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THE EFFECTS OF LOW OXYGEN ATMOSPHERES ON THE POWDERPOST BEETLE, *LYCTUS BRUNNEUS* (STEPHENS)

Mark Gilberg and Alex Roach

Abstract—The effect of low oxygen atmospheres on all life stages of the powderpost beetle, *Lyctus brunneus* (Stephens), was investigated. Artificial diet blocks infested with immature stages of *Lyctus brunneus* were exposed to a low oxygen atmosphere (0·4% oxygen, balance nitrogen) at 30°C and 70% RH for varying lengths of time. Mortality counts for immature stages were based on the relative number of adult beetles emerging from the treated and untreated diet blocks. One·hundred percent mortality was observed for eggs, larvae and pupae exposed to low oxygen atmospheres for six, eight and 12 days, respectively. Adult beetles similarly exposed to low oxygen atmospheres all died within three days.

1 Introduction

Oxygen-deficient atmospheres have been shown to be a viable alternative to conventional chemical fumigants for the treatment of insect-infested museum objects [1–4]. Lethal exposure times for a number of common museum pests have been determined [5–7]. Few studies, however, have been devoted to the effect of low oxygen atmospheres on wood-borers such as *Lyctus brunneus* (Stephens) [7, 8]. *Lyctus brunneus* is a common museum pest which infests the sapwood of many species of hardwood [9]. Given that wood-borers are internal feeders, and are thus subject to a restricted air supply, it is possible that *Lyctus brunneus* may tolerate low oxygen environments for prolonged periods of time [10]. In the following study the efficacy of low oxygen atmospheres against adult *Lyctus brunneus* and its developmental stages is investigated. The potential application of low oxygen atmospheres as a fumigation technique for the treatment of wooden objects infested with wood-borers will be discussed in terms of the results of these experimental trials.

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2 Methods and materials

2.1 Stock cultures [11]

Stock cultures of *Lyctus brunneus* were obtained from the Forest Research Division of the Forestry Commission of New South Wales, Sydney. Adult insects were reared on an artificial diet consisting of soluble starch (144g), wheatmeal (240g), yeast extract (8g), bacteriological peptone (16g) and water (200ml). To prepare the artificial diet the dry ingredients were thoroughly mixed in a bowl and distilled water was slowly added to form a dough. After kneading, the wet dough was pushed into the wells of a cookie tin and baked at 86°C for approximately 8–10 hours in a convection oven. Upon cooling, the diet blocks were transferred to a large glass jar fitted with a filter paper lid and placed in an incubator and conditioned at 26°C and 70% RH for two weeks. Using this method a total of 14 diet blocks each weighing approximately 15g was prepared.

Wide-mouthed glass jars (2·5 liters) were used as culture vessels. Each glass jar was fitted with a screw-top lid containing a breathing hole covered with coarse filter paper. The filter paper was sprayed with a 0·1% solution of Dicofol (Kelthane) in alcohol to prevent the invasion of parasitical mites. A disk of filter paper was also placed on the bottom surface of each jar to support the diet block and adult beetles.

Stock cultures were prepared by releasing 100 young adult beetles into a culture vessel containing 14 diet blocks (approximately 200g of artificial diet). The culture vessel was then incubated at 26°C and 70% RH until the emergence of the next generation of adult beetles (70 days post oviposition). These were then collected by hand and added to fresh diet blocks to establish a new stock culture. All stock cultures were prepared in this manner.

To make the preparation of new stock cultures easier, the incubator was illuminated immediately following the emergence of the first adult beetles from the old stock culture. This forced the emerging beetles to remain in their

pupal chambers, thus minimizing oviposition on the old diet blocks from which they had emerged. The emerging adults were then collected a week later for use in establishing the new stock culture.

2.2 Test cultures

Adult and immature stages of *Lyctus brunneus* were obtained by placing 100 adult beetles collected from stock cultures onto 14 diet blocks for three days (eggs) and seven days (larvae, pupae, adults) at 26°C and 70% RH. The adult beetles were then removed and the diet blocks transferred to individual glass jars (550ml) fitted with filter paper lids. The cultures were then incubated as above until the desired life-stage of the developing progeny was obtained. This was estimated on the basis of results obtained from preliminary studies by one of the authors (MG) in which the number of days from oviposition was correlated with developmental stage. For this study the ages of the adult and immature stages infesting the diet blocks at the start of exposure were as follows:

0–3 days: eggs
40–47 days post oviposition: larvae ('young larvae' [11])
60–67 days post oviposition: pupae
0–7 days post emergence: adults.

Given the experimental conditions (26°C and 70% RH) a life cycle of approximately 70 days from egg to adult was observed. Because the designated stages were estimates based on the number of days of development from the start of oviposition, some overlapping in developmental stages would be expected.

2.3 Exposure procedure

Test and control cultures were exposed to low oxygen atmospheres at 30°C and 70% RH for a period of one to 12 days. The glass jars containing the infested diet blocks were sealed with a metal screw-top lid fitted with an inlet and outlet valve. Test cultures were continuously purged with a commercially prepared mixture of oxygen and nitrogen delivered under pressure from a compressed gas cylinder (0·4% oxygen, balance nitrogen). The gas mixture was humidified by passing the gas through a Dreschel gas-washing bottle containing a glycerine/water mixture to obtain a relative humidity of 70%. A constant temperature was maintained by placing both the gas-washing bottles and the glass jars inside a convection oven maintained at 30°C.

The temperature and humidity of the exposure chambers were monitored by the inclusion of a thermometer and relative humidity indicator card. The oxygen content of the gas mixture leaving the exposure chamber was monitored using a Beckman Model 715 Process Oxygen Monitor.

Tests on eggs, larvae, pupae and adults were conducted without the use of replicates because of difficulties in securing large numbers of these developmental stages at any one time. However, all tests were repeated three times for each developmental stage to confirm the results. Parallel tests with controls purged with air were similarly conducted for each developmental stage. In total, seven test cultures and seven control cultures were purged simultaneously.

Immediately after exposure the glass jars containing the treated and untreated diet blocks were transferred from the exposure chamber, refitted with filter paper lids and cultured as before at 26°C and 70% RH in an incubator. Counts of adult beetles emerging from treated and untreated diet blocks began when the first emergence was observed in the untreated control blocks and ended approximately three months later. Mortality counts for immature stages were based on the relative number of adult beetles emerging from the treated and untreated diet blocks. An average of 66, 107 and 96 adult beetles emerged from the untreated control diet blocks infested with eggs, larvae and pupae, respectively.

Adult mortality was based on counts of 10 adult beetles exposed in each test. In total, 30 adults were exposed, as each test was repeated three times.

3 Results and discussion

The results of the exposure tests for *Lyctus brunneus* are given in Table 1 and Table 2.

The order of tolerance to low oxygen atmospheres was pupae > larvae > eggs > adults. One hundred percent mortality was observed after six days for eggs, eight days for larvae, 12 days for pupae and three days for adults.

Delayed emergence of adult beetles from infested diet blocks exposed to low oxygen atmospheres was evident for all developmental stages. Adult emergence from diet blocks infested with

Table 1 Percent mortality of immature stages of the powderpost beetle,* Lyctus brunneus, *after exposure to low oxygen atmospheres at 30°C and 70% RH for 1–12 days*

Stage	Age (days)	Exposure period in days											
		1	2	3	4	5	6	7	8	9	10	11	12
Eggs	0– 3	0	51	71	95	99	100	100	—	—	—	—	—
Larvae	40–47	12	9	—	39	—	87	—	99	—	100	—	100
Pupae	60–67	10	13	—	24	—	57	—	92	—	99	—	100

*Average of three exposures.

Table 2 Percent mortality of adult beetles of* Lyctus brunneus *after exposure to low oxygen atmospheres at 30°C and 70% RH for 0–4 days*

Stage	Age (days)	Exposure period in days					
		0·25	0·5	1	2	3	4
Adult	0–7	0	10	20	90	100	100

*Average of three exposures.

larvae and pupae which had survived exposures of six days to low oxygen atmospheres occurred several weeks later relative to control cultures.

4 Conclusion

Exposure to low oxygen atmospheres (0·4%) for 12 days at 30°C and 70% RH proved lethal to all life stages of *Lyctus brunneus*. With slight modification, these experimental conditions may be readily translated into practice for the treatment of museum objects.

Though the experimental trials were conducted at 70% RH, exposure to lower relative humidity has been shown to enhance the action of low oxygen atmospheres [12, 13]. It would therefore be unnecessary to elevate the relative humidity above ambient conditions when exposing a museum object to low oxygen atmospheres. However, it would be necessary to maintain a temperature of 30°C throughout the exposure period. The action of low oxygen atmospheres is strongly temperature-dependent. Raising the temperature greatly decreases the exposure time needed to obtain 100% mortality [12, 13].

It is highly unlikely that exposure at 30°C for 12 days would adversely affect most museum objects, particularly in the absence of oxygen. Any dimensional change would be minimal given the buffering capacity of many museum materials.

Though the exposure period needed to produce effective control of all developmental stages is considerably longer than that generally associated with chemical fumigants, this is more than offset by the inherent health and safety benefits of using low oxygen atmospheres.

From a practical standpoint, continuous purging with nitrogen is an inefficient means of generating low oxygen atmospheres for the disinfestation of museum objects. Alternatively, chemical oxygen scavengers may be used to achieve and maintain a sufficiently low oxygen concentration throughout the entire exposure period [3, 4].

The relationship between wood thickness and the efficacy of low oxygen atmospheres against *Lyctus brunneus* is problematical. Little information exists with regard to the diffusion of oxygen through wood as a result of the development of concentration gradients across its surface. The rate of diffusion of non-swelling gases, such as oxygen, through wood varies from species to species; it is greater in sapwood than heartwood and occurs primarily in the axial direction [14]. In general, this diffusion is very rapid [14] and it is highly unlikely that sufficient residual oxygen will remain in the wood to support insect activity. Moreover, insect respiration will also contribute to the removal of any residual oxygen if the oxygen concentration within the wood is not reduced to a level which is lethal during the exposure period [15]. Nonetheless, the 12-day exposure period recommended above for wood infested with *Lyctus brunneus* should be considered a minimum. Longer treatment times may, in fact, be necessary for the disinfestation of relatively large wooden objects.

Materials and suppliers

Kelthane: Rohm and Haas Co., 969 Burke Road, Camberwell, Victoria 3124, Australia.

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Résumé—On a étudié l'effet d'atmosphères à basse teneur en oxygène sur les divers stades de vie de la lycte, *Lyctus brunneus* (Stephens). Des blocs de nourriture artificielles infestés par le *Lyctus brunneus* immatures ont été exposés à une atmosphère pauvre en oxygène (0,4% d'oxygène dans l'azote) à 30°C et 70% HR pendant des durées variables. L'evaluation de la mortalité des sujets immatures était basée sur le nombre relatif des individus adultes sortant des blocs traités et non traités. On a observé cent pour cent de mortalité dans les oeufs, les larves et les chrysalides exposées à cette atmosphère, respectivement au bout de six, huit, et 12 jours. Les adultes exposées à la même atmosphère sont tous morts dans les trois jours.

Zusammenfassung—Der Beitrag beschreibt den Einfluß einer Atmosphäre mit niedrigem Sauer stoffgehalt auf alle Lebensstadien des Braunen Splintholzkäfers, *Lyctus brunneus* (Stephens).

Hierzu wurden mit unreifen Stadien dieses Holzschädlings befallene künstliche Nährsubstrate einer niedrigen Sauerstoffatmosphäre (0·4% Sauerstoff, ausgetauscht gegen Stickstoff) bei 30°C und 70% relativer Feuchte für bestimmte Zeiten ausgesetzt. Die Mortalitätsrate für die unreifen Stadien bezieht sich dabei auf die relative Anzahl fertiger Insekten, die jeweils aus den behandelten oder unbehandelten Nährsubstraten ausschlüpften. Eine Sterblichkeitsrate von 100% wurde für die Eier, Larven und Puppen beobachtet, die sechs, acht oder 12 Tage dem niedrigen Sauerstoffgehalf ausgesetzt waren. In ähnlichen Versuchsreihen starben alle fertigen Insekten innerhalb von drei Tagen.