## The effect(s) of carbaryl-treated seed on body maintenance and survival of the multi-mammate mouse, Mastomys natalensis (sensu lato)

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#### **ABSTRACT**

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Maintenance, expressed as change in daily body mass, and survival rates of *Mastomys natalensis* (sensu lato) were recorded from May to September 1994 in laboratory feeding trials, to investigate the short-term effects of a carbaryl insecticide on these variables. Individuals were subjected to seeds treated with carbaryl insecticide in three different treatments (5, 10 and 20 g of carbaryl/kg of seeds). Carbaryl did not have short-term adverse effects on growth and survival of this species when the seeds were kept in the laboratory and when they were exposed to environmental conditions between measurements. This suggests that the ingestion of carbaryl-treated seeds is not the cause of the decline in density of *M. natalensis* on rehabilitating coastal dune forests at Richards Bay.

Keywords: Body maintenance, carbaryl-treated seed, effect(s), Mastomys natalensis, mouse, survival

### INTRODUCTION

The post-mining rehabilitation of coastal dunes north of Richards Bay by Richards Bay Minerals (RBM) comprise the spreading of topsoil collected prior to mining on restructured dunes, followed by the sowing of a seed mixture containing various exotic and indigenous species (Camp 1990). The seeds are treated with carbaryl (Carbaryl [1-Naphthalenyl methyl carbamate-(CI2H12O2N)] is the active chemical in the commercial insecticide Karbadust that is used to treat seed at RBM's Ecology Centre) during storage (Andrew Denton 1995, personal communication).

The multi-mammate mouse, *Mastomys natalensis* (sensu lato), dominates small mammal communities

after 8 weeks of rehabilitation (Van Aarde, Ferreira, Kritzinger, MacMahon, Miller & Leumann 1993) and reaches densities of  $\approx 50$  animals/ha about 6 months after the start of rehabilitation. The peak in densities is followed by a sharp decline to  $\approx 15$  animals/ha after 8 months of rehabilitation (S.M. Ferreira 1994, unpublished data).

Studies on the population dynamics of *M. natalensis* suggest that the decline in density could be associated with the species' poor competitive abilities (Coetzee 1967; Swanepoel 1972; Meester, Lloyd & Rowe-Rowe 1979). Although this species has the advantage of wide habitat tolerances and extreme adaptability (Skinner & Smithers 1990), it may, within months of colonizing recently disturbed areas, be replaced by species with better competitive abilities when more complex plant communities become established. On the other hand, seed availability or quality

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(Carothers & Fabian 1984) may explain the drastic decline in their density.

Although M. natalensis is omnivorous, it feeds predominantly on seeds and fruit augmented by smaller portions of insects (Skinner & Smithers 1990). Detrimental effects of insecticides may result directly from ingestion of the treated seed and insects killed by the insecticide (Sullivan 1990), or indirectly from dermal penetration resulting from contact (Shah, Monroe & Guthrie 1983). The population decline could thus be a result of insecticide ingestion. Furthermore, seed and insect eaters are susceptible to pesticide accumulation in their bodies (Greig-Smith 1987), which may cause death in small mammals. Although dermal penetration seems to be less important as being toxic for most animals, dermal penetration of carbaryl was higher in mice than in other non-target species that were tested (Shah et al. 1983). This is consistent with results from studies on Australian mammals by McIlroy (1982) who suggested that rodents were highly sensitive to poison.

In rodents that were exposed to pesticides and then starved, the pesticide concentrations in fat and body tissues increased (Dale, Gaines & Hayes Jr 1962), which may eventually lead to death. Bernard & Gaertner (1964) argue that pesticides ingested in small concentrations by small animals are retained in sufficient quantities to produce death, and cause a decline in fertility in rats and mice. Insecticides may thus have an effect on the demographic parameters of rodent populations. To the best of our knowledge it is unknown whether carbaryl has the same effect on the multimammate mouse, a species which is known to occur in high numbers in disturbed environments and which is also of considerable economic importance to crop-producing farmers in southern Africa (Skinner & Smithers 1990).

The present study reports on the effect(s) of carbaryl-treated seed on the body maintenance and survival of *M. natalensis* and relates the results to patterns of population change of *M. natalensis* at Richards Bay. We argue that animals need to maintain themselves individually before reproducing. An effect on survival rates and body maintenance will indicate an impact on individual maintenance with subsequent repercussions for reproductive output. The present study was therefore designed to quantify the effect(s) of treated seed on individual maintenance.

## **MATERIALS AND METHODS**

## Study animals

Study animals (12 males and 16 females) were collected at the Richards Bay study area (Van Aarde et al. 1993) during December 1992 and were subsequently kept at the Mammal Research Institute (MRI)

of the University of Pretoria. Additional animals (four males and 11 females) were obtained from the Medical Research Council (MRC) and others were caught on the university's experimental farm with Sherman livetraps from May to August 1994 (27 males and 15 females). The animals were housed individually in separate cages, with enough water to drink and food to eat (Epol mouse cubes, Premier Food Industries Ltd) while they were not exposed to trial conditions. Individuals were not considered for the present study unless adult body masses had been attained (Skinner & Smithers 1990).

## Food provision

Four types of seed (*Pennisetum glaucum, Crotolaria juncea, Helianthus annuus* and *Cynodon dactylon*) were used in the experimental trials and were obtained from Gunson Seed Suppliers (Ltd). They were mixed in equal proportions. These seeds were the same as those sown on dunes as part of the rehabilitation programme (A. Denton 1995, personal communication).

Two groups, consisting of six animals each, received treated seed (treated with 5 g of carbaryl/kg of seed) and untreated seed, respectively, along with adequate water which was provided *ad lib* throughout all experiments. The experiment continued for 30 d, each individual receiving 10 g of seed daily. Seed was provided in plastic containers (3 x 4 x 6 cm) attached to the bottom of the cage with Prestik. The amount of seed eaten was determined daily by weighing the seed left over in the container. None of the individuals ate more than 10 g/d. Only 45,9 % of individuals survived in both groups of animals, suggesting a nutrient deficiency of some kind.

The experiment was repeated with four groups of animals consisting of six animals each. One group received untreated seed (10 g/d). A second group received untreated seed (10 g/d), supplemented with mouse cubes (10 g/d). A third group received treated seed (10 g/d of seed treated with 5 g of carbaryl/kg of seed), while the last group received treated seed (10 g/d of seed treated with 5 g of carbaryl/kg of seed) and treated mouse cubes (10 g/d of mouse cubes treated with 5 g of carbaryl/kg of mouse cubes). Seed and mouse cubes were provided separately in round tins (6 cm diameter and 3 cm high), which replaced the plastic containers. Adequate water was provided as previously. After 15 d, mortality was recorded amongst these individuals receiving seed only, irrespective of whether the seed had been treated or not. In subsequent experiments which continued for 15 d, all individuals were provided with both seed and mouse cubes.

#### **Experimental phases**

Once an acceptable food ration had been established, the investigation consisted of two phases.

During phase 1, the food (seed and mouse cubes) was stored in sealed plastic buckets and tins that were kept in the laboratory where the trials were conducted. During this phase the trials for the different treatments (5 g of carbaryl/kg of food, 10 g of carbaryl/kg of food and 20 g of carbaryl/kg of food) were carried out at different times, which resulted in three trials lasting for 15 d, each with its own control and treatment group.

During phase 2, the food was spread in steel trays and left outside the laboratory where it was exposed to ambient environmental conditions. The daily rations were taken from this food. The procedure was considered to mimic the dilution of carbaryl concentration with time, on seeds sown on dunes at Richards Bay. This phase consisted of one trial conducted with one control group and three treatment groups (5 g of carbaryl/kg of food), 10 g of carbaryl/kg of food, 20 g of carbaryl/kg of food). This trial lasted for 15 d.

# Body maintenance, survival and food consumption

Variations in daily body mass were used as an indicator of body maintenance. The animals were weighed daily with a DB600 Digital Balance (Universal Weight Enterprise Co., Ltd). Mean changes in body mass of individuals in the control and the treatment groups were compared statistically. Survival was recorded as the number of days that the animals survived during a feeding trial. Food consumption was recorded as the amount of food (seed and mouse cubes) consumed in g/d.

## Data analyses

Students' *t*-tests were used to test for significance in differences between means when two groups were compared, and analyses of variance as well as Tukey multiple-range tests were used when more than two groups were compared (Sokal & Rohlf 1969). Significance was taken at the 95% confidence level.

Cochran's C-test was used to investigate the homogeneity of variances (Caughley & Sinclair 1994) and Bartlett's test, to test for the normality of the distribution of the data (Sokal & Rohlf 1969). For data sets characterized by heterogeneous and/or non-normal data, log transformations were used for more than two groups, while the approximate t-test ( $t_s'$ ) (Sokal & Rohlf 1969), was used for two groups. The differences were considered significant when the critical value for type one error,  $t_{0,05}'$ , was less than the absolute value of  $t_s'$ .

The relationships between the amount of food consumed and the initial body mass, and growth rate and the initial body mass, were investigated by means of correlation analyses (Sokal & Rohlf 1969). Significance was taken at the 95% confidence level.

### **RESULTS**

## Food not exposed to the environment (phase 1)

There were no significant differences in initial body mass between control and treatment groups during phase 1 ( $F_{3,30}$  = 2,199, P = 0,11). The change in mean daily body mass recorded for the control and treatment groups did not differ significantly when 5 g of carbaryl/kg of food was used ( $t_8 = -1,678$ ; P =0,13), 10 g of carbaryl/kg of food ( $t'_{10} = -0.343; t'_{0.055} = 2,447$ ) and 20 g of carbaryl/kg of food ( $t_{10} = 1.475;$ P = 0.17) (Table 1). It is interesting to note that change in daily body mass was negative at 20 g of carbaryl/kg of food. All individuals survived to the end of the experiments in which 5 g of carbaryl/kg of food and 10 g of carbaryl/kg of food were used, while two of the six individuals exposed to 20 g of carbaryl/kg of food, died during the trial. However, the difference in survival between the control and treatment groups was not significant when 20 g of carbaryl/kg of food  $(t'_{10} = 0.645; t'_{0.05} = 2.447)$  was used (Table 1).

There was no significant difference between the food consumption of the control and the treatment groups when 5 g of carbaryl/kg of food ( $t_8$  = 0,754; P = 0,47) and 20 g of carbaryl/kg of food ( $t'_{10}$  = -0,422;  $t'_{0,05}$  = 2,447) were used. The food consumption of the treatment group given 10 g of carbaryl/kg of food was significantly higher than that of the control group ( $t_{10}$  = -3,543; P < 0,05) (Table 1).

## Food exposed to the environment (phase 2)

Body masses for the different groups were similar  $(F_{3,20}=0,157,\ P=0,92)$  at the onset of the trial. Change in daily body mass did not differ significantly between the treatment groups  $(F_{3,20}=0,407,\ P=0,45)$ , while all individuals survived to the end of the experiment (Table 2). Food consumption did not differ significantly between treatment groups  $(F_{3,20}=1,284,\ P=0,31)$  (Table 2). Fig. 1 illustrates no decline in daily body mass and survival when there is an increase in carbaryl concentration.

## Comparison between the treatments of phase 1 and phase 2

Change in daily body mass did not differ significantly between phase 1 and phase 2 for the control ( $t_{21} = -1.57$ ; P = 0.13), the 5 g of carbaryl/kg of food treatment ( $t_9 = -0.325$ ; P = 0.754), the 10 g of carbaryl/kg of food treatment ( $t_{10}' = -0.494$ ;  $t_{0.05}' = 2.447$ ) and the 20 g of carbaryl/kg of food treatment ( $t_{10}' = -0.594$ ;  $t_{0.05}' = 2.447$ ) (Tables 1 and 2).

There was no difference between the survival rates during phase 1 and phase 2 for the control group, the 5 g of carbaryl/kg of food treatment and the 10 g of

TABLE 1 Mean change in daily body mass (g/d), survival (number of days survived) and food consumption (g/d) during phase 1 (seed not exposed to environmental conditions)

Change in daily body mass (g/d)	Carbaryl concentration						
	5 g/kg		10 g/kg		20 g/kg		
	Control	Treatment	Control	Treatment	Control	Treatment	
n Mean SE	5 0,081 0,06	5 0,253 0,10	6 0,181 0,10	6 0,293 0,31	6 0,225 0,16	6 -1,087 0,88	
Survival (d)							
n Mean SE	5 15 0	5 15 0	6 15 0	6 15 0	6 15 0	6 10,83 2,64	
Food consumption (g/d)						1	
n Mean SE	5 6,206 0,740	5 5,536 0,490	6 6,450 0,230	6 7,615 0,230	6 5,717 0,330	6 7,502 1,380	

n = Sample size

SE = Standard error of the mean

TABLE 2 Mean change in daily body mass (g/d), survival (number of days survived) and food consumption (g/d) during phase 2 (seed exposed to environmental conditions)

Change in deily hady mass (a/d)	Carbaryl concentration					
Change in daily body mass (g/d)	0 (control)	5 g/kg	10 g/kg	20 g/kg		
n Mean SE	6 0,367 0,120	6 0,319 0,140	6 0,332 0,090	6 0,206 0,090		
Survival (d)						
n Mean SE	6 15 0	6 15 0	6 15 0	6 15 0		
Food consumption (g/d)						
n Mean SE	6 5,670 0,280	6 5,550 0,380	6 4,939 0,430	6 4,990 0,170		

n = Sample size

SE = Standard error of the mean

carbaryl/kg of food treatment. Neither did the survival rates differ significantly for the groups on 20 g of carbaryl/kg of food treatment ( $t'_{10} = 0,645; t'_{0,05} = 2,447$ ) in phase 1 and phase 2 (Tables 1 and 2).

Change in daily body mass and the survival rate declined with an increase in carbaryl concentration in phase 1 while no decline was recorded in phase 2 (Fig. 1).

## DISCUSSION

The effects of pesticides on mammals have been receiving attention since the early sixties (Dale et al. 1962; Bernard & Gaertner 1964). Pesticides have on occasion been used for the control of rodents, but most frequently their effects on mammals and other vertebrates have been a secondary product of attempts to eradicate insect pests. If carbaryl has any

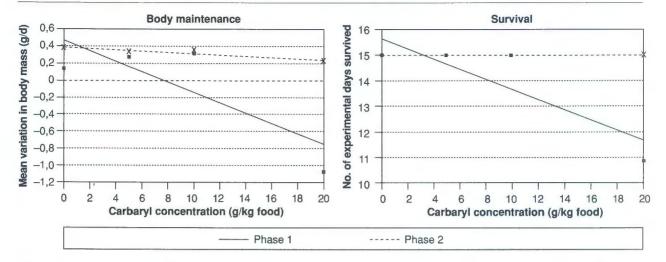


FIG. 1 The effect of carbaryl concentration on maintenance (change in daily body mass) and survival of *Mastomys natalensis*. Phase 1 refers to seed not exposed to the environment, while phase 2 refers to seed exposed to the environment. All these relationships were significant at the 90% level, but none of the slopes differed significantly from zero

direct effects on the demographic parameters of *M. natalensis*, such effects should be observed in the laboratory when individuals of the species are subjected to a diet treated with carbaryl.

The mortalities in both the control and the treatment groups, observed during the food-provision trial when the animals received seeds only, could be due to many factors. One of these could be related to *M. natalensis* being omnivorous, feeding predominantly on seeds and fruit, their diet being augmented with smaller portions of insects (Skinner & Smithers 1990). This implies that the seed provided as food to the animals, although part of their diet, may have been insufficient for maintenance.

Our analyses illustrate that no marked differences in maintenance (change in daily body mass) and survival resulted from carbaryl ingestion at various concentrations. However, the decline in maintenance and survival with an increase in carbaryl concentration on seed not exposed to environmental conditions, does not exclude any effect. The suggestion is rather that higher concentrations than those used in this study will affect individual maintenance and survival of *M. natalensis* negatively.

Environmental exposure of treated food reduced the effect of carbaryl on the maintenance and survival of *M. natalensis* during the present study. Furthermore, the lowest treatment used in this study was higher than that used for treating seed at the RBM Ecology Centre (A. Denton 1994, personal communication). Direct effects of carbaryl on the maintenance and survival of *M. natalensis* are therefore not the cause of the rapid decline recorded for numbers on rehabilitating dunes between the 6–8 months following the initiation of habitat rehabilitation (S.M. Ferreira 1994, personal observation).

The results of this study are consistent with studies on the effects of other pesticides on other small mammals. Swanepoel (1972) found that dieldrin coverspraying did not affect the growth, reproduction or survival of M. natalensis. Although no generalization can be made about the effects of pesticides on wildlife, this result is consistent with results from the evaluation of forest herbicides on small mammals (Sullivan 1990). In contrast, studies on dermal penetration by insecticides (including carbaryl) showed that dermal penetration is higher in mice than in any other species that was tested (Shah et al. 1983). which may result in high concentrations in the body tissues, leading to death. George, McEwen & Fowler (1992) concluded that carbaryl bait treatments have a minimal direct toxic effect on birds and small mammals, although they may have an indirect effect on insectivores and seed eaters. The indirect effect of pesticides may result from reduction of fertility (Bernard & Gaertner 1964), delayed maturity or induction of resorption and abortion of foetuses.

It follows that indirect effects could be masking the carbaryl effect on patterns of variation in *M. natalensis* population parameters on rehabilitating dunes 6–8 months after the initiation of habitat rehabilitation at Richards Bay. These indirect effects need time to realize their full potential and we therefore feel that ecological factors rather than the effect of carbaryl, cause rapid declines in *M. natalensis* numbers, following colonization of rehabilitating dunes.

### CONCLUSION

Seeds treated with carbaryl insecticide have little or no direct short-term adverse effect on the maintenance and survival of *M. natalensis* when such seeds are

ingested from the sand-dune surface or dug from below the surface. The recorded post-colonization decline in density of *M. natalensis* after 8 months of rehabilitation at Richards Bay (S.M. Ferreira 1994, unpublished data) is not due to the effects of carbaryl on growth and survival rates resulting from ingestion of seeds treated with carbaryl insecticide. This does not, however, entirely rule out carbaryl as a potential cause of the decline, since its effects on the fecundity, foetus development, age at maturity and breeding behaviour were not investigated in this study.

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