

# The influence of temperature on the sensitivity of two nitidulid beetles to low oxygen concentrations

J. E. Donahaye, S. Navarro and M. Rindner\*

## Abstract

In laboratory experiments, larval, pupal, and adult stages of the nitidulid beetles *Carpophilus hemipterus* (L.) and *Urophorus humeralis* (F.) were exposed to simulated burner-gas concentrations at three temperatures of 26°, 30°, and 35°C. The gas concentrations were: 1%O<sub>2</sub>, 85%N<sub>2</sub>, 14%CO<sub>2</sub>; 2%O<sub>2</sub>, 84.7%N<sub>2</sub>, 13.3%CO<sub>2</sub>; 3%O<sub>2</sub>, 85%N<sub>2</sub>, 12%CO<sub>2</sub>—all at 75% relative humidity. For all insects submitted to the modified atmosphere (MA) containing 3%O<sub>2</sub> at 26°C, exposure time to produce 95% mortality was 196 hours. To obtain the same mortality level with the MA containing 1%O<sub>2</sub> at 35°C 60 hours was required. Comparison of exposure times required to produce 50% kill (LT<sub>50</sub>) showed that the effect of temperature on treatment efficacy was most pronounced at the 1% O<sub>2</sub> level where for the three stages of both species tested, values of LT<sub>50</sub> at 26°C were about half those at 35°C. However, at 3%O<sub>2</sub> and 35°C, LT<sub>50</sub> levels were only marginally reduced.

## Introduction

The problems of field infestations of nitidulid beetles in dates at harvest time in Israel have been addressed in previous investigations (Donahaye and Navarro 1989; Donahaye et al. 1991 a,b). For commercial scale fumigations, the addition of 20% carbon dioxide (CO<sub>2</sub>) in air to methyl bromide (MB) has enabled recommended MB dosage rates to be reduced to 12 g/m<sup>3</sup> and reduction in exposure time from 16 to 6 hours (Navarro et al. 1989). However, decisions have already been made by some regulatory organisations to phase out the use of MB as a fumigant due to its involvement in depletion of the stratospheric ozone (USEPA 1993).

A further incentive for the introduction of non-toxic chemical control procedures into the date industry has been for the treatment of organically grown dates using MAs. At present we are experimenting with treatments in a fumigation chamber of 36.1 m<sup>3</sup> capacity attached to a controlled atmosphere generator using a catalytic converter running on butane or propane. This produces in-chamber concentrations of 1–3% oxygen (O<sub>2</sub>) and 12–14% CO<sub>2</sub> within 6 hours. It is highly effective in causing insect emigration, but as a control treatment it is slow, and if quick turnaround is required, short exposure times may not be sufficient to produce complete mortality of residual infestations.

Recent studies (Soderstrom et al. 1992) have shown that the sensitivity of *Tribolium castaneum* (Herbst) to hypoxia and hypercarbia is strongly influenced by temperature. This laboratory study was undertaken to investigate whether treatments of the burner-gas concentrations could be applied as an alter-

native to the present MB/CO<sub>2</sub> fumigations, by optimising temperatures to obtain complete kill within an acceptably short exposure period.

## Materials and methods

### Modified atmospheres

Three combinations of atmospheric gases were chosen to cover the range of atmospheres frequently obtained in the burner-gas treatment chamber, namely: 1%O<sub>2</sub>, 85%N<sub>2</sub>, 14%CO<sub>2</sub>; 2%O<sub>2</sub>, 84.7%N<sub>2</sub>, 13.3%CO<sub>2</sub>; 3%O<sub>2</sub>, 85%N<sub>2</sub>, 12%CO<sub>2</sub>. These compositions were obtained from supply cylinders of O<sub>2</sub>, N<sub>2</sub> and CO<sub>2</sub> using a gas-mixing apparatus described by Donahaye (1992). This consisted of component gases supplied in tubing at rates regulated by a series of valves and gas-flow meters, that enabled the components to be mixed in the desired combinations. After the gas supplies converged, gas in the common supply-line was led to temperature controlled incubators, adjusted to the desired temperature, and passed through a wash-bottle containing sulphuric acid to obtain a constant relative humidity (r.h.) of 75%. Finally the gas mixtures were delivered via a distribution chamber to a series of 100 mL Erlenmeyer flasks that served as exposure chambers arranged in-parallel.

### Temperatures

The three exposure temperatures of 26° 30° and 35°C were chosen as being within the proven feasible range for burner-gas treatments in the chamber. The ambient temperatures during the harvest season were in the lower range, and when the cooling system was not operated, the heated gases from the exothermic converter contributed to heating of the chamber at a rate of about 1°C per hour. Any additional heat required to raise the temperature to 35°C could be supplied from an electric heater installed on the chamber wall.

### Insects

The beetles *Carpophilus hemipterus* (L.) and *Urophorus humeralis* (F.) served as test insects. Larval, pupal and adult stages were obtained from cultures reared at 26°C and 70% r.h. on a synthetic food medium (Donahaye and Navarro, 1989). Larvae were taken from culture jars 7 days after egg hatch. Pupae were exposed to the treatments 1–2 days after pupation, and were obtained by daily removal of pupae from culture jars. Newly emerged adults were collected daily and held on culture medium for 7 days before exposure to treatments.

### Experimental procedure

Six groups of 30–50 insects were placed in exposure flasks together with approximately 2 g synthetic food medium, and linked to the gas mixture apparatus. An additional flask served as control. Periodic removal of flasks was based on preliminary trials to cover the time ranges over which insect mortality

\* Department of Stored Products, Agricultural Research Organization, P.O. Box 6, Bet Dagan, 50250 Israel.

was found to occur for each species, stage, gas-mixture, and temperature. Due to heterogeneity of response, each set of exposures was repeated five times. At the end of each exposure time a flask was removed from the apparatus and held in a constant temperature room at  $30 \pm 1^\circ\text{C}$  and  $60 \pm 5\%$  r.h. Mortality was determined after 10 days, with larvae that failed to pupate and pupae that failed to reach adult emergence being considered as dead. Experimental results were subjected to probit analysis using a program written by Daum (1979).

## Results

The exposure times required to produce 95% mortality ( $LT_{95}$ ) for *C. hemipterus* and *U. humeralis* are given in Figures 1 and 2. Values missing in the larval stages of both species are

because mortality was prolonged, and heterogeneity of response was so great that significant regression could not be obtained.

From both figures it can be seen that the effect of temperature was most pronounced at the 1%  $O_2$  and 2%  $O_2$  levels, where in most cases, at  $35^\circ\text{C}$ , less than half the time was required to produce 95% mortality than at  $26^\circ\text{C}$ . At the 3%  $O_2$  level, adult mortalities of both species were hardly affected by temperature with more than 12 days exposure required to produce  $LT_{95}$ . For *C. hemipterus*, mortalities below 48 hours at the  $LT_{95}$  level were recorded only for adults (18 hours) and pupae (46 hours) whereas for *U. humeralis* all  $LT_{95}$  mortalities were more prolonged than 48 hours.

For a comparison of sensitivity between species to the different treatments, results are given in Table 1 at the  $LT_{50}$  level.

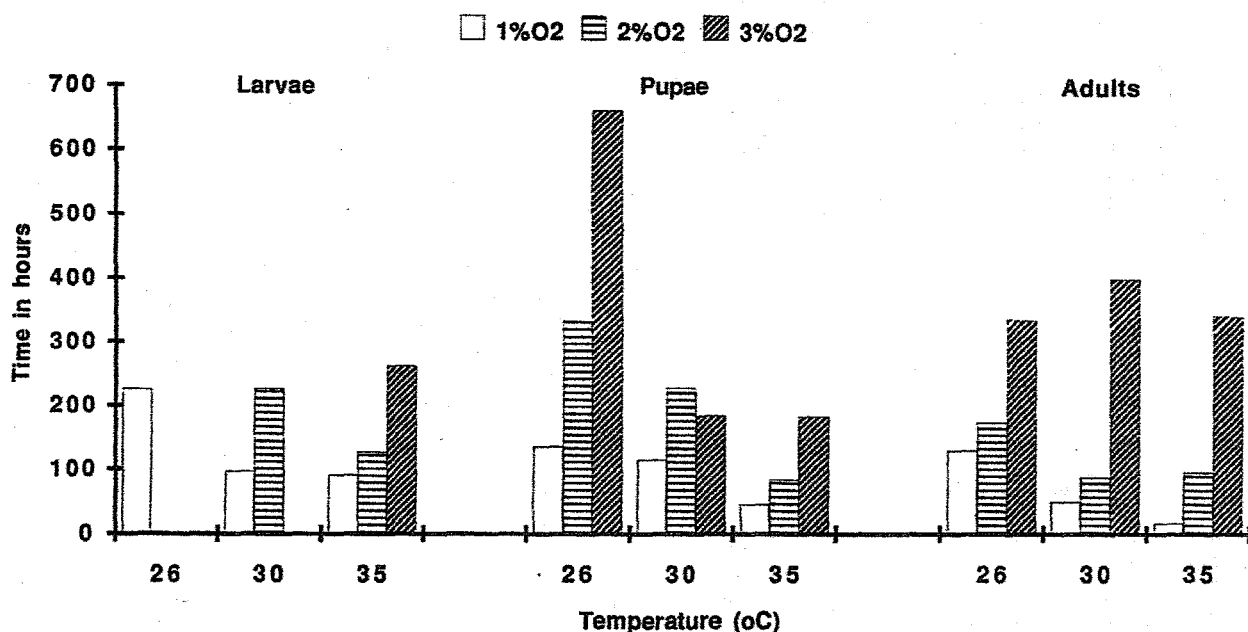


Fig. 1. Mortalities at the  $LT_{95}$  level for three stages of *Carphophilus hemipterus* at three temperatures.

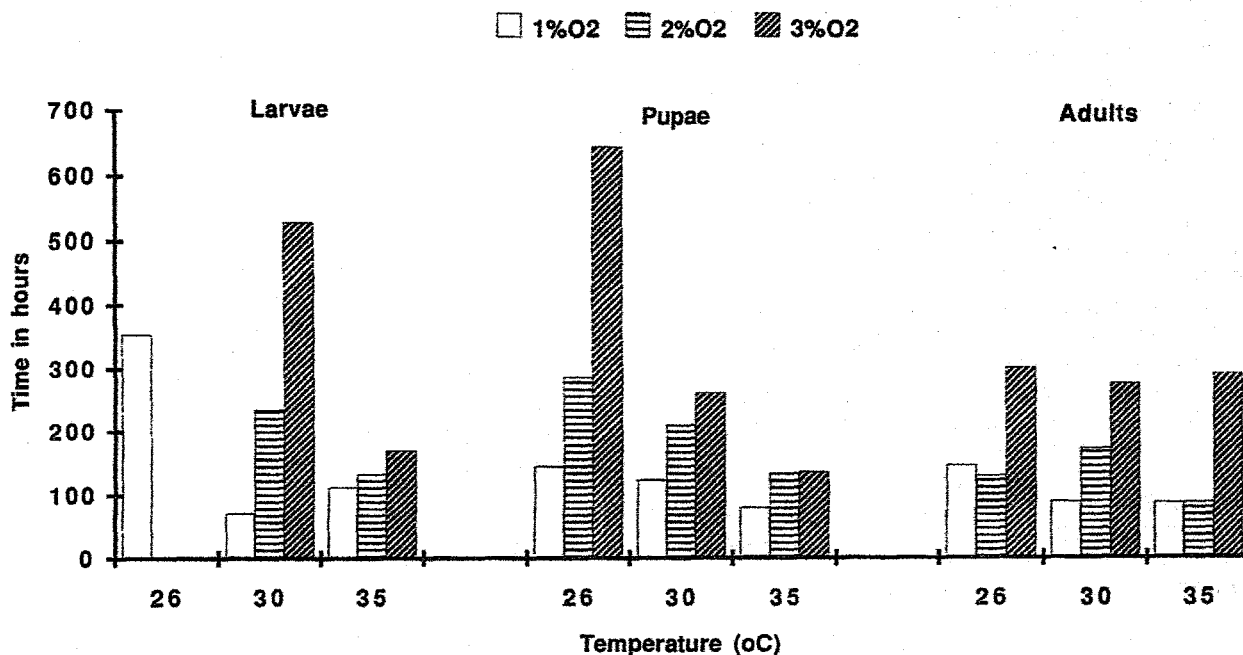


Fig. 2. Mortalities at the  $LT_{95}$  level for three stages of *Urophorus humeralis* at three temperatures.

**Table 1.** Comparison of sensitivities at the LT<sub>50</sub> level, of larvae, pupae and adults of *Carpophilus hemipterus* and *Urophorus humeralis* to three burner-gas concentrations at three temperatures. (NS = non-significant regression, numbers in brackets represent 95% confidence limits.)

Gas mixture	Stage	Temperature					
		26°C		30°C		35°C	
		<i>C. hemipterus</i>	<i>U. humeralis</i>	<i>C. hemipterus</i>	<i>U. humeralis</i>	<i>C. hemipterus</i>	<i>U. humeralis</i>
1%O <sub>2</sub>	Larva	112(97–136)	112(73–913)	60(51–69)	36(23–46)	60(54–67)	57(51–63)
85%N <sub>2</sub>	Pupa	78(62–109)	102(83–116)	24(17–30)	50(44–58)	26(23–30)	45(40–49)
14%CO <sub>2</sub>	Adult	69(61–77)	66(58–72)	29(25–32)	48(42–53)	11(10–12)	38(32–44)
2%O <sub>2</sub>	Larva	NS	NS	105(88–138)	89(72–123)	69(61–81)	82(70–91)
84.7%N <sub>2</sub>	Pupa	137(61–313)	109(95–126)	43(12–59)	71(64–82)	36(31–40)	60(51–68)
13.3%CO <sub>2</sub>	Adult	81(21–141)	83(77–88)	50(5–91)	82(76–87)	36(31–41)	66(58–72)
3%O <sub>2</sub>	Larva	NS	NS	317(275–405)	218(186–289)	104(82–121)	144(139–149)
85%N <sub>2</sub>	Pupa	183(134–286)	149(95–54847)	108(101–114)	139(121–156)	108(99–121)	94(90–99)
12%CO <sub>2</sub>	Adult	151(134–185)	196(165–232)	138(119–180)	173(153–219)	176(152–233)	157(142–178)

## Discussion

Soderstrom et al. (1992) examined the influence of temperature over the range 38–42°C on the influence of hypoxia and hypercarbia on *T. castaneum* adults for 6-hour exposures. Although the different experimental conditions make comparison difficult, their results clearly indicate that raised temperatures could be used to reduce treatment duration. Possibly the fact that the nitidulid beetles in this experiment are also field pests that develop normally at high temperatures had an attenuating influence on the effect of temperature on insect mortality under exposure to the burner-gas modified atmospheres. These results would not enable CA treatments using burner-gas to replace conventional fumigations when quick turn-around of the fruit in the treatment chamber is required.

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