THE INFLUENCE OF FLUOROACETATE PRODUCING PLANTS UPON SEED SELECTION BY SEED HARVESTING ANTS

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Introduction

Sodium monofluoroacetate (compound 1080) is a highly toxic substance which is used for the eradication of mammalian vermin[2,3,7]. Once absorbed into the body, the toxic manifestations of fluoroacetate (monofluoroacetate) arise from its in vivo transformation into fluorocitrate[11,19]. Fluorocitrate then inhibits the mitochondrial tricarboxylic acid enzyme aconitate hydratase (E.C.4.2.1.3.) with resultant accumulation of citrate[19,20]. In addition, recent studies suggest that fluorocitrate may also inhibit citrate transportation into and out of mitochondria[11]. The consequences of fluoroacetate toxicity are similar in a wide range of organisms and result in a dysfunction of the tricarboxylic acid cycle leading to energy deprivation and death.

Fluoroacetate occurs naturally in several plant genera[14], in particular, in thirty-three species of **Gastrolobium** and **Oxylobium**. Indigenous mammalian herbivores inhabiting areas where such vegetation occurs have a higher tolerance to fluoroacetate than animal populations which have not had evolutionary exposure to fluoroacetate-bearing vegetation[16,24]. The range of these fluoroacetate-tolerant species correlates with the distribution of fluoroacetate-bearing vegetation .

Sodium fluoroacetate can act as a systemic insecticide and it was patented as a moth-proofing agent in the 1930's[22]. Sodium fluoroacetate is known to have good insecticidal action against mustard beetles[5] and aphids [5]. Very little is known, however, about the fluoroacetate susceptibility of invertebrate fauna from areas whose vegetation includes fluoroacetate-bearing plants.

It has been established that many Australian ants are seed harvesters. Harvester and decorator ants may remove large quantities of seed from forests and agricultural pastures for food or for nest building materials[12,15,25].

As fluoroacetate is toxic to invertebrates and because seed harvesting ants co-exist with fluoroacetate-bearing vegetation in some areas of Western Australia, it becomes interesting to speculate on how these invertebrates avoid fluoroacetate intoxication. Investigations were therefore conducted to assess whether the presence of fluoroacetate in the seed of toxic species of **Gastrolobium** influenced the seed selection by harvesting ants in an area where such vegetation occurs.

Methods

The study was conducted during March 1983, in the Dryandra State Forest near Narrogin (32°56'S, 117°11'E) which is approximately 160 km south-east of Perth. The climate is warm mediterranean with an average yearly rainfall of 508 mm.

The vegetation is a typical **Eucalyptus wandoo** open woodland with the understorey dominated by **Gastrolobium microcarpum**, locally present to levels of 80 per cent cover. **Acacia pulchella**, **Astroloma epacridis**, **G. oxyloboides** var. **angustifolia**, and **Lomandra leucocephala** were occasionally present.

All ants mentioned in this study have been sorted to species level. Species names were given where possible. When these were unavailable, they were either coded with Western Australian Institute of Technology (J.D.M.) code numbers or, if voucher specimens are deposited there, with Australian National Insect Collection (ANIC) codes.

Description of Seed

Ants were provided with seed from three leguminous species. These were **G. microcarpum** (Papilionaceae) (mean mass from 10 seeds, 5.4 mg), **Bossiaea eriocarpa** (Papilionaceae) (mean mass, 1.5 mg) and **Acacia pulchella** (Mimosaceae) (mean mass, 6.9 mg). **G. microcarpum** elaborates fluoroacetate (400-600 mg kg^{-1})[1] whereas the others do not. The seed of these species offered ants a choice between toxic and non-toxic seed and a range in seed mass. Seed was previously collected from Dryandra in December, 1982 and all three genera produce seed with elaiosomes[4].

Seed Taking

To determine which ants were seed takers and also their relative capacity to remove seed, twenty seed depots (20 x 15 cm Masonite boards) were placed out in a 5 x 4 grid spaced 5 m apart. In the early morning twenty seeds of each species were placed on each depot and at 2h intervals, for a 24 h period, a count of the number of seeds removed per depot was made. Seed removed from depots was not replenished. The soil temperature (depth 1-2 cm) was also recorded at 2 h intervals. Any ants removing and/or feeding on seed were collected and the time of day recorded.

Determination of the Fate of Harvested Seed

To assess whether the toxic seeds of **G. microcarpum** were removed but later rejected by ants, a seed depot with twenty marked **G. microcarpum** seeds was placed at the nest entrance of a known seed taking ant species (located outside the main study plot). Over a 48 h period, brief inspections of seed removal were made. After approximately 72 h, a 15 x 15 x 25 cm soil core was taken around each nest and the seed content analysed. Seeds were separated from the nests using a combination of sieving and hand sorting. The nest midden was treated separately from the below-ground nest.

Annual Report 6 Mulga Research Centre 1982, Published August 20th 1983 75-80

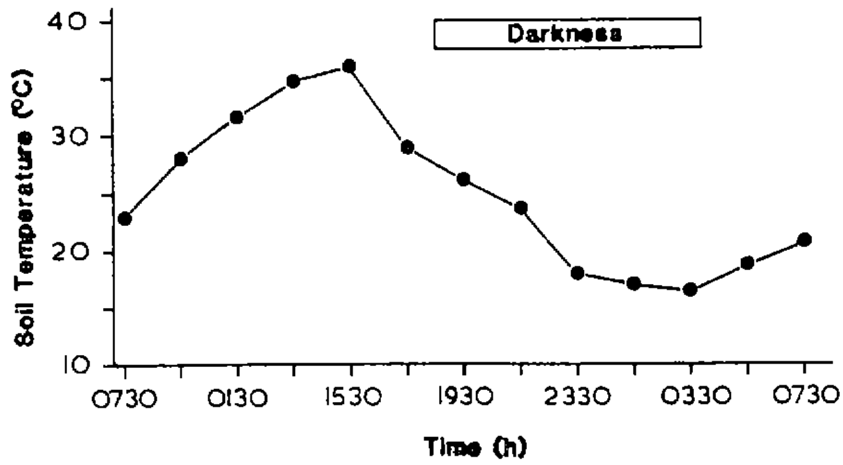


FIGURE 1 Soil Temperature (0°C) for top 1-2 cm of Soil during Seed Taking Trial.

Results

The range in soil temperature during the 24 h seed taking trial is presented in Figure 1.

Fourteen species of ants from nine genera were found in the study plot; six of these species were active seed harvesters (Table 1). Other species may have been present but were insufficiently common or conspicuous to observe. The periods when ants were active is shown in Figure 2. With the exception of **Melophorus** sp. J.D.M. 117 and **Camponotus** sp. J.D.M. 549, which were diurnal, the foraging behaviour of most ant species was mainly nocturnal with foraging extending into the cooler morning and evening (Figures 1 and 2). **Melophorus** sp. J.D.M. 117 was only active during the period when soil temperature was at a maximum (Figures 1 and 2).

Table 1. Ant species recorded during seed taking experiment

Species	Collection frequency
Brachyponera lutea	1
Camponotus sp. J.D.M. 182	1
Camponotus sp. J.D.M. 183	5*
Camponotus sp. J.D.M. 549	2*
Iridomyrmex sp. J.D.M. 200	2
Iridomyrmex sp. J.D.M. nov.	1
Melophorus sp. J.D.M. 117	3*
Meranoplus sp. 2 (ANIC)	1
Meranoplus sp. J.D.M. 400	4*
Monomorium sp. 1 (ANIC)	3*
Rhytidoponera sp. J.D.M. 121	2*
Stigmacros sp. J.D.M. 113	1
Stigmacros sp. J.D.M. 375	1
Tapinoma sp. J.D.M. 134	1

* Known seed harvester

Seed Taking

Thirty-six point five percent, 33 percent and 63.5 percent of the **G. microcarpum**, **A. pulchella** and **B. eriocarpa** seed respectively, were harvested by seed-taking ants (Figure 3). This represents 44.3 per cent of the total seed offered. Most of this seed was taken during the early morning when soil temperatures were moderate (Figures 1 and 3a).

The rate of seed removal for all three seed species was significantly different with respect to each other ($p < 0.05$, Table 2). The order of magnitude of seed removal was; **B. eriocarpa** > **G. microcarpum** > **A. pulchella**. Rank of seed mass, **A. pulchella** > **G. microcarpum** > **B. eriocarpa**, was inversely correlated with the quantity of seed taken.

Fate of Harvested Seed

No marked **G. microcarpum** seeds were recovered from the extracted ant nests. However, two of the four **Melophorus** sp. J.D.M. 117 nests

Table 2. Analysis of Variance and Critical Differences for Seed Removed Over 24 h period.

Species	Difference between means (Scheffe test p 0.05, CD 2.00) A.p.	B.e.
G. microcarpum		
A. pulchella	2.03*	
B. eriocarpa	-10.59*	-12.63*

Analysis of Variance F = 3.17, F p 0.05 3.68

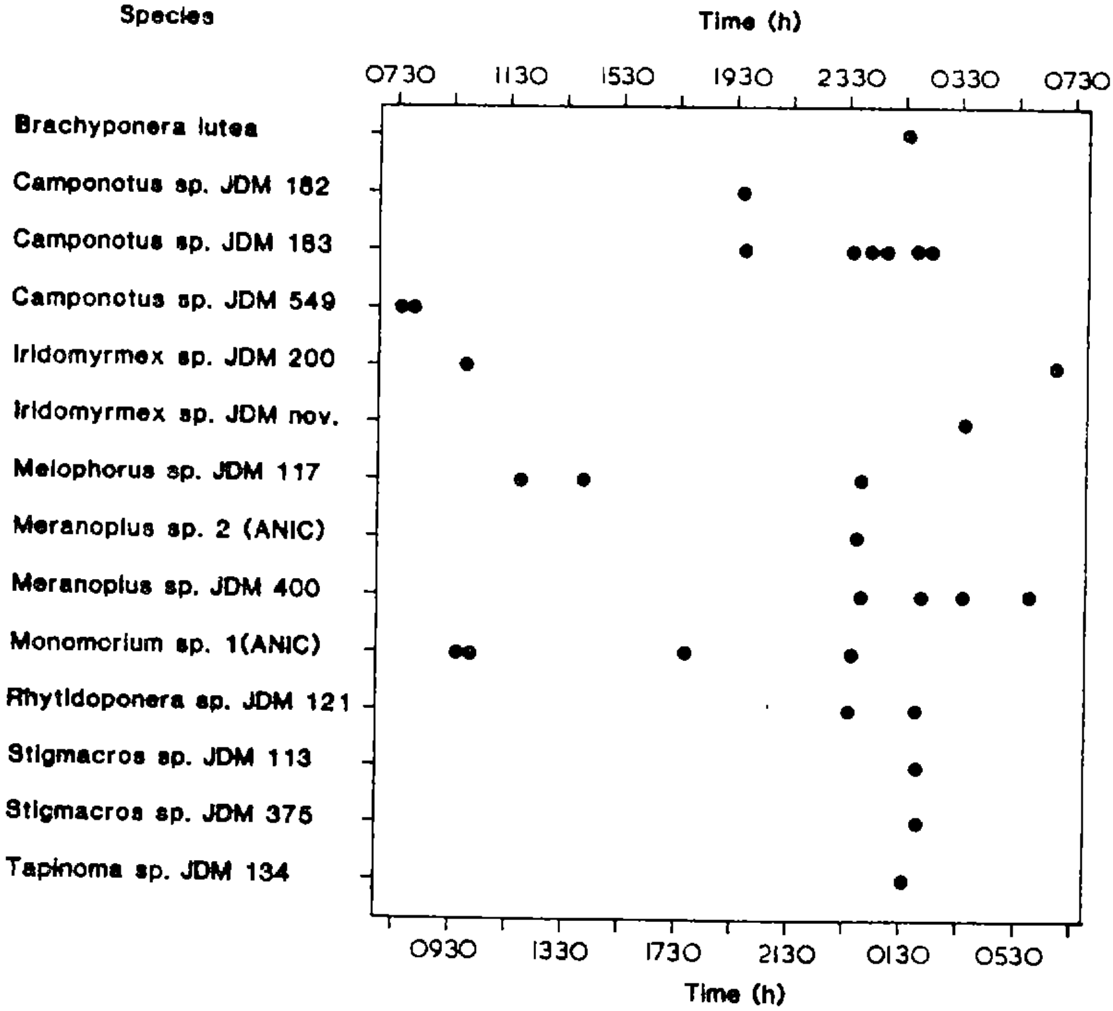


FIGURE 2 Times When the Various Ant Species were Foraging.

examined were found to contain small numbers of G. microcarpum seed (Table 3). Most of these seeds lacked the elaiosome.[4] No seeds of any plant species were recovered from the nest middens.

Table 3. Seeds recovered from inside nests

Nest species	Number of Seed Recovered		
	+ Elaiosome	- Elaiosome	Total
Melophorus sp. J.D.M. 117	2	5	7
Melophorus sp. J.D.M. 117	-	12	12
Melophorus sp. J.D.M. 117	-	-	-
Melophorus sp. J.D.M. 117	-	-	-
Camponotus sp. J.D.M. 182	-	-	-

Discussion

The number of ant species recorded for Dryandra indicates the existence of a populous and relatively species-rich ant community[13]. **Camponotus** sp. J.D.M. 183, **Melophorus** sp. J.D.M. 117, **Meranoplus** sp. J.D.M. 400, **Monomorium** sp. 1 (ANIC) and Rhytidoponera sp. J.D.M. 121 were active seed harvesters. However, due to insufficient observations and because the abundance of seed taking ants in the Narrogin/Katanning area declines during late summer/early autumn[25] it was not feasible to rank seed taking ants.

The level of seed removal over the 24 h period (44 percent) was of similar magnitude to that recorded in Australian pastures and grasslands (61 percent after 3d[21], 35 percent after 24 h[8], 20-30 percent after 24 h[25]) and in Western Australian forests (48-66 percent after 24 h[12]). These figures may well be inflated due to seed being placed in concentrated patches. Naturally spread seed would produce a more dispersed distribution of seeds which would probably be gathered less efficiently by ants.

The influence of soil temperature upon ant foraging activity is well documented[8,17,23]. At Dryandra for example, **Melophorus** sp. J.D.M. 117

was only active when soil temperatures were above 30°C while some species of **Camponotus** which were mainly nocturnal, were only active when temperatures were below 25°C (see Figures 1 and 2).

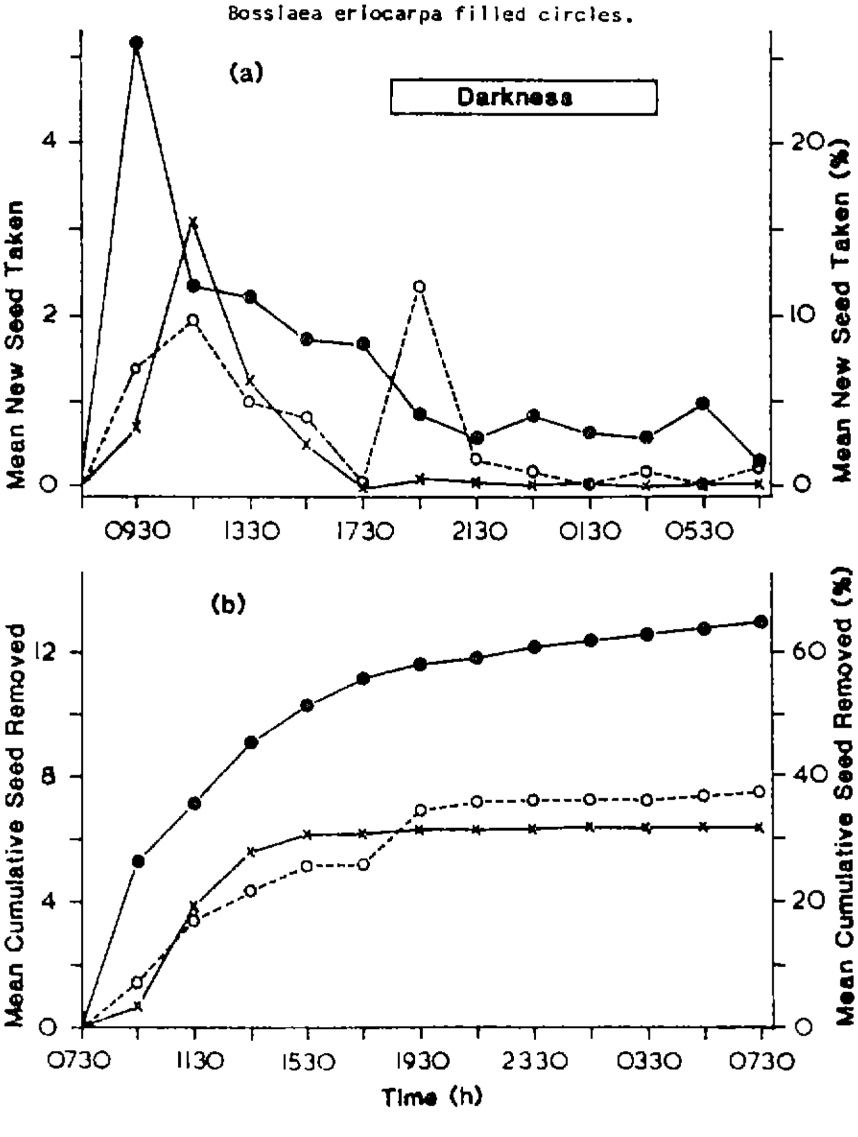
The differences in the rate of seed removal of the three seed species are best explained by seed size selection rather than by the absence or presence of fluoroacetate in the seed. **B. eriocarpa**, the lightest seed, was preferentially harvested. **Monomorium** sp. 1 (ANIC), which is a relatively small ant, appeared to lack the physical ability to remove the larger seed, but was frequently observed removing **B. eriocarpa**. **Monomorium** sp. 1 (ANIC) was also one of the more densely populated species. In Australian grasslands, the relatively small and lighter seeds (such as ryegrass, **Lolium rigidum** seed) have also been shown to be preferentially removed by seed taking ants[15,23]. The lack of selection for, or against, fluoroacetate-bearing seed is further exemplified by the very low Scheffe value when comparing the theft rates of the two larger seed species, **G. microcarpum** and **A. pulchella** (see Table 2).

It is interesting to note that the **G. microcarpum** seed retrieved from the extracted ant nests mainly had elaiosomes removed (see Table 3). While some ants are known to feed exclusively on elaiosomes[4,13] no definitive conclusions on the fate of the elaiosomes on seed extracted from ant nests at Dryandra can be made. These elaiosomes may have been removed by the soil sieving process. The sieving may have also removed the white paint used to mark the **G. microcarpum** seed. This may account for the failure to retrieve marked **G. microcarpum** seed from the extracted ant nests.

Whether fluoroacetate is evenly distributed through the seed and elaiosome, and the effect of weathering upon the fluoroacetate content of

FIGURE 3 Seed Taken During the Trial

(a) Mean new seed taken per depot
(b) Cumulative seed taken
Gastrolobium microcarpum open circles,
Acacia pulchella crosses, and
Bossiaea eriocarpa filled circles.



these components, is not known at present. Fluoroacetate may be less concentrated in the elaiosome or it may be leached more quickly from the fleshy elaiosome than the fluoroacetate of the seed. This could be one of the factors influencing the selection of seed by harvesting ants in areas where toxic plants occur. However, it is more likely that, as with the indigenous mammalian herbivores from the south-west of Western Australia, the invertebrate fauna from areas whose vegetation includes fluoroacetate-bearing plants are less susceptible to fluoroacetate.

G. microcarpum was by far the most dominant plant species in the understorey at Dryandra and as G. microcarpum seed is a nutritious and readily obtainable food source, it is unlikely that the ants would ignore G. microcarpum seed. This implies interaction between the toxic plants and the ants. In order to regularly consume fluoroacetate-bearing seed, the ants would need to have evolved mechanisms for circumventing the toxic manifestations of fluoroacetate. Such mechanisms have been suggested by recent studies on the fluoroacetate sensitivity of Bag Moth larvae (Family: Noctuidae) from Dryandra. Caterpillars of the bag moth have been shown to tolerate 100–200 mg 1080 kg-1 body weight (Twigg, unpublished data) which suggests the invertebrate fauna in areas containing fluoroacetate-bearing vegetation may be less susceptible to fluoroacetate.

The fluoroacetate susceptibility or tolerance of indigenous invertebrate fauna from areas in Western Australia, where toxic species of **Gastrolobium** and **Oxylobium** are present, is currently being investigated. Studies are also being conducted on the concentration and distribution of fluoroacetate within the seed and elaiosome of toxic species of **Gastrolobium**. It is hoped that these investigations may provide answers to the questions posed during the current study.

Summary

Little is known about the influence of fluoroacetate-bearing seed upon the selection of seed by harvesting ants. The response of ants to seed containing fluoroacetate, which were placed out in artificial depots in an **Eucalyptus wandoo** open woodland, suggested that the presence of this substance had very little influence upon the seed selection of harvesting ants. Of the total seed offered, 44.3 percent was removed in 24 h. The principal ant species observed taking seed were **Camponotus** sp. J.D.M. 183, **Melophorus** sp. J.D.M. 117, **Meranoplus** sp. J.D.M. 400, **Monomorium** sp. 1 (ANIC) and **Rhytidoponera** sp. J.D.M. 121. Small numbers of **Gastrolobium** microcarpum (fluoroacetate producing) seed were recovered from two of five extracted ant nests.

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