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INFLUENCE OF QUEEN MOTHERS ON THE HONEYBEE HIVE VARROA MITE POPULATION IN THE YEAR 1998

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ABSTRACT

Varroa mite population was studied in 317 hives located in 40 apiaries in July 1998. Queens originated from 46 mother queens were reared at 16 different rearing locations in Slovenia. Diagnostic treatment represented 10 ml of 85 % formic acid applied on the bottom board. Fallen mites were counted 48 hours after treatment. We found a highly significant effect of the apiary on the varroa infestation by using the analysis of variance. Queen rearing locations and mother queens nested within queen rearing locations did not affect a level of varroa infestation significantly. LSMEAN estimate extracted two mother queens with extremely small varroa population (Kropivšek b/97 and Vozelj 1/97). There was no evidence of a colony having a significantly higher level of varroa mite infestation than tested population of bees. Highly infested colonies yielded approximately 2 kg less honey than less infested bee colonies.

Key words: honey bees / queens / origin / selection / disease / varroa / Slovenia

VPLIV MATER ČEBELJIH MATIC NA POPULACIJO VAROE V ČEBELJIH PANJIH V LETU 1998

IZVLEČEK

Proučevali smo prisotnost varoe v 317 panjih. Matice so po poreklu izvirale od 46 mater, zrejene so bile v 16 različnih zrejališčih v Sloveniji. Panjsko populacijo varoe smo ugotavljali na 40 različnih mestih (čebelnjakih) v juliju 1998. Uporabili smo diagnostično metodo zdravljenja čebel z 10 ml 85 % mravljinčne kisline na podnici panjev. Mrtve odpadle varoe smo šteli po 48 urah. Analiza variance je pokazala visoko značilen vpliv čebelnjaka in neznačilne vplive zrejališč in mater čebeljih matic znotraj zrejališč. LSMEAN ocena je izločila dve materi čebeljih matic z izjemno maloštevilno populacijo varoe (Kropivšek b/97 in Vozelj 1/97). Nismo našli nobene matere matic z značilno večjo panjsko populacijo varoe od povprečja vseh mater. V zelo okuženih čebeljih družinah lahko pričakujemo za okoli 2 kg manj medu kot v najmanj okuženih panjih.

Ključne besede: čebele / matice / poreklo / selekcija / bolezni / varoza / Slovenija

INTRODUCTION

Disease caused by *Varroa jacobsoni* Oudemans is one of the central problems that influences honeybee population. Smirnow (1978) reported severe damage in honeybee industry caused by losses of colonies in China in the sixties. Varroa is nowadays spread all over the world except in Australia and New Zealand. Varroa caused losses of more than 100.000 colonies in Argentina (Dietz, 1986), 300.000 in Spain (Gomez, 1988, quoted by Martin, 1998). It has a devastating effect on the beekeeping industry of USA. More than 75 % of colonies were lost in some districts of California (Kraus and Page, 1995, quoted by Martin, 1998), and 71 % in Arizona respectively (Loper, 1995). The first honeybee colonies in Slovenia were infected in 1980 (Rihar, 1987). Many different ways of varroa control procedures have been used among Slovenian beekeepers since that time. They have had to manage their bees with much greater effort and costs since that time. Large damages have been prevented by Apistan, which became ineffective in the recent time (Trouiller, 1998). Specialists in bee diseases have been trying to find an alternative way to control varroa, primarily with essential oils which are mainly used as the registered product Apilife VAR (Gregorc and Jelenc, 1996).

The study of honeybee varroa resistance has been running under the program of the Carniolan Honeybee Selection at the Agricultural Institute of Slovenia. 1072 young honeybee queens were distributed in July 1997. The next season, in 1998, queens were introduced into the colonies of 78 apiaries to test their production. We decided to conduct the varroa mite treatment with formic acid at the beginning of July 1998. Some benefits were expected from that action: the Slovenian Animal Health Center should give very useful information about the level of varroa mite infestation in Slovenia so that beekeepers will be warned on time. The Central Republic Selection Service would receive correct information on genetic sources of honeybee varroa mite tolerance. The impact of the level of varroa mite infestation on honey yields, the swarming tendency and the colony defensive activity is discussed.

MATERIAL AND METHODS

Varroa control treatment was carried out from 5 to 15 July 1998. We visited 39 beekeepers and 317 honeybee colonies were investigated. The queens originated from 46 queen mothers. 10 ml of 85 % formic acid on the hive bottom board was applied as a diagnostic treatment of tested colonies. After the formic acid treatment each hive was carefully isolated. Dead varroa mites fell down under the bottom board wire mesh. Dead varroa were sampled after 48 hours. Data were processed using the SAS GLM model:

$$Y_{ijk} = \mu + a_i + b_j + c_{jk} + e_{ijkl} \qquad \text{(Model 1)}$$

Y_{ijk} = Estimated value of varroa population in tested honeybee colonies. Estimating includes the influence of apiary, queen rearing location and mother nested within the queen rearing location)
μ = mean of population
a_i = influence of the apiary on varroa mite population
b_j = influence of the queen rearing location on varroa mite population
c_{jk} = influence of queen mother nested within queen rearing location on varroa mite population
e_{ijkl} = error

The model included the main source of variance, apiary, which sums up many particular influences of local environment and beekeeper activities. The second source of variance was the origin of queens and the influence of place where the queens were reared. The third source of variance was the influence of the queen mother nested within the rearing location. Least square means values for queen mothers nested within the queen rearing location were estimated after that (option lsmean pdiff). The differences between them were carefully studied, afterwards the queen mother lines with significant difference to other queen mothers were sampled. All the values of p H0 >: $lsmean_i = lsman_j$ smaller as 0.05 were used as a criterion.

Data on the production of bee colonies were collected in September 1998. Honey yields were weighed in kg, swarming expression was estimated on four scale system (1 – the colony swarm, 4 – the colony expresses no signals of swarming, Ruttner, 1988). The aggressive behaviour was estimated on four scale system too (1 – the colony is extremely aggressive, 4 – the colony is extremely gentle). The influence of all known independent variables on honey yields of honeybee colonies was studied using the following model:

$$Y_{ijk} = \mu + a_i + b_j + b1\,(x_k - x) + b2\,(x_l - x) + b3\,(x_m - x) + e_{ijklmn} \qquad \text{(Model 2)}$$

Y_{ijk} = Estimated value of honey yield in kg as the result of apiary, origin of queen and regressions of varroa mite infestation level, swarming expression and aggressive behaviour of bees
a_i = influence of apiary on honey yield
b_j = influence of origin of queen on honey yield
x_k = the value of regression of varroa mite infestation - honey yield
x_l = the value of regression of colony swarming expression - honey yield
x_m = the value of regression of aggressive behaviour - honey yield
e_{ijklmn} = error

RESULTS AND DISCUSSION

Many methods to enhance varroa resistance (Harbo *et al.*, 1997) are included in selection programs of honey bees in several countries. Erickson and others (1998) reported about very promising selective honeybee varroa tolerant breeding in Arizona. An extensive program is running in Austria (Boigenzahn and Willam, 1999). Selection goal, i.e. to increase the honey bee varroa tolerance, can be achieved by using particular mechanisms of varroa mite resistance:

- Grooming and cleaning behaviour of worker bees can have a strong influence on the hive varroa population. Božič and Valentinčič (1995) demonstrated that social grooming behaviour of the Carniolan honey bee is well developed but not oriented toward the varroa mites. On the other hand, Ruttner and Haenel (1992) reported the presence of active worker fight (aggression) against varroa mites which resulted in damaged mites. Thakur and others (1997) observed the attack of bee workers on the varroa mite situated on the comb cells while varroa mites situated on the bodies of adult bees were of no interest to other bees.
- Removing infested brood from honeybee nest (hygienic behaviour). This behaviour could be well related to the detection of varroa mites in sealed brood cells (Spivak, 1996, Correa Marques, 1998). Hygienic behaviour is extremely well developed in Africanized honey bee in South America.
- Worker and drone brood attractiveness to the female varroa mites could be well related to the number of nurse bee visits (Message and Goncalves, 1995). Drone brood cells are more attractive than worker brood cells (Boot *et al.*, 1995). Attractiveness of the pupae to the varroa mite is the consequence of some esters which are synthesized in honeybee brood immediately before cell capping (Trouiller *et al.*, 1994). Rosenkranz and others (1993) proved the influence of juvenile hormone on the varroa reproduction rate. This is much more expressed in the Apis cerana bees than in the Apis mellifera bees.
- Reproduction rate of varroa mite is reduced (Fuchs, 1994).
- Short post capping period of worker brood is related to reduced reproduction rate of varrroa females (Martin, 1997).
- Other resistant factors, such as smaller reproduction rate of parasitised drones (Cacho *et al.*, 1996), nest temperature regulation of honey bees (Le Conte *et al.*, 1990), and the influence of climate (Garcia – Fernandez, 1995).

The sum of all particular factors of honeybee varroa resistance is reduction of growth of varroa population. Varroa infestation of colonies varies strongly within the same location (with similar environmental conditions) at the same time. The differences between several location in varroa infestation are even larger.

The values of dead varroa mites fallen on the bottom board of 317 hives ranged from 0 to the maximum value of 719. The mean ±SD was 28.47, ± 79.74. The positive value of skewness (+5.20) indicated that the data were clustered more to the left of the mean. The positive value of kurtosis (+31.74) indicated that the distribution of most data was more peaked than it would be true for a normal distribution.

The model 1 is highly significant (P = 0.0001). More than 84.79 % of all variance influence is explained.

Table 1. Significance of ANOVA components of variance included in the model 1. Independent variable is the level of varroa mite infestation

Source of variance	Degree of freedom	F ratio	P value
Apiary	38	29.78	0.0001
Queen rearing location	15	0.95	0.5097
Queen mother influence	30	1.15	0.2779
Error	233		

The effect of the apiary on varroa infestation was highly significant. Two other sources of variance (queen rearing location and queen influence nested within queen rearing location) did not affect varroa infestation significantly. Smaller P value of the queen mother influence than the p value of queen rearing location indicated some significant differences among the least squares means. The queen mother line Kropivšek b/97 expressed a significantly smaller hive varroa population than 44 other queen mother lines. The queen mother line Vozelj 1/97 expressed a significantly smaller hive varroa population than 36 of other queen mother lines. The third mother line Dremelj 60/97 showed an tendency of increased varroa tolerance. We did not detect any colony with significantly higher varroa resistance than the majority of colonies.

The analysis of influence of hive varroa mite population in correlation to other colony characteristics on honey yield (model 2) confirmed a statistically significant relationship between honey yield and predictor variables at 99 % confidence level. The model explained 71.81 % of the variability of the independent variable honey yield.

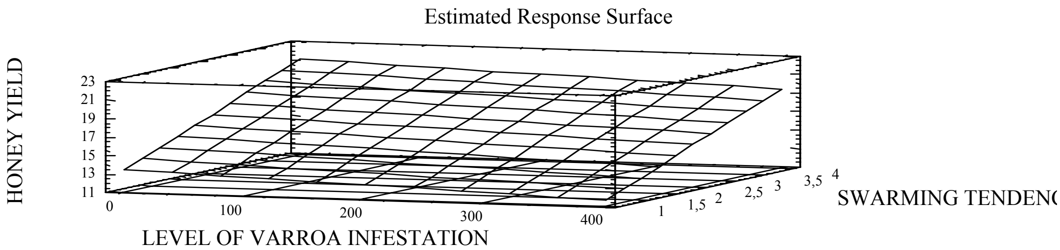
Table 2. Significance of ANOVA components of variance in the model 2. Independent variable is honey yield

Source of variance	Degree of freedom	F ratio	P value
Apiary	35	11.16	0.0000
Origin of queen	14	1.15	0.3146
Level of varroa mite infestation	1	0.06	0.8072
Swarming expression	1	26.80	0.0000
Aggressive behaviour	1	1.32	0.2514
Error	197		

The highest P value in Table 2 belongs to the level of varroa mite infestation. Consequently the component of varroa mite infestation can be excluded from the model. The origin of queen and aggressive behaviour were not significantly correlated to the honey yield. By longer observation an increase of significance can be expected.

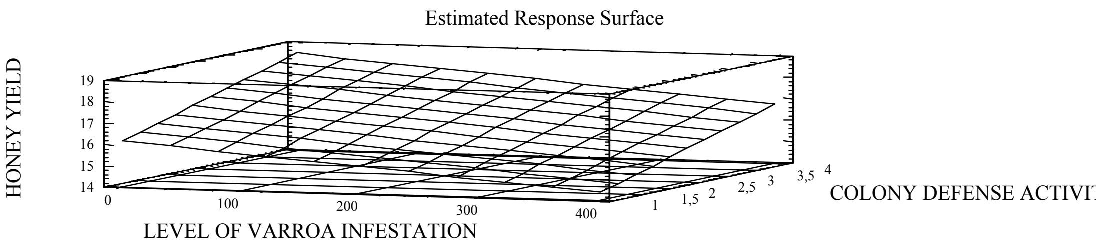
Boigenzahn and Willam (1999) investigated varroa – honey yield relation in a larger population in Austrian honeybee population. They found some phenotypic correlations of + 0.15 (s.e. 0.03) and a negative genetic correlation of -0.23 (s.e. 0.18). A relatively large standard error of genetic correlation indicates that this correlation has not been exactly determined yet. The population in our study is too small to give any relevant results.

Instead of insignificant influence of the level of varroa mite infestation and colony defense activity on honey yield, it is of interest to represent the estimated response surface 3 – d graph between honey yield as dependent variable and all three independent regression variables included in the model 2.



Graph 1. Estimated response surface as interactions between honey yield, the level of varroa mite infestation and swarming tendency.

It should be repeated that the level of varroa mite infestation in model 2 is not a significant component of variance. There could be some evidence of slight correlation between varroa infestation and honey yield. A more infested colony yielded approximately 2 kg less honey. This is evident from Graphs 1 and 2.



Graph 2. Estimated response surface as interactions between honey yield, the level of varroa mite infestation and colony defensive activity.

The correlation between varroa infestation and honey yield should be further investigated. Population of more than 1.000 queens was distributed in summer 1998 to test their production abilities in the season 1999. The group is prepared for hive varroa population test that will be carried out in summer 1999. Experiences gained by the present study should help to make a certain step forward to the varroa resistant bees.

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POVZETEK

Varoza čebel je eden od največjih problemov v čebelarstvu v svetovnem merilu. Slovensko čebelarstvo pesti od leta 1980 dalje. V vmesnem obdobju so imeli čebelarji na razpolago zelo učinkovita zdravila na osnovi piretroidov, vendar so ta postala neučinkovita v zadnjih letih. Uvajanje novih alternativnih načinov preprečevanja varoze čebel vključno z načrtno odbiro čebel s povečano stopnjo odpornosti je vse bolj nujno.

V analizo vpliva izvora matic na številčnost panjske populacije varoze smo vključili skupino 317 matic. Matice so bile zrejene v 16 zrejališčih, po poreklu so izhajale iz 46 mater. Testirali smo jih pri 39 čebelarjih. Čebelje družine smo testirali z enkratno diagnostično aplikacijo 10 ml 85 % mravljinčne kisline na podnici panja. Po 48 urah smo prešteli odpadle varoe. Poskus je potekal v prvi polovici julija 1998. Zbrane podatke smo statistično obdelali. Odpadlo je od 0 do 719 varoj, srednja vrednost je bila 28,47, standardna deviacija 79,74. V nadaljevanju smo izločili statistično visoko značilni vpliv mesta testiranja in določili vpliv izvornega zrejališča in vpliv mater znotraj posameznih izvornih zrejališč. Analizirali smo razlike med posameznimi materami znotraj zrejališč in izločili vse matere, ki so najbolj odstopale od ostalih mater. Pri tem smo izločili dve materi z izjemno nizko stopnjo panjske varoe (Kropivšek b/97 in Vozelj 1/97) in eno mater z nakazano tendenco vpliva na nizko populacijo panjske varoe (Dremelj 69/97). Velikost panjske populacije varoe ni značilno vplivala na letni pridelek medu, nakazana pa je bila tendenca negativne regresije med panjsko populacijo varoe in zmanjšanjem pridelka medu. V najbolj okuženih čebeljih družinah so čebelarji pridelali za okoli 2 kg manj medu kot pa v najmanj okuženih.

REFERENCES

Boigenzahn, C./ Willam, A. Schaetzung von Populationsparametern fuer die Varroatoleranz der Honigbiene (Apis mellifera carnica). Bienewelt 41(1999)1, 3-7.

Boot, W.J./ Schoenmaker J./ Calis J.N.M./ Beetsma J. Invasion of Varroa jacobsoni into drone brood cells of the honey bee, Apis mellifera. Apidologie 26(1995), 109-118.

Božič, J./ Valentinčič, T. Quantitative analysis of social grooming behavior of the honey bee Apis mellifera carnica. Apidologie 26(1995), 141-147.

Cacho, E.D./ Marti, J.I./ Rosa, A./ Quilez, J./ Sanchez-Acedo, C. Effect of Varroa jacobsoni parasitization in the glycoprotein expression on Apis mellifera sprematozoa. Apidologie 27(1996), 87-92.

Correa-Marques, M.H./ De Jong, D. Uncapping of worker bee brood, a component of the higienic behavior of Africanized honey bee against the mite Varroa jacobsoni Oudemans. Apidologie 29(1998), 283-289.

Dietz, A. The geographical distribution and levels of infestation of the mite Varoa jacobsoni Oud. in honey bee colonies in Argentina. Am. Bee J. 126(1986), 49-51.

Erickson, E.H./ Atmowidjojo, A.H./ Hines, L. Can we produce varroa - tolerant honey bees in the United States? American Bee Journal, 138(1998), 828-832.

Fuchs, S. Non reproducing Varroa jacobsoni Oud in honeybee worker cells - status of mites or effect of brood cells? Exp & Appl Acarol 18(1994), 309-317.

Garcia-Fernandez, P./ Rodrigez, R.B./ Orantes-Bermejo, F.J. Ifluence of climate on the evolution of the population dynamics of the Varroa mite on honeybees in the south of Spain. Apidologie, 26(1995), 371-380.

Gregorc. A./ Jelenc, J. Control of Varroa jacobsoni Oud. In the honeybee colonies using Apilife VAR. Zbornik Veterinarske fakultete Univerze v Ljubljani 33 (1996), 2, 255-259.

Harbo, J.R./ Hoopingarner, R.A./ Harris, J.W. Evaluating honey bees for resistance to varroa mites: procedures and results. Am Bee J. 137(1997), 223-224.

Le Conte, Y./ Arnol, G./ Desenfant, P. Influence of the brood temperature and hygrometry varations on the development of the honeybee ectoparasite Varroa jacobsoni. Environ Entomol 19(1990), 1780-1785.

Loper, G.M. A documented loss of feral bees due to mite infestation. Am. Bee J. 135(1995), 823-824.

Martin, S.J. Varroa jacobsoni population biology research in the UK. American Bee Journal, 137(1997), 382-385.

Martin, S.J. A population model for the ectoparasitic mite Varoa jacobsoni in honey bee colonies. Ecological modelling 109(1998), 267-281.

Message, D./ Goncalves, L.S. Effect of size of worker brood cells of Africanized honey bees on infestation and reproduction of the ectoparasitic mite Varroa jacobsoni Oud. Apidologie 26(1985), 381-386.

Rihar, J. Varoza čebel. ČZP Kmečki glas, Ljubljana, 1987, 58 p.

Rosenkranz, P./ Tewarson, N.C./ Rachinsky, A./ Strambi, A./, Strambi, C./ Engels, W. Juvenile hormone titer and reproduction of Varroa jacobsoni in capped brood stages of Apis cerana indica in comparison to Apis mellifera ligustica. Apidologie 24(1993), 375-382.

Ruttner, F. Zuchttechnik und Zuchtauslese bei der Biene - 6. neubearbeitete Auflage. Ehrenwirth Verlag Muenchen, 1988, 146 p.

Ruttner, F./ Haenel, H. Active defense against Varoa mites in a Carniolan strain of honey bee. Apidologie 23(1992), 173-187.

Smirnov, A.E. Research results obtained in USSR concerning aetiology, pathogenesis, epizootiology, diagnosis and control of Varoa disease in bees. Apiacta 13(1978), 149-162.

Spivak, M. Honey bee hygienic behavior and defense against Varoa jacobsoni. Apidologie 27(1996), 245-260.

Thakur, R.A./ Bienefeld, K./ Keller, R. Varroa defense behavior in A mellifera carnica. Am. Bee J. 137(1987), 143-148.

Trouiller, J./ Arnold, G./ Chappe, B./ Le Conte, Y./ Billion, A./ Masson, C. The kairomonal esters attractive to the Varroa jacobsoni mite in the queen brood. Apidologie, (1994)25, 314-321.

Trouiller, J. Monitoring Varroa jacobsoni resistance to pyretroids in western Europe. Apidologie, 29(1998), 537-546.