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New observations on camphor – an old insect repellent – as a relatively safe candidate fumigant against nine insect species

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Exposure of the adults of *Musca domestica* (L.), *Plodia interpunctella* (HB.), *Ephestia kühniella* ZELL., *Galleria mellonella* (L.), *Cydia pomonella* (L.), *Pieris brassicae* (L.), *Rhizopertha dominica* (F.), *Tenebrio molitor* L., and *Acanthoscelides obtectus* (SAY) to different concentrations of camphor in air-tight jars resulted, at the highest concentration of 40 mg/l, in 100% knock-down of the 9 insect species within 3 h or less, e. g. 15 min in the case of *P. interpunctella*. The latter and the other pyralide moths were also completely knocked down with the lowest concentration of 10 mg/l. A 6 h exposure of the insects to the highest concentration of camphor, followed by an overexposure to the normal atmosphere for 1 to 4 days, killed 100% of the insects within the overexposure time. 100% mortality of the pyralide moths was also achieved with the lowest concentration of camphor (in the case of *P. interpunctella* within 24 h).

When post-war agriculture became more and more industrialized, the fast growing demand for insecticides could no longer be satisfied by the production of natural insecticides, such as nicotine, rhyania, rotenone, pyrethrum, and camphor. Research for insecticides and insect repellents therefore concentrated on synthetic substances promising either higher insecticidal or repellent activity or – in comparison to nicotine – safer handling. Today a trend towards more biological methods in agriculture can be observed. Natural substances are considered to be integral parts of the biosphere and therefore less disruptive to the ecological balance. Furthermore, natural substances used in traditional medicine have a long record of safety for human beings. One of the candidate substances is camphor. It has a long tradition as a repellent against household insects, especially the clothes moths, and some of the older records also point out its potential for killing insects (SCOTT *et. al.*, 1918; PRATT *et. al.*, 1933; STÄGER, 1933; BROWN, 1951; GUNTHER & JEPSON, 1960). The physicochemical and medicinal properties of camphor have been described by ABIVARDI & ZAREH (1971); they shall not be treated here.

Historical review

DÖRFFURT (1793), while explaining the history, purification, and behaviour of camphor, also described some methods of its application. According to this author, spiders, scorpions, flies, wasps, fleas, mosquitoes, bugs, lice, ants, and caterpillars as well as beetles and their larvae, damaging wheat kernels and other seeds, were killed when exposed to camphor. DÖRFFURT also recommended using it, instead of heat, for killing the silkworm pupae inside the cocoons. Experimenting all the year round he found mortality caused by camphor to occur faster in summer, i. e. at high temperatures. He also observed that the clothes moths,

although repelled by the scent of camphor, were resistant to the insecticidal action of the substance.

Throughout the nineteenth century, camphor – besides tobacco, pyrethrum, derris, hellebore, quassia and turpentine – was one of the most important natural products used as insecticide and/or repellent. According to BROWN (1951) insecticidal joss sticks were prepared by dipping one end of the sticks into plant gums containing camphor or pyrethrum. Up to 1925 various oils and other natural products, including alcoholic solutions of camphor, were the only generally used repellents against biting flies, gnats, and mosquitoes (BUNKER & HIRSCHFELDER, 1925; GUNTHER & JEPPSON, 1960).

The first systematic research on camphor was conducted by SCOTT *et al.* (1918) who got 80–100% control of the larvae and adults of the dermestide beetles *Attagenus piceus* (OLIV.) and *Anthrenus scrophulariae* L. within one month at a concentration of about 0.75 g camphor per liter of trunk capacity. Even though this result could not be considered very positive, PRATT *et al.* (1933) started once more systematic research on the action of camphor against the convergent lady-beetle, *Hypodamia convergens* GUER, and found its toxicity to be higher than that of chloropicrine and nicotine. Camphor had also an auxiliary effect on HCN to kill scale insects. Similarly, STÄGER (1933) observed 100% mortality of ants exposed to camphor for 3.5 h. However, these systematic investigation coincided with the development of the more potent synthetic insecticides and repellents. In 1932 the fumigant methyl bromide was patented, in 1936 and 1938 the repellents dihydropyrone and dimethyl-phtalate respectively, in 1940 DDT, and in 1942 HCH. The great success of DDT against the vectors of typhoid fever and malaria during the Second World War and the development of highly effective synthetic repellents against blood sucking insects during this period as well as the needs of modern agriculture led to a concentration of research on synthetic insecticides and repellents as mentioned before, whereas research on natural insecticides and repellents was almost completely abandoned. This is also true for camphor, though a chlorinated product of camphor was developed to the well known insecticide Toxaphene, patented in 1945 (BROWN, 1951; STETTER, 1977).

The revival of the use of camphor seemed to be possible when olfactometer tests by ARNOLD (1957) revealed the high repellency of camphor against *Attagenus piceus* larvae at a concentration of 0.64 mg/l of air, and WOLCOTT (1957) as well as KASSAB *et al.* (1960) reported on the resistance of the camphor tree to four species of termites. However, camphor showed almost no repellency when field-tested or investigated by split-arena tests (CHAMBERLAIN, 1957).

Recent investigations

Considering the safety of camphor for man as reflected by its oral and dermal application in modern medicine, especially the use of preparations containing up to 50% camphor for dermal treatments, and the great needs for finding safe pesticides, ABIVARDI & ZAREH (1971) took up research on camphor once more. They have shown that camphor may completely control *Callosobruchus chinensis* at a concentration as low as 12 g/m³ of stored dried beans without changing the odour, taste, baking quality or germination rate of the treated seeds (ZAREH, 1973; ABIVARDI, 1978). Camphor also highly reduces the rate of oviposition (ABIVARDI & ZAREH, 1971; ABIVARDI & RAHIMIAN, 1977) and completely inhibits the embryonic and postembryonic development of this insect (ABIVARDI, 1977). Further-

more, camphor causes 100% mortality of some other insects (ABIVARDI, 1974, 1976) and has a fungicidal and/or fungistatic effect on seven important soil-born fungi (ABIVARDI, 1979).

Considering the promising results of these studies, and in order to find out more about the spectrum of the insecticidal activity of camphor, further experiments were carried out with a relatively wide range of economically important insect species, belonging to 3 orders, 7 families, and 9 genera. This paper reports on the results with the adults of these insects.

MATERIAL AND METHODS

Insects

The insecticidal activity of camphor was evaluated against the adults of the house fly, *Musca domestica* L. (Dipt.: Muscidae), the Indian meal moth, *Plodia interpunctella* (HB.) (Lep.: Pyralidae), the tobacco moth, *Ephestia kühniella* ZELL. (Lep.: Pyralidae), the greater wax moth, *Galleria mellonella* (L.) (Lep.: Pyralidae), the codling moth, *Cydia pomonella* (L.) (Lep.: Tortricidae), the large cabbage white, *Pieris brassicae* (L.) (Lep.: Pieridae), the lesser grain borer, *Rhizopertha dominica* (F.) (Col.: Bostrychidae), the yellow meal-worm, *Tenebrio molitor* (F.) (Col.: Tenebrionidae), and the common bean weevil, *Acanthoscelides obtectus* (SAY) (Col.: Bruchidae), obtained from laboratory colonies. All insects used in these experiments were unsexed adults and, with the exception of *T. molitor*, were 1–3 days old. *Tenebrio* adults were about one month old.

Bioassay

The insects, except the coleoptera, were first anaesthetized with CO₂. Ten insects per unit (in the experiment with *R. dominica*, infested wheat kernels, each containing an adult insect) were laid on the bottom of 500 ml air-tight glass jars. The insects were then exposed to filter paper discs (Ø 55 mm) impregnated with 0, 5, 10 or 20 mg of pure camphor, corresponding to a concentration of 0, 10, 20 or 40 mg/liter.

In order to prepare the filter discs, camphor crystals (d-camphor of Siegfried Company, Zofingen, Switzerland) were first dissolved in pure acetone and diluted to the concentrations of 1 : 5, 1 : 10, and 1 : 20 (wt./vol.). Each filter paper disc was treated with 0.1 ml of pure acetone or one of the above-mentioned solutions under a hood at 22°C and then transferred to one of the jars with insects, after the solvent had been allowed to evaporate for 3 min. The air-tight lid of the jar was then closed and the jar placed in a rearing chamber of 25 ± 1 °C and 60% RH.

The knock down rates were recorded at 15, 30, 60, 120 and 180 min after treatment. After a 6 h exposure to the camphor vapours, the insects of each replicate were transferred to a one liter recovery glass jar covered with a fine screen (0.5 mm mesh width) and kept at 25 °C and 60% RH. The insects were thus overexposed to the normal atmosphere for 1 to 4 days and also provided with appropriate food. During the overexposure period the stored-product insects were kept in the dark, while the other insects were subjected to a 16L : 8D light regime. The experiments were conducted with a completely randomized design and replicated 4 times.

RESULTS

Table 1 summarizes the effects of three concentrations of camphor on the rates of knock-down of the adults of 9 insects, after a 15-180 min exposure to the

Table 1: Knock-down effect of different concentrations of camphor on the adults of nine insect species after different exposure times in air-tight jars.

Insect species and concentrations of camphor (mg/l)	Knock-down ^a in % after				
	15 min	30 min	60 min	120 min	180 min
<u>M. domestica</u>					
10	0	25	50	73	73
20	25	80	85	100	-
40	60	85	95	100	-
<u>P. interpunctella</u>					
10	0	20	100	-	-
20	45	65	100	-	-
40	100	-	-	-	-
<u>E. kühniella</u>					
10	25	50	60	80	100
20	25	50	60	87	100
40	25	50	70	100	-
<u>G. mellonella</u> ^b					
10	0	0	0	80	100
20	0	0	0	100	-
40	0	0	0	100	-
<u>C. pomonella</u>					
10	0	0	0	58	58
20	0	0	45	70	100
40	0	0	70	93	100
<u>P. brassicae</u>					
10	0	20	30	40	55
20	0	35	53	100	-
40	40	47	75	100	-
<u>R. dominica</u>					
10	0	0	0	0	0
20	0	0	0	0	20
40	0	0	20	48	100
<u>T. molitor</u>					
40	- ^c	-	-	-	100
<u>A. obtectus</u>					
40	-	-	-	-	100

a Means of 4 replicates (10 insects/replicate). No knock-down was observed in controls.

b Highly stimulated for at least one hour, so that no records on knock-down could be made.

c Not recorded.

substance in air-tight jars at 25 °C. All of the insects were completely knocked down when exposed to camphor at concentrations of 20 or more mg/l for a maximum of 3 h. The treated insects experienced three distinct phases, which may be

Table 2: Effect of three concentrations of camphor on the mortality of the adults of nine insect species after a 6 h exposure to the substance in air-tight jars, followed by exposure to the normal atmosphere for 1-4 days (d).

Insect species and concentrations of camphor (mg/l)	% mortality ^a after		
	1 d	2 d	4 d
<u>M. domestica</u>			
10	80	- ^b	-
20	95	-	-
40	100	-	-
<u>P. interpunctella</u>			
10	100	-	-
20	100	-	-
40	100	-	-
<u>E. kühniella</u>			
10	64	-	80
20	75	-	100
40	90	-	100
<u>G. mellonella</u>			
10	30	35	52
20	50	70	75
40	50	84	100
<u>C. pomonella</u>			
10	0	-	40
20	57	-	93
40	90	-	100
<u>P. brassicae</u>			
10	80	-	-
20	100	-	-
40	100	-	-
<u>R. dominica</u>			
10	0	0	10
20	55	60	95
40	100	-	-
<u>T. molitor</u>			
40	-	-	100
<u>A. obtectus</u>			
40	-	100	-

a Means of 4 replicates (10 insects/replicate). Less than 2% mortality was observed in controls.

b Not recorded.

respectively named stimulation, disorientation, and agitation phase before knock-down. In all treatments camphor first stimulated the activity of the insects, reflected by rapid climbing the walls of the jars by the house flies and moths, leaving the wheat kernels by *R. dominica*, and higher rates of movement in the remaining insects. Then the flies and the moths stopped climbing, *P. brassicae* lost the resting position, and the beetles made disoriented movements. Finally the insects were highly agitated, and the flies and the Lepidoptera started a strong vibration of the wings so that some of the legs were detached from the insects during this period. This phase was eventually terminated in a complete knock-down, recognized by a stationary position. The agitation phase was clearly observed in the house flies and the Lepidoptera and lasted longer than an hour in *G. mellonella*, so that no data on the rates of knock-down of this insect could be recorded in this period (Tab. 1).

The lowest concentration of camphor, 10 mg/l, also resulted in 100% knock-down of *P. interpunctella*, *E. kühniella*, and *G. mellonella* after respectively 1, 2, and 3 h exposure, showing a very high sensitivity of the three members of the family Pyralidae to camphor. The differences in the required exposure time for a complete knock-down of these insects, may partly be due to the distinct differences in their size (see below).

Table 2 reports the effects of camphor on the mortality of different insects, after a 6 h exposure in air-tight jars, followed by overexposure to the atmosphere for 1, 2, or 4 days, at 25 °C and 60% RH. All of the insects exposed to the highest concentration of camphor (40 mg/l) were killed after a maximum of 4 days aeration, while no knock-down and less than 2% mortality were observed during the 6 h exposure period in the controls. The same degree of control was achieved already at the lower concentration of 20 mg/l in *E. kühniella* and *P. brassicae*, and of 10 mg/l in *P. interpunctella*. The latter species suffered already 100% mortality within 1 day after exposure to the lowest concentration of camphor. The effects of camphor were not reversed during the periods of overexposure. The effective concentrations for killing 95–100% of six stored-product insects, after a 6 h exposure to camphor are summarized in Fig. 1.

DISCUSSION

Whereas the high concentrations of camphor used in human medicine (NEUGEBAUER & MORANT, 1983) show the safety of camphor for man, the effective concentrations for killing insects reveal its strong insecticidal activity. The adults of all nine insect species tested were killed already after a short period of exposure to the substance. Thus, although the effective concentrations are acceptable, they may be further reduced by increasing the exposure time.

On the other hand, due to the fumigant action of camphor, the effective control of *M. domestica*, *C. pomonella*, and *P. brassicae* in our tests is not believed to be of practical value. However, the high sensitivity of these and the other insects to camphor reveals the broad spectrum of its insecticidal action.

It is evident that the fumigant action of camphor renders this substance especially suitable for the control of stored-product insects. Long-term studies on such insects, including species not killed by camphor after short exposure times, are therefore required to evaluate its practical value. According to ADHAOO (1980) camphor is traditionally used in India to keep insects from stored products. The most effective method of application seems to consist in burning the camphor.

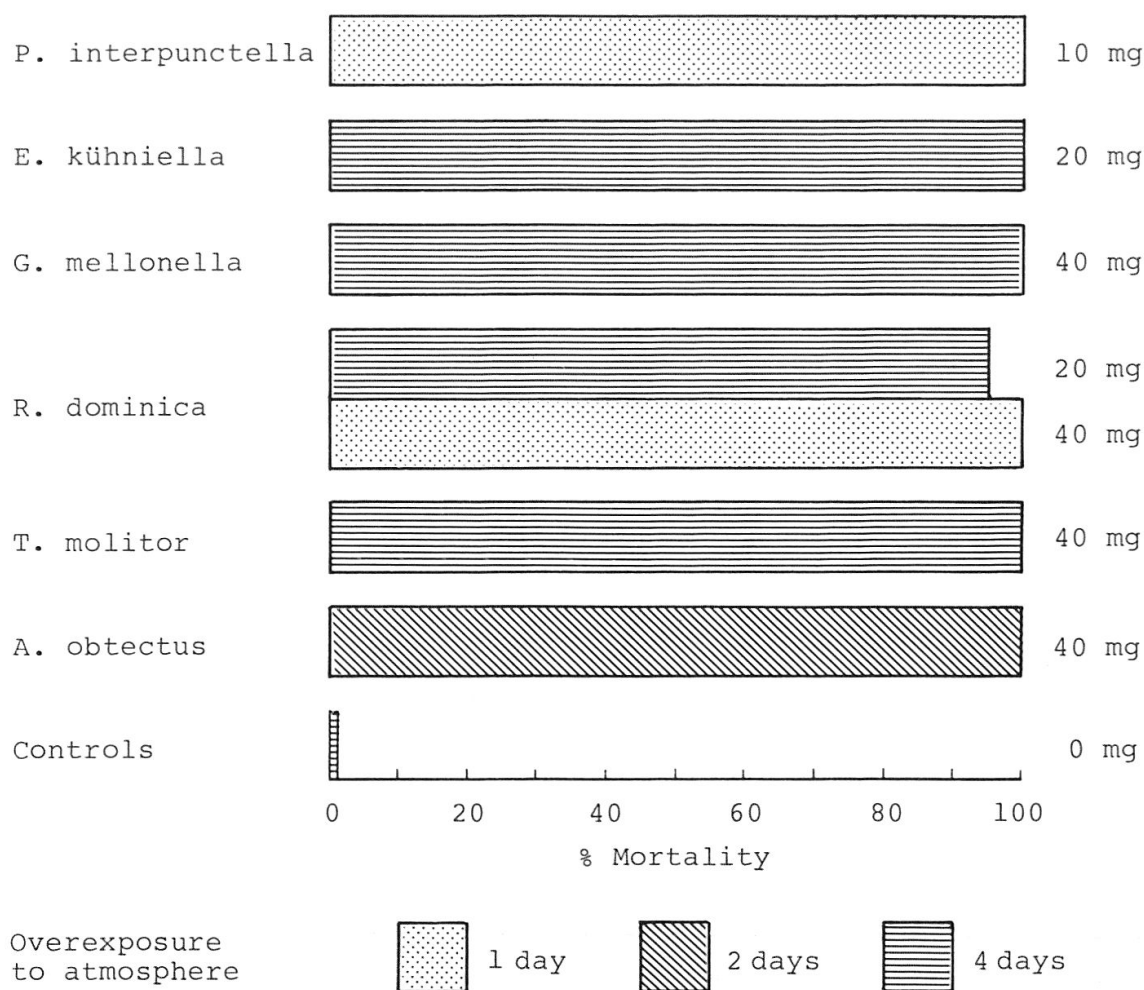


Fig. 1: Effective concentrations (mg/l) for 95–100% mortality of six stored-product insects, assessed after a 6 h exposure of the adults to camphor in air-tight jars, followed by overexposure to the normal atmosphere for 1 to 4 days. Only one concentration has been tested with *T. molitor* and *A. obtectus* (s. Tab. 2).

As mentioned before, the differences in the exposure times required for the complete knock-down of the three pyralide moths reported in this paper may be due to the differences in the size of the three species, the large *G. mellonella* being the least susceptible. A similar correlation of camphor sensitivity to insect weight has also been observed in four members of the beetle family Bruchidae (ABIVARDI, 1976). However, the high resistance to camphor of the small *Tribolium confusum* DU VAL (ABIVARDI, 1976) and the susceptibility of the large *Tenebrio molitor*, both within the family Tenebrionidae, as well as the low sensitivity of some small coleoptera (*A. obtecta* and *R. dominica*) and the relatively high susceptibility of the larger Lepidoptera reported in this paper, reveal that sensitivity to camphor is not only governed by weight and/or family relationship.

Although plant produced insecticides have been developed in the course of evolution for the protection of plants, there has always been a coevolution of plants and herbivores, allowing the survival of both. Insects may thus adapt to plant constituents. Increased resistance of insect populations as a result of selection may therefore develop to natural as well as to synthetic insecticides. In our experiments the five lepidopterous species (belonging to three families) were

easily killed by camphor – probably because they were never exposed to this plant substance in their natural environment and therefore have not developed resistance mechanisms. This seems not to be true for the clothes moth, *Tineola bisselliella* (HUMMEL) (Lep.: Tineidae), which, according to DÖRFFURT (1793) was resistant to the insecticidal action of camphor. However, since already at DÖRFFURT's time camphor was excessively used against clothes moths, their resistance may have resulted from selection, rather than represent a specific character of the species.

Because of its low human toxicity, camphor may be regarded as a desirable substitute for synthetic pesticides used for the protection of stored products. However, it must be kept in mind that the production of natural substances is much more limited than that of synthetic substances. Since the basic camphene used for the synthesis of toxaphene is an isomerization product of α -pinene from turpentine oil (STETTER, 1977), this may also be a way for increasing the camphor production.

ZUSAMMENFASSUNG

Adulte Hausfliegen (*Musca domestica* L.), Dörrobstmotten (*Plodia interpunctella* HB.), Kakaomotten (*Ephestia kühniella* ZELL.), Grosse Wachsmotten (*Galleria mellonella* L.), Kohlweisslinge (*Pieris brassicae* L.), Getreidekapuzinerkäfer (*Rhizopertha dominica* F.), Mehlkäfer (*Tenebrio molitor* L.) und Speisebohnenkäfer (*Acanthoscelides obtectus* SAY) wurden in luftdichten Gläsern bei einer Temperatur von 25 °C während 6 h Konzentrationen von 0, 10, 20 oder 40 mg Kampfer pro Liter Luft ausgesetzt und anschliessend während 1–4 Tagen in gelüfteten Gläsern bei 25 °C und 60% relativer Feuchtigkeit gehalten. Alle Insektenarten wurden innerhalb von 3 h immobilisiert, wenn sie 20 oder mehr mg/l Kampfer ausgesetzt wurden (bei den Pyraliden genügten sogar schon 10 mg/l). Spätestens nach 4 Tagen waren alle während 6 h der höchsten Konzentration ausgesetzten Insekten gestorben (bei den Pyraliden genügte dafür auch die geringste Konz.).

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