

Survival of *Varroa jacobsoni* Oud. (Acari: Varroidae) away from
its living host *Apis mellifer* L.

by

L.I. De Guzman, T.E. Rinderer and L.D. Béaman

Reprinted from

experimental & applied acarology

**Purchased by the
United States
Dept. of Agriculture
for official use**

VOLUME 17 No. 4 1993

Science and Technology Letters
PO Box 81, Northwood, Middlesex HA6 3DN, UK

experimental & applied acarology

EDITORS

W. Helle and L.P.S. van der Geest
*Dept. of Pure and Applied Ecology,
Section Population Biology,
University of Amsterdam,
Kruislaan 320,
1098 SM Amsterdam,
The Netherlands*

REVIEW ARTICLE EDITOR

M. Sabelis
Amsterdam, The Netherlands

EDITORIAL ADVISORY BOARD

G. Alberti, *Heidelberg, Germany*
H.M. André, *Tervuren, Belgium*
L.G. Arlian, *Dayton, OH, USA*
D. Ben-Yakir, *Bet Dagan, Israel*
B.A. Croft, *Corvallis, OR, USA*
C.E. Desch, *West Hartford, CT, USA*
U. Gerson, *Rehovot, Israel*
J. Gutierrez, *Montpellier, France*
M.A. Hoy, *Gainesville, FL, USA*
F. Jongejan, *Utrecht, The Netherlands*
O. Kahl, *Berlin, Germany*
W.R. Kaufman, *Edmonton, Alta., Canada*
C.O. Knowles, *Columbia, MO, USA*
W. Knülle, *Berlin, Germany*
G.W. Krantz, *Corvallis, OR, USA*
D. Kropczynska, *Warsaw, Poland*

PRODUCTION EDITOR

S. Nash
*33 St John's Court,
Beaumont Ave,
St Albans, Herts,
AL1 4TS, UK*

E. E. Lindquist, *Ottawa, Ont., Canada*
M. Luxton, *Liverpool, UK*
J.A. McMurtry, *Riverside, CA, USA*
P.W. Murphy, *Lymington, UK*
R.A. Norton, *Syracuse, NY, USA*
R.A.I. Norval, *Gainesville, FL, USA*
W.B. Nutting, *Amherst, MA, USA*
D.R. Penman, *Canterbury, New Zealand*
D.E. Sonenshine, *Norfolk, VA, USA*
A. Takafuji, *Kyoto, Japan*
L.K. Tanigoshi, *Pullman, WA, USA*
G. Uilenberg, *Maisons-Alfort, France*
A. Veerman, *Amsterdam, The Netherlands*
D.L. Wrensch, *Columbus, OH, USA*
M. Wysoki, *Bet Dagan, Israel*

GENERAL INFORMATION

Aims and scope. The journal is concerned with the publication of original scientific papers in the field of experimental and applied acarology. It aims to bring together basic and applied research on various acarine groups, in order that acarologists may more easily keep abreast of developments in related fields. The scope encompasses different aspects of working on agricultural mites, stored-product mites, parasitic mites (ticks, *Varroa*, etc.) and mites of environmental significance. Subject matter dealt with may originate from various disciplines, such as general biology, reproduction, physiology, genetics, evolution and speciation, behaviour, ecology, epidemiology, and all aspects of control, e.g. toxicology and pesticide resistance, immunology, etc. Papers on morphology will be considered when the emphasis is placed on the functional aspects of that morphology. Papers on taxonomy will be considered only in so far as they are relevant to experimental or to control studies.

Submission of articles. Manuscripts should be submitted in triplicate to the Editorial Secretariat, *Experimental & Applied Acarology, Section Population Biology, University of Amsterdam, Kruislaan 320, 1098 SM Amsterdam, The Netherlands.* A detailed guide for authors is available on request.

Publication schedule. *Experimental & Applied Acarology* has 12 issues per volume, volume 17 appearing in 1993.

Subscriptions. Subscription price for 1993 is £350 (US\$512). Send your order to your usual supplier or direct to Science and Technology Letters, P.O. Box 81, Northwood, Middlesex HA6 3DN, UK. Tel. 0923 823586. Fax. 0923 825066.

EXPERIMENTAL & APPLIED ACAROLGY HAS NO PAGE CHARGES

Survival of *Varroa jacobsoni* Oud. (Acari: Varroidae) away from its living host *Apis mellifera* L.

Lilia I. De Guzman^a, Thomas E. Rinderer^b And Lorraine D. Beaman^b

^aDepartment of Entomology, Louisiana Agricultural Experiment Station, Louisiana State University Agricultural Center, Baton Rouge, LA 70803, USA

^bU.S. Department of Agriculture, Agricultural Research Service, Honey-Bee Breeding, Genetics and Physiology Laboratory, 1157 Ben Hur Road, Baton Rouge, LA 70820, USA

ABSTRACT

De Guzman, L.I., Rinderer, T.E. and Beaman, L.D., 1993. Survival of *Varroa jacobsoni* Oud. (Acari: Varroidae) away from its living host *Apis mellifera* L. Exp. Appl. Acarol., 17: 283–290.

Mite survival was measured on eight different substrates: cloth, wax comb, dead drones, dead workers, metal, wood, pollen and control. Trials were conducted at both 26°C and 13°C.

Substrate type, temperature and their interactions significantly affected the survival of *V. jacobsoni*. Longest survival (71 ± 1.1 h) was recorded on dead worker bees (*Apis mellifera* L.) held at 26°C. Shortest survivals were observed on cloth (21 ± 1.3 h), no substrate (21 ± 1.3 h), metal (20 ± 1.1 h) and pollen (18 ± 1.3 h), all maintained at 13°C.

INTRODUCTION

Among the parasitic mites associated with honey bees, *Varroa jacobsoni* Oud. is the most economically serious worldwide. It was first discovered as a parasite of the eastern honey bee (*Apis cerana* F.) in Java, Indonesia (Oudemans, 1904) reportedly having no destructive effect on infested colonies (Koeniger and Koeniger, 1983). With the introduction of the western honey bee (*Apis mellifera* L.) into Asia around 1950, *V. jacobsoni* extended its host range by shifting to the introduced *Apis* species (Crane, 1979). It has subsequently been distributed to all continents except Australia and New Zealand (Griffiths and Bowman, 1981).

In the United States, *V. jacobsoni* was first discovered in Wisconsin in September 1987. By using multivariate morphometric analysis on various samples of the mite from different countries, Delfinado-Baker and Houck (1989) claimed that the North American population was probably introduced from South America. Since its discovery in the USA, 49 states have confirmed infestations. Spread of the mite has probably been enhanced by movement of bee colonies from infested areas for honey production and pollination services (Ruttner, 1983).

Among colonies, drifting is an important mechanism of mite dispersal (Sakofski and Koeniger, 1986). When colonies were too weak to defend themselves, Sakofski (1988, 1990) observed that robbing can introduce an average of 35.4% infestation in an uninfested colony after 24 hours. Robbing of empty hives however, has not been reported as a contributor to *V. jacobsoni* dispersal.

Although *V. jacobsoni* is known to be specific to *Apis* species, live specimens have been observed on wasps robbing honey bee colonies (Gerig, 1987). *V. jacobsoni* is also reported to survive on flowers for 4 days under artificial conditions (Gromyko, 1982) while Hartwig and Jedruszuk (1987) reported 6-day survival of the mite on flowers visited by bees. This ability of the mite to survive away from its host may contribute to its dispersal among colonies. This study was conducted to establish the survival of *V. jacobsoni* on some materials associated with bee-keeping and to determine their potential as agents of dispersal for this mite species.

MATERIALS AND METHODS

This experiment was conducted in Ebro, Florida in March 1991. The substrates used for this experiment were: cloth, wax comb, dead drones, dead worker bees, metal, pollen, wood, and control (no substrate). All substrates were obtained from Louisiana (which did not have mites at that time) to ensure that they were mite-free. Except for pollen and dead bees (*A. mellifera*), the substrates were cut into small pieces ($\sim 5 \times 4$ cm) and placed in insect rearing cups made of cardboard coated with paraffin wax with two pieces per cup. The pieces of metal were cut from used queen excluder while the wood came from a used frame. Both substrates had a little beeswax and propolis on them. For the pollen treatment, one tablespoonful of dried pollen pellets was used per cup. Twenty bees per cup were utilized for the dead bee (drone and worker) treatments. Bees were frozen for storage and thawed before use. Inoculum mites were collected from a highly infested colony by removing capped drone and worker brood from brood combs having varied developmental stages and brushing mites into experimental cups. All adult female mites contained in infested cells were employed in the experiment, with 33 to 71 mites introduced per cup. Thus, inoculum mites used were not of uniform ages.

Inoculated substrates were maintained at 26°C (room temperature) and 13°C (inside a refrigerator with a volume of 1.6 ft³). Room temperature was regulated using the building's central heating and room thermostat and monitored with a thermometer hung on the wall. Likewise, temperature inside the refrigerator was monitored using a thermometer throughout the experimental period. After the experiment, but under similar climatic conditions, temperature and relative humidity readings were determined using a hygrothermograph. The average room temperature was $23 \pm 1.2^\circ\text{C}$ with a relative humidity of $74 \pm 13\%$. Inside the refrigerator, the temperature averaged $9 \pm 0.7^\circ\text{C}$ with $73 \pm 3.6\%$ relative humidity. The discrepancy in temperature readings, especially for the refrigerator, was probably due to the absence of experimental cups, which may have interfered with the flow

of cold air inside the refrigerator. The refrigerator's volume was not sufficient to accommodate both the hygro-thermograph and the rearing cups.

All room temperature treatments were replicated five times except for the dead drone treatment, which had only three replications. All 13°C treatments were replicated three times.

Mite mortality was observed every six hours until the last mite died. At the time of observation, all dead mites were counted and removed from the rearing cups. All mites maintained at 13°C were held at room temperature for ten minutes before dead individuals were counted. Moribund mites or mites that showed leg movements but were not able to crawl when touched with an insect brush were considered dead.

Data were analyzed using ANOVA (factorial experiment with a nested error term) and means were compared using multiple *t*-tests.

RESULTS

Substrates differed significantly ($P < 0.0001$) in their ability to support mite survival irrespective of temperature (Table 1; Figs 1 and 2). Among the eight substrates used, the longest survival period of *V. jacobsoni* was observed on dead workers (Table 1). Temperature likewise displayed a significant ($P < 0.0001$) effect on mite survival. Overall, *V. jacobsoni* survived significantly longer ($P < 0.0001$) at 26°C than at 13°C (Table 1).

Mite survival at 26°C varied on different substrates (Fig. 1). Median mite mortality was 24 hour on both metal and no substrate. However, the last mite survived on these substrates up to 60 and 78 hours post-inoculation, respectively. On wood and wax comb, 50% mite mortality was observed after 29 and 35 hours,

TABLE 1

Average survival (Mean±SE hours) of *Varroa jacobsoni* on eight substrates at two different temperatures.

Substrates	Room temperature (26°C)	Cold temperature (13°C)	Mean
Control	27±0.97 ^g	21±1.29 ^{hi}	26±0.81 ^e
Metal	26±1.00 ^g	20±1.13 ^{hi}	23±0.76 ^e
Wood	32±1.04 ^f	24±1.27 ^{gh}	28±0.82 ^d
Pollen	41±0.96 ^d	18±1.28 ⁱ	33±0.80 ^d
Cloth	48±1.01 ^c	21±1.27 ^{hi}	37±0.81 ^c
Comb	37±1.01 ^e	35±1.31 ^{ef}	36±0.83 ^c
Dead drones	56±1.41 ^b	33±1.35 ^{ef}	44±0.98 ^b
Dead workers	71±1.06 ^a	35±1.39 ^{ef}	58±0.88 ^a
Mean	41±0.38 ^a	25±0.46 ^b	

Means with the same letter (columns and row) do not differ significantly ($P < 0.01$; multiple *t*-tests).

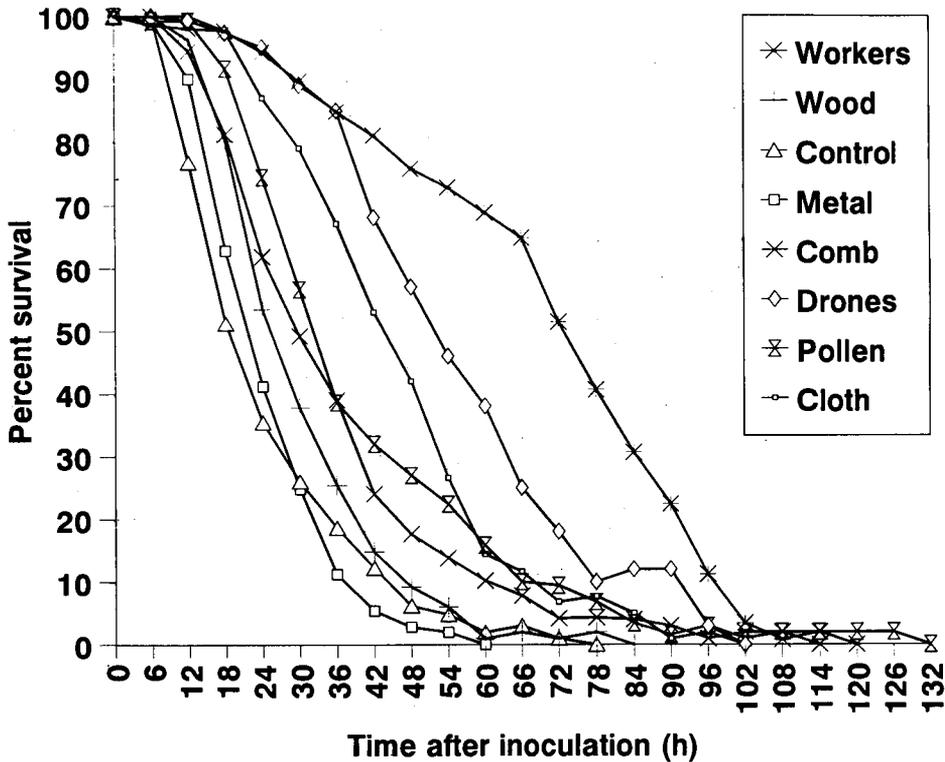


Fig. 1. Survival of *Varroa jacobsoni* on eight substrates at (26°C).

respectively. The death of the last mite was 84 hours post inoculation on wood and 102 hours on wax comb. Median mortality on pollen was 39 hours, and maximum survival was 132 hours. On cloth, 50% mortality occurred at 45 hours while maximum longevity was 102 hours. The longest median mortalities were observed on dead bee treatments; 50% mortality on dead drones was reached earlier (53 hours) than on dead workers (68 hours). Maximum survival on these substrates was 114 and 120 hours, respectively.

At 13°C, the shortest median survival time was on the pollen substrate (15 hours), followed by metal (17 hours)(Fig. 2). No mite survivor was present on this substrate 84 and 48 hours post-inoculation, respectively. On cloth and control, the median survival time was 18 hours. The last survivors lived 60 hours on cloth and 66 hours on control. Median mortality on wood was 21 h with last survivors lived up to 60 hours. Dead drones and wax comb provided 30 and 32 hours median mortality, respectively. Mites on both substrates recorded a maximum survival of 78 hours. Dead workers proved to be the best medium at 13°C with a median mortality of 33 hours and maximum survival of 102 hours.

We also noted some preferences for location by mites on the dead bees. Most

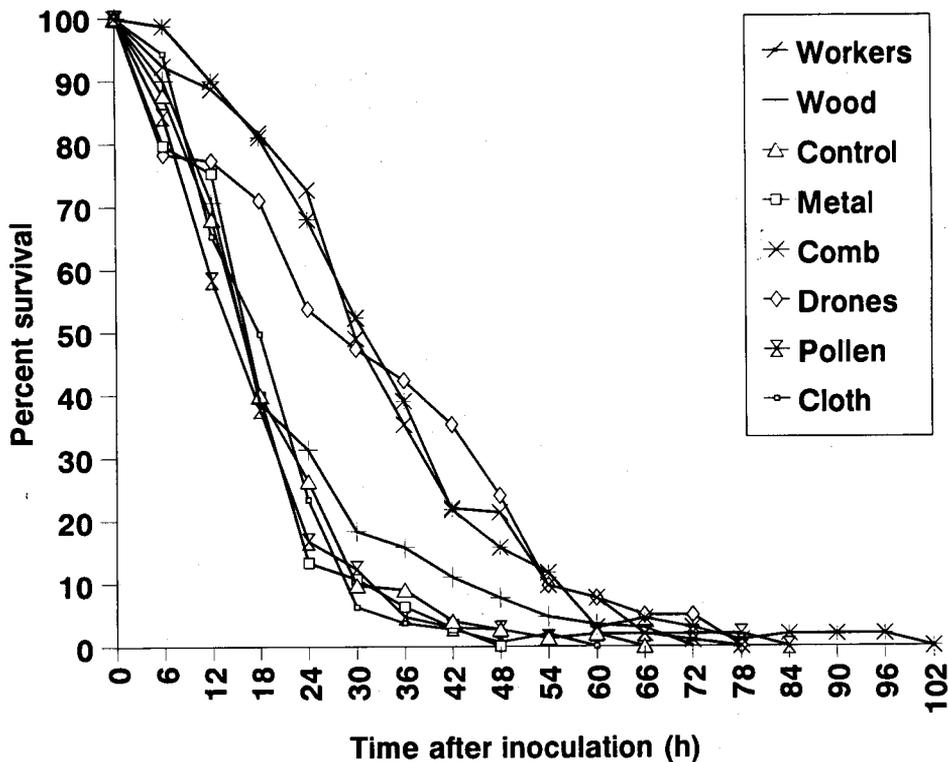


Fig. 2. Survival of *Varroa jacobsoni* on eight substrates at (13°C).

mites were found on the wings, legs, between the thorax and abdomen or between the head and thorax. These locations may have provided protection for the mite from lower temperature since only a few mites were observed on the walls of the container. No mites were found between the intersegmental membranes therefore, feeding may have not occurred.

DISCUSSION

Dead bees were found to be more favourable for the survival of *V. jacobsoni* than any of the other substrates tested. However, dead worker bees supported significantly longer survival than dead drones. The significant survival of *V. jacobsoni* on dead workers over dead drones is probably of biological importance. Workers are the main occupants of the hive while drones are seasonal and few in numbers when present. The present study suggests that dead bees can be good sources of mite contamination, which may happen when bees rob dying or dead colonies. According to Sakofski (1988) robber bees carry dead bees out of the robbed colonies. Further, the author observed a substantial decrease in the infestation rate

of *V. jacobsoni* on dead bees collected outside the hives and later postulated that the transported mites came from the dead bees.

Mean survival of mites on cloth and wax comb were not significantly different. Both substrates may have the ability to provide protection for the mites. These observations support the view that bee suits used in manipulating *Varroa*-infested colonies and empty combs removed from infested colonies are not safe for immediate use in mite-free colonies.

Wood provided survival comparable with that of pollen. On wood, mite survival was probably due to the presence of some beeswax and the mites' ability to hide in cracks for protection from dehydration. This observation suggests that *V. jacobsoni* can live on bottom boards, frames and supers for up to a few days. The reason for the relatively long survival of mites on pollen was most likely due to the ability of the mites to hide under or between the pollen pellets to prevent dehydration. Hence, using fresh pollen from pollen traps and empty combs with pollen may also spread *V. jacobsoni*.

Shortest survivals were recorded on metal and in the control which may reflect the inability of either situation to provide protection for the mites. Nonetheless, some mites survived up to 3 days under these conditions. Mite survival may be increased, however, with the presence of beeswax/burr combs on the metal substrate and in the control.

The small reduction in the ability of the mites to survive on wax combs kept at low temperature may indicate that cells can provide insulation for the mites. Most of the inoculated mites were seen inside the cells throughout the experimental period, where they may have experienced limited dehydration. For both temperature treatments, the longest survival of mites was on the dead worker substrate.

V. jacobsoni has a strong attraction to a heat source and has the ability to discriminate a temperature difference of 1.2°C (Le Conte and Arnold, 1987). This attraction reinforces the idea that the mite has an optimum temperature for survival. Reduced mite survival at 13°C may indicate that mites cannot tolerate this temperature well. According to Pätzold and Ritter (1989), the tolerable temperature for mites collected during winter ranges from 16–43°C, which is higher than the temperature in the refrigerator we used. Mites collected in summer can withstand temperatures ranging from 24–43°C with a preferred temperature range of 25–40°C. Further, these authors reported that exposure of mites above or below these ranges significantly impaired their mobility. Their observation may explain the increased survival of the mites at room temperature.

V. jacobsoni can evidently survive away from a living host for several hours under many conditions any time of the year. Engels (1992, personal communication) claimed that *V. jacobsoni* can survive 5–10 days without food intake when provided with high humidity and temperatures between 20–25°C. All of the substrates that we tested were potential dispersal agents for *V. jacobsoni* if they were transferred and used in uninfested locations, apiaries or colonies. Foraging bees can acquire *V. jacobsoni*, especially when drifting and robbing (Sakofski, 1988 and

1990; Sakofski and Koeniger, 1986; Ruttner, 1983; Rademacher, 1991). Since empty beekeeping equipment may also be subjected to robbing, it must be considered a potential source of *V. jacobsoni* contamination for uninfested colonies.

RECOMMENDATIONS

With the observed ability of *V. jacobsoni* to survive 18-71 h on the tested substrates, we recommend the following precautionary measures to slow the spread of the mite from location to location, apiary to apiary or colony to colony:

(1) Tools, gloves and bee suits used in examining mite-infested colonies should be washed or stored for at least two weeks before using with uninfested bees.

(2) Trucks used in hauling infested colonies should not be used immediately for hauling uninfested colonies without cleaning. Dead bees should be removed since they can still have living mites on them. The presence of honey on the trucks will stimulate robbing, which may enhance the contact of uninfested robber bees with dead infested bees.

(3) Supers, queen excluders, and empty frames with or without pollen that were recently removed from infested hives should be stored at least two weeks prior to using them in clean hives.

ACKNOWLEDGMENT

We thank Dr. Steve Buco of Statistical Resources, Inc. for data analyses. This paper has been approved for publication by the Director of the Louisiana Agricultural Experiment Station as manuscript number 92-17-6275.

REFERENCES

- Crane, E., 1979. Fresh notes on the *Varroa* mite. *Bee World*, 60:8.
- Delfinado-Baker M. and Houck, M. A., 1989. Geographic variation in *Varroa jacobsoni* (Acari, Varroidae): Application of multivariate morphometric techniques. *Apidologie*, 20:345-358.
- Engels, W., 1992. Personal communication.
- Gerig, L., 1987. Vespen als Varroaträgerinnen. *Schweizerische Bienen-Zeitung*, 110: 341-345.
- Griffiths, D. A. and Bowman, C. E., 1981. World distribution of the *Varroa jacobsoni*, a parasite of honey bees. *Bee World*, 62:154-163.
- Gromyko, G. I., 1982. Vyzhyvayemost' samok *Varroa* vnie pchelinoi semii. *Pchelevodstvo*, 5: 16-17.
- Hartwig, A. and Jedruszuk, V. S. A., 1987. Survival of *Varroa jacobsoni* on flowers. Proceedings of the XXXI International Congress on Apiculture, Warsaw, Poland, pp 229-233.
- Koeniger, N. and Koeniger G., 1983. Observations on mites of the Asian honeybee species (*Apis cerana*, *Apis dorsata*, *Apis florea*). *Apidologie*, 14(13): 197-204.
- Le Conte, Y. and Arnold, G., 1987. Influence de l'age des abeilles et de la chaleur sur le comportement de *Varroa jacobsoni*. *Apidologie*, 18: 305-320.
- Oudemans, A. C., 1904. On a new genus and species of parasitic acari. *Notes Leyden Mus.*, 24(8):216-222.
- Pätzold, S. and Ritter, W., 1989. Studies on the behaviour of the honey-bee mite, *Varroa jacobsoni* O., in a temperature gradient. *J. Appl. Ent.*, 107:46-51.
- Rademacher, E., 1991. How *Varroa* mites spread. *Am. Bee J.*, 131: 763-765.

- Ruttner, F., 1983. Varroaosis in honeybees: extent of infestation and effects. In: *Varroa jacobsoni* Oud. Affecting Status and Needs, Proc. Meet. EC Expert Group. R. Cavalloro (ed.). pp 7-13. Wageningen, Balkema, Rotterdam.
- Sakofski, F., 1988. Transfer of *Varroa jacobsoni* by robbing. In: Present Status of Varroaosis in Europe and Progress in the *Varroa* Mite Control, Proc. Meet. EC Expert Group. R. Cavalloro (ed.) pp. 177-181. Udine, Italy.
- Sakofski, F., 1990. Quantitative investigations on transfer of *Varroa jacobsoni* Oud. In: Proc. of the Int. Symposium on Recent Res. on Bee Path. W. Ritter (ed.) pp 70-72. Gent, Belgium.
- Sakofski, F. and Koeniger, N., 1986. Natural transfer of *Varroa jacobsoni* among honey bee colonies in autumn. In: European Research on Varroaosis, Proc. Meet. EC Expert Group. R. Cavalloro (ed.) pp. 81-84. Bad Homburg, Balkema, Rotterdam.