Navarro, S. and Donahaye, E. [Eds.](1993) Proc. Int. Conf. Controlled Atmosphere and Fumigation in Grain Storages, Winnipeg, Canada, June 1992, Caspit Press Ltd., Jerusalem, pp. 115-120.

LOW OXYGEN ATMOSPHERES FOR THE CONTROL OF *Callosobruchus maculatus* (FABRICIUS) AND *Acanthoscelides obtectus* (SAY)

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ABSTRACT

Bruchids are major pests of stored legume seeds and are currently controlled most frequently by the use of chemical fumigants and protectants. The use of controlled atmospheres (CAs) has gained popularity in recent times for the control of stored-product pests. This study was undertaken to investigate the influence of CAs on the control of bruchids. Mortality of eggs, larvae, pupae, and adults of the cowpea seed beetle, *Callosobruchus maculatus* (Fabricius) and the bean bruchid, *Acanthoscelides obtectus* (Say) was tested under three nitrogen atmospheres with low-oxygen (O_2) contents of 1%, 2%, and 3%, at two different temperatures: 25°C and 32°C, and at 70% relative humidity. Complete mortality of all eggs, young larvae, old larvae, pupae, and adults of both bruchid pests was obtained at both temperatures within 1-4 days, 3-9 days, 5-13 days, 5-15 days, and 1-5 days, respectively. Mortality of the life stages occurred generally faster at the higher temperature. *A. obtectus* was generally more susceptible to the low-O_2 atmospheres than was *C. maculatus*.

INTRODUCTION

Bruchids are well-known pests of stored legume seeds, frequently causing severe losses in many tropical and subtropical areas (Singh, 1990). The damage is the result of larvae feeding inside the seed, causing weight losses, reduced seed quality, and poor seed viability. Storage losses due to bruchids as high as 70% in field beans (Khamala, 1978) and 60% in cowpeas (Caswell, 1981), have been reported. Bruchid control in stored legumes using chemical fumigants and protectants is effective, but there may be problems of objectionable residues on treated commodities, handling hazards, and insect development of resistance to the chemicals. Atmospheres with low-O_2 content offer a safe, residue-free alternative to chemical fumigants and protectants for controlling insects infesting stored grain and grain products (McGaughey and Akins, 1989). Hitherto, studies on the effects of O_2

deficient atmospheres appear to have concentrated on insects that attack stored cereals (Bailey and Banks, 1975; 1980; Annis, 1987; Banks and Annis, 1990). In this study, we investigated the mortality of eggs, larvae, pupae, and adults of two major bruchid pests, the cowpea seed beetle, *Callosobruchus maculatus* (F.) and the bean bruchid, *Acanthoscelides obtectus* (Say), in three low-O_2 atmospheres at two different temperatures and constant relative humidity (r.h.).

MATERIALS AND METHODS

The study was carried out at the Federal Biological Research Centre for Agriculture and Forestry, Institute for Stored Product Protection, Berlin, Germany. The *C. maculatus* and *A. obtectus* were obtained from cultures maintained at 25°C and 70% r.h. on blackeye cowpea. Both insects were tested at the following developmental stages: eggs (< 2 days old), immature larvae (7-10 days old as larvae), mature larvae (14-17 days old as larvae), pupae (0-4 days old as pupae) and adults (> 1 day old). *C. maculatus* was treated in batches of 50 eggs on cowpea seeds or 50 larvae or pupae in seeds or 20 adults, placed in separate wire cages (5.0 cm length; 1.5 cm diameter) capped with foam rubber stoppers. *A. obtectus* was treated likewise, except that its eggs were wrapped carefully with a few seeds in tissue paper for treatment. The low O_2 atmospheres to which the insects were exposed were: $1\%O_2/99\%N_2$, $2\%O_2/98\%N_2$, and $3\%O_2/97\%N_2$. The O_2 and N_2 were mixed using a Calibrated Gas Mixture Generator. The insects were exposed to the gas mixtures in gastight connected Dressel flasks. The overall range of exposure was from 1-15 days and at 25°C and 32°C respectively, under a humidity regime of approximately 70% r.h.. Exposure of the respective developmental stages of the insects to the different gas mixtures was replicated three times, along with controls. Adult mortality was recorded at the end of each exposure period with ample allowance for possible recovery. For eggs, larvae, and pupae, the treated infested seeds were placed in separate Petri dishes and sufficient time allowed for adult emergence at 25°C. Percentage mortality was calculated and corrected using the formula of Abbott (1925).

RESULTS AND DISCUSSION

Figures 1 and 2 summarize the time in days required for 100% mortality of the various life stages of *C. maculatus* and *A. obtectus* when exposed to CAs containing $1\%O_2/99\%N_2$, $2\%\ O_2/98\%N_2$, or $3\%O_2/97\%N_2$, and at 25°C and 32°C respectively. For all three CAs and temperatures, complete mortality of both eggs and adults was achieved in 1-5 days and 1-4 days for *C. maculatus* and *A. obtectus* respectively (Fig. 1). For all CAs and temperatures, all immature larvae, mature larvae, and pupae of both species

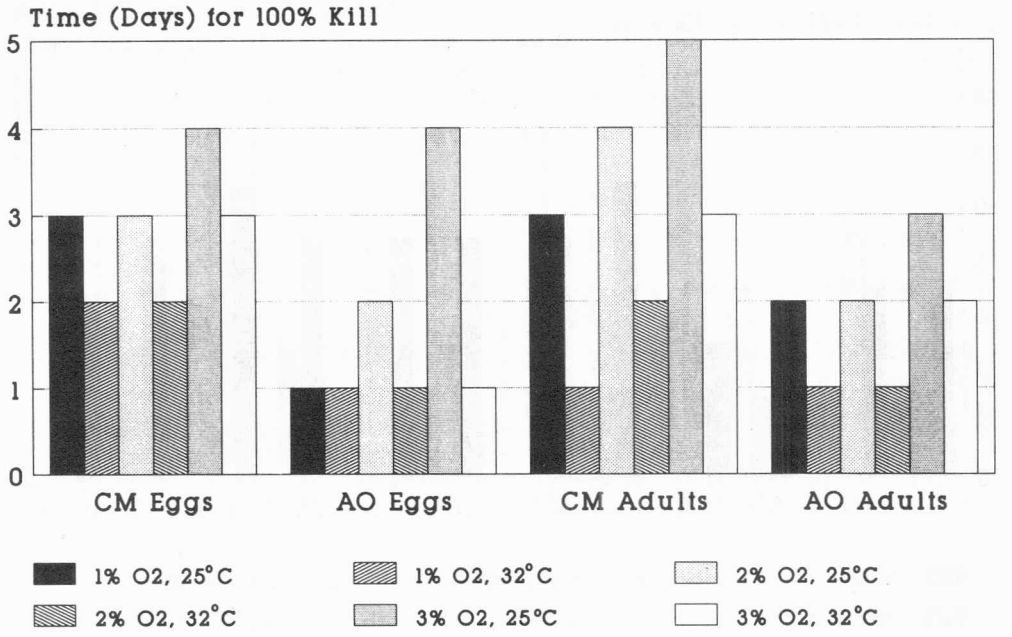

Fig. 1: Time for 100% kill of eggs and adults of two bruchid species in low-oxygen atmospheres, at 70% r.h. (CM = *C. maculatus*; AO = *A. obtectus*).

were killed within 3-9 days, 5-13 days, and 5-15 days, respectively (Fig. 2). Clearly, the mature larvae and pupae of both species were more tolerant to the low-O_2 atmospheres than were the egg and adult stages. The exposure times needed for complete mortality of *C. maculatus* in the $1\%O_2/99\%N_2$ atmosphere recorded here are generally consistent with earlier observations reported by Storey (1978). Generally, the lower the O_2 content in N_2, the shorter was the exposure period to obtain 100% mortality. For 100% mortality of *C. maculatus* pupae, 9 days were needed in the $1\%O_2$ atmosphere at 25°C, whereas the same mortality at the same temperature was recorded in only approximately 15 days in the 3% O_2 atmosphere. This trend has also been observed with some other stored-product pests and varies with insect species and developmental stage (Navarro, 1978; Rameshbabu *et al.*, 1991). Mortality of the different stages of the pests was also generally faster at the higher temperature. For instance, while all the *A. obtectus* eggs were killed within 1 day of exposure to the $3\%O_2/97\%N_2$ atmosphere at 32°C, approximately 4 days were required for the same level of mortality to be obtained at 25°C. Several other studies (Bailey and Banks, 1980; Reichmuth, 1987; Rameshbabu *et al.*, 1991) have also reported that the effect of low-O_2

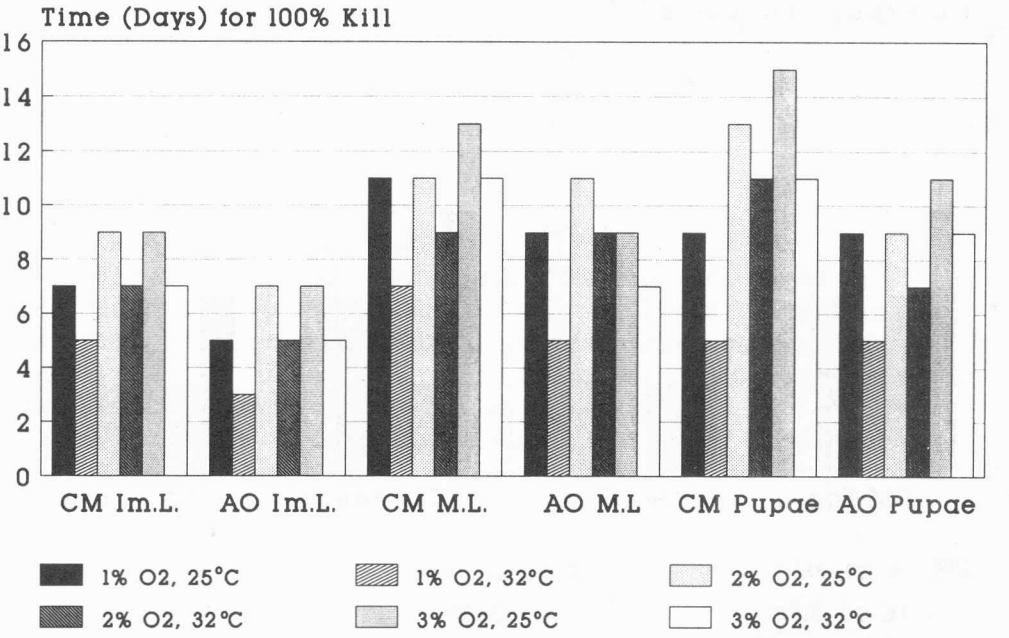

Fig. 2: Time for 100% kill of larvae and pupae of two bruchid species in low-oxygen atmospheres, at 70% r.h. (CM = *C. maculatus*; AO = *A. obtectus*; Im.L.= Immature larvae; M.L. = Mature larvae).

atmospheres is strongly dependent on temperature. For example, with *Sitophilus granarius* in 1.3% O_2 in N_2, Bailey and Banks (1980) reported that almost complete mortality (> 99.5%) was obtained in 2 weeks at 29.4°C, but in 3 weeks only at 23.9°C. Figures 1 and 2 also indicate that *A. obtectus* was generally more susceptible to the low-O_2 atmospheres than was *C. maculatus*. This apparent difference between the action on these two bruchids supports the conclusion of Navarro (1978), that generalization from one insect species to another may be subject to error. Therefore, the action of these low O_2 atmospheres should be tested against other major bruchid species that attack stored legume seeds.

The currently recommended exposure schedule by Banks *et al.* (1991) for low-O_2 CAs for insect control in stored grain is 14-21 days in 1% O_2 at 25-29°C. For legume seeds infested with either *C. maculatus* or *A. obtectus* or both, this recommended exposure period will be effective in controlling the pests even if the O_2 content reaches 3%. Furthermore, for commodities stored at high temperatures (> 30°C) that prevail often during harvest in many legume seed producing countries, low-O_2 CAs (3% O_2 or less) can provide a rapid means of disinfestation with exposures of 11 days or even less.

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REFERENCES

Abbott, W.S. (1925) A method of computing the effectiveness of an insecticide. *J. econ. Ent.* **18,** 265-267.

Annis, P.C. (1987) Towards rational controlled atmosphere dosage schedules: a review of current knowledge. In: Proc. 4th Int. Work. Conf. Stored-Product Protection, Tel Aviv, Israel. (Edited by Donahaye, E. and Navarro, S.), pp. 128-148.

Bailey, S.W. and Banks, H.J. (1975) The use of controlled atmospheres for storage of grain. In: Proc. 1st Int. Work. Conf. Stored-Product Entomology, Savannah, GA, USA, pp. 362-374.

Bailey, S.W. and Banks, H.J. (1980) A review of recent studies of the effects of controlled atmospheres on stored product pests. In: Controlled Atmosphere Storage of Grains, (Edited by Shejbal, J.), pp. 100-118. Elsevier Sci. Publ. Co., Amsterdam, Holland.

Banks, H.J. and Annis, P.C. (1990) Comparative advantages of high CO_2 and low O_2 types of controlled atmospheres for grain storage. In: Food Preservation by Modified Atmospheres, (Edited by Calderon, M. and Barkai-Golan, R.), CRC Press, Boca Raton, FL, USA. pp. 93-122.

Banks, H.J., Annis, P.C. and Rigby, G.R. (1991) Controlled atmosphere storage of grain: The known and the future. In: Proc. 5th Int. Work. Conf. Stored-Product Protection, Bordeaux, France, Sept. 1990. (Edited by Fleurat-Lessard, F. and Ducom, P.), pp. 695-706.

Caswell, G.H. (1981) Damage to stored cowpea in the northern part of Nigeria. *Samaru J. Agric. Res.,* **1,** 11-19.

Khamala, C.P.M. (1978) Pests of grain legumes and their control in Kenya. In Pests of Grain Legumes: Ecology and Control, (Edited by Singh, S.R., van Emden, H.F. and Taylor, T.A.), Academic Press, London, UK., pp. 127-134.

McGaughey, W.H. and Akins, R.G. (1989) Application of modified atmospheres in farm grain storage bins. *J. stored Prod. Res.* **25,** 201-210.

Navarro, S. (1978) The effects of low oxygen tensions on three stored product insect pests. *Phytoparasitica,* **6,** 51-58.

Rameshbabu, M., Jayas, D.S. and White, N.D.G. (1991) Mortality of *Cryptolestes ferrugineus* (Stephens) adults and eggs in elevated carbondioxide and depleted oxygen atmospheres. *J. stored Prod. Res.* **27,** 163-170.

Reichmuth, Ch. (1987) Low oxygen content to control stored product insects. In: Proc. 4th Int. Work. Conf. Stored-Product Protection, Tel Aviv, Israel. (Edited by Donahaye, E. and Navarro, S.), pp. 194-207.

Singh, S.R. (1990) Insect Pests of Tropical Food Legumes. John Wiley & Sons, Chichester, UK., 451 pp.

Storey, C.L. (978) Mortality of cowpea weevil in a low oxygen atmosphere. *J. econ. Ent.* **71,** 833-834.