. Subsequently, *Varroa* mite will increase quickly and may reach damage levels shortly. In addition to that, only worker brood cells shortly before emergence “about 11.5 days after capping” were included in this analysis. This could be the direct reason for the increased reproductive rate per fertile female mite

In our study, we could show that the host genotype did not affect total mite progeny per mother mite, while in other situations host genotype was found to affect mite reproduction (ROSENKRANZ & ENGELS, 1994; GUZMAN NOVA et al, 1996).

Although, the post-capping periods differ significantly due to bee race, this small difference (1.2h) should have no influence on the total progeny or the number of viable daughter produced between both races. On the other hand, other bee races found to have shorter post-capping period.

From our study it is clear that reduced mite fertility does not exist in colonies of *Apis m. syriaca* and the mite is able to reproduce in these colonies as successful as in colonies of *Apis m. carnica*. It should be clarified, if differences in mite genotype influence fertility and reproductive rate.

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