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OBSERVATIONS ON THE FEEDING PREFERENCES OF TWO SPECIES OF TASMANIAN TERRESTRIAL AMPHIPODS (CRUSTACEA: AMPHIPODA: TALITRIDAE)

by H.P. Morton and A.M.M. Richardson
Department of Zoology, University of Tasmania

(with three tables)

ABSTRACT

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The food preferences of two species of terrestrial amphipod, *Talitrus (Keratroides) vulgaris* and *Talitrus (Keratroides) angulosus*, from near Tooms Lake, Tasmania were investigated. Both species preferred the leaves of *Olearia argophylla* to those of *Eucalyptus obliqua*, decomposed leaves to fresh leaves, and leaves artificially hydrolysed in hydrochloric acid to fresh leaves. Sterilization did not affect their preferences. No differences were detected in the preferences of the two species. These terrestrial amphipods do not appear to be attracted to living bacteria and fungi, but rather to the hydrolytic effect which they have on the leaves.

INTRODUCTION

Terrestrial amphipods are common members of the litter fauna in the wetter forests of Australia, New Zealand, Japan and a number of localities in the Pacific (Bousfield 1982). They share this habitat with a large number of species which often exhibit a low degree of trophic specialization (Anderson 1975). Recent work (Friend 1980) has shown that the amphipod fauna in Tasmanian forests is diverse and that it is not uncommon to find three or four species present together in the litter at a single site. This raises the problem of how these species avoid competition with each other, and although spatial separation has been demonstrated between two Tasmanian species (Friend & Richardson 1977), the possibility that they have different food preferences must be considered.

In temperate forests, part of the litter consists of a collection of leaves in various stages of decay, more or less reflecting the proportions of leaf species in the canopy. The process of decay begins while the leaf is still on the tree (Dickinson & Pugh 1974), but when the leaf reaches the floor the change in microclimate allows a rapid colonization of the surface of the leaf by fungi and bacteria which primes the leaf for attack by macro-arthropods, including amphipods. Laboratory experiments have shown not only clear preferences by mites (Mitchell & Parkinson 1976) collembolans (Visser & Whittaker 1977) and aquatic amphipod species (Barlocher & Kendrick 1973a) for leaves colonised by specific fungi, but also increased growth and fecundity when arthropods are fed on the preferred fungi (Barlocher & Kendrick 1973b, Booth & Anderson 1979). Changes in available nitrogen and phosphorus levels following microbial attack (Satchell & Lowe 1967) may also alter leaf palatability, and therefore the extent of microbial attack.

Thus to state simply that a macro-arthropod feeds on decomposing plant material ignores the subtle differences inevitably present between substrates. When these are recognised, and the great heterogeneity of leaf litter is considered, the apparent similarities in food preferences of coexisting mesofauna may only be superficial.

Friend (1980) studied two species of amphipod in a wet gully near Tooms Lake in eastern Tasmania. The two species, *Talitrus (Keratroides) vulgaris* Friend, 1979 and *Talitrus (Keratroides) angulosus* Friend, 1979, both apparently feed on decaying leaves, preferring these to other components of the litter. This paper records a further investigation of these two species, using animals and food from the same site.

Feeding Preferences of Tasmanian Terrestrial Amphipods

Preferences for partly decomposed leaves over sterile or freshly fallen ones have been recorded for millipedes (Lyford 1943), earthworms (Satchell & Lowe 1967), aquatic caddis fly larvae (Mackay & Kalff 1973) and aquatic amphipods (Kaushik & Hynes 1971). These preferences can be attributed to either the physical presence of microbial communities, or the chemical changes in the leaf resulting from their hydrolytic actions. As well as examining the effect of leaf species on palatability, we have attempted to separate these two effects and to determine their influence on palatability.

METHODS

Several choice experiments of the same basic design were used to assess food preferences. The experiments were performed in Petri dishes with a base of moistened Plaster of Paris. The leaves used were collected from the Tooms Lake site and stored at 10°C until use. Discs of the leaves to be tested (1.5 cm diameter, cut to avoid the mid-rib) were oven dried at 60°C for 48 hours, and weighed individually prior to use. The test substrates were arranged, underside down, around the edge of the dishes. Duplicates of each substrate were placed in each dish, positioned opposite each other to eliminate spatial preferences. All substrates were sprayed with water before the animals were added to the dish.

Approximately ten animals of the selected species were weighed and added to each dish, where they were allowed to feed for 3-5 days at 10°C. The substrates were then removed, dried as before, and reweighed. The weight losses were compared with those from identical discs which had not been exposed to the animals. Consumption was expressed as mg consumed. g $animal^{-1}.day^{-1}$.

In the first series of experiments, the animals were given a choice between the leaves of the two principal tree species at the site, *Olearia argophylla* F. Muell. and *Eucalyptus obliqua* L'Herit., at two stages of decomposition. Freshly fallen leaves (green, rigid, and with little sign of microbial attack) were compared with leaves at an intermediate stage of decomposition (browning but still rigid, with signs of moderate microbial attack). It was not possible to compare leaves at a later stage of decomposition, since the *Eucalyptus* leaves were too fragile to handle. For each amphipod species, ten trials were run in which all test substrates were offered.

To separate the effects of the presence of microbial communities from their catalytic effects, the preferences of the animals for partly decomposed leaves were compared with those for similar leaves autoclaved at 125°C for 1 hour (Howard & Frankland 1974). Fifteen trials were conducted on both amphipod species.

To distinguish between the purely chemical changes in the leaves resulting from microbial attack and structural, biochemical or biological changes, the effect of an artificial hydrolysing agent (hot HCl) on the palatability of *Olearia* leaves and filter paper was examined. Preweighed discs of *Olearia* and filter paper were added to beakers of HCl (1.0N for filter paper, 2.0N for leaves) or distilled water. The treatments used were identical to those used by Barlocher & Kendrick (1975) and the higher concentrations of acid used on the leaves reflects their greater resistance to hydrolysation. The beakers were incubated at 90°C for 4 hours (leaves) or 45 minutes (filter paper). Duplicate beakers with identical contents were left at room temperature. The discs were then rinsed repeatedly in distilled water before use in the experiments. Ten trials, using all substrates (leaves and filter paper treated with hot HCl, cold HCl, hot water and cold water) were made for each amphipod species.

RESULTS

The mean consumption rates of each leaf species at two stages of decomposition are shown in table 1a. A three-way analysis of variance (table 1b) revealed significant preferences for *Olearia* leaves, and partly decomposed leaves. There is a significant interaction between these two factors, which is due to the higher consumption of fresh *Olearia* by *T. vulgaris*.

TABLE 1

EFFECT OF LEAF SPECIES AND STAGE OF DECOMPOSITION ON AMPHIPOD FEEDING RATE.
(a) Effect of leaf species and stage of decomposition on the rate of consumption by the two amphipod species, expressed as mg of leaf consumed.g $animal^{-1}.day^{-1}$ at 10°C, (x±SD), n=10.
(b) Summarised analysis of the variance of the above data.

(a)

Amphipod species	*Talitrus vulgaris*		*Talitrus angulosus*	
Leaf species	*Olearia*	*Eucalyptus*	*Olearia*	*Eucalyptus*
Freshly fallen	45.80±9.49	0.01±0.01	10.59±2.28	0.02±0.01
Partly decomposed	30.13±4.19	27.30±3.71	28.65±3.68	10.23±2.36

(b)

Factor	F	d.f.	Significance
Leaf species	13.46	1,72	P < 0.001
Decomposition stage	5.83	1,72	P < 0.05
Amphipod species	0.03	1,72	NS
Leaf species × decomposition stage	4.39	1,72	P < 0.05

The effect of sterilization on consumption rate is shown in table 2a. Analysis of variance (table 2b) showed that although substrate type did not influence consumption rates significantly, overall consumption by *T. angulosus* was significantly greater than by *T. vulgaris*.

TABLE 2

EFFECT OF LEAF STERILIZATION ON AMPHIPOD FEEDING RATE.
(a) Effect of leaf sterilization on the feeding rate of the two amphipods expressed as mg leaf consumed. g $animal^{-1}.day^{-1}$ at 10°C (x±SD), n=15.
(b) Summarised analysis of variance of the above data.

(a)

K	*Talitrus angulosus*	*Talitrus vulgaris*
Natural leaves	40.76±12.24	31.45±9.50
Autoclaved leaves	43.56±17.65	29.21±13.00

(b)

Factor	F	d.f.	Significance
Substrate type	0.006	1,56	NS
Amphipod species	11.66	1,56	P < 0.01

The mean consumption rates for each of the hydrolysed substrates are shown in table 3a and analysis of variance (table 3b) showed that changes in the composition of the leaves and filter paper produced by hydrolysis significantly increased palatability, but no significant differences in preferences existed between the two amphipod species, or in their preferences for leaves or filter paper.

TABLE 3

EFFECT OF HYDROLISATION ON AMPHIPOD FEEDING RATE
(a) Effect of hydrolisation on the feeding rate of the two amphipod species expressed as mg consumed.g $animal^{-1}.day^{-1}$ at 10°C (x±SD), n=10.
(b) Summarised analysis of variance of the above data.

(a)

Amphipod species	*Talitrus angulosus*				*Talitrus vulgaris*			
Treatment	Hot HCl	Cold HCl	Hot H_2O	Cold H_2O	Hot HCl	Cold HCl	Hot H_2O	Cold H_2O
Olearia leaves	30.18 ±11.45	1.60 ±3.08	0.15 ±0.34	0.01 ±0.01	31.64 ±11.42	4.45 ±8.09	0.66 ±1.59	0.01 ±0.01
Filter paper	28.19 ±8.53	0.40 ±1.26	1.43 ±4.29	0.02 ±0.01	31.30 ±10.09	1.23 ±2.11	3.59 ±6.03	0.15 ±0.46

(b)

Factor			Significance
Substrate treatment	243.45	3,144	$P < 0.001$
Substrate type	0.05	1,133	NS
Amphipod species	1.88	1,144	NS

DISCUSSION

The clear preference of both amphipod species for *Olearia* over eucalypt leaves illustrates the importance of leaf characteristics in determining palatability. In a study of millipede food choices, Lyford (1943) found a strong preference for basswood leaves compared to birch, maple and oak, and although he attributed this to differences in their calcium content, he recognised that also involved were the organic structure of the leaf, the proportion of lignins, cutins and similar structural compounds. Variation in protein and sugar content (Laverack 1960), tannin and polyphenol content (King & Heath 1967), nitrogen and carbohydrate content (Satchell & Lowe 1967) are among other chemical factors implicated in leaf palatability, but the relative importance of these factors undoubtedly varies with the species of macro-arthropod decomposer involved.

Very little work seems to have been done on the preferences of leaf litter arthropods for the leaves of Australian trees. Using a behavioural approach, Devitt (1981) showed that three species of terrestrial amphipod preferred the leaves of *Bedfordia salicina* to those of *Eucalyptus obliqua*. The thick, waxy cuticle of eucalypt leaves, and the high polyphenol content and the presence of complex aromatic oils (Kelly 1969) probably act as a deterrent to macro-arthropod attack. Casual observation in the laboratory suggests that the thickness of the cuticle may be important in determining preferences. Amphipods generally attacked *Bedfordia* leaves from the underside, where the cuticle was thinner, and the material between the veins was consumed in preference to vascular bundles. Since the cuticle of eucalypt leaves covers both sides more or less equally, this must reduce their attractiveness to the animals.

Preference for partly decomposed leaves has been recorded in a number of other macro-arthropod decomposers. Kheirallah (1979) found that the preferences of millipedes for various leaf species changed as the leaves decomposed. Similarly, Lofty (1974) found that the preferences of *Lumbricus terrestris* reversed after leaves had weathered. Dickinson & Pugh (1974) suggested that changes in leaf palatability during decomposition may be attributed to the following factors: (1) decrease in polyphenol content, (2) decrease in C:N ratios, (3) hydrolysis of surface sugars, and (4) the presence of bacterial and fungal colonies. Since leaves will differ in their structure and chemical composition, the decomposition process will act differentially on leaf species.

Two major mechanisms are involved in leaf conditioning by micro-organisms: (1) the conversion of plant tissue into microbial tissue, and (2) the partial digestion of plant tissue into sub-units digestible by macro-arthropod grazers (Barlocher & Kendrick 1975). Either, or both, of these mechanisms may be instrumental in explaining the preference of most grazers for "conditioned" leaves.

Sterilizing techniques have been used previously to assess the importance of microbial production in determining preferences for "conditioned" leaves. The effects of autoclaving may not be confined simply to killing the microbial colonies, since Howard & Frankland (1974) also noted chemical changes in autoclaved leaf litter. However, table 3 shows that autoclaving leaves did not significantly alter their palatability. In similar experiments, Kaushik & Hynes (1971) and Mackay & Kalff (1973) found that the detritus feeders which they studied preferred natural to autoclaved leaves. This was interpreted as confirmation of the importance of microbial production in determining leaf palatability, leaf attractiveness being influenced by microbial growth, not just the physical presence of microbial communities.

This conclusion cannot be supported from the present study, where it appears that amphipods do not discriminate between leaves with viable microbial colonies and those with dead colonies. Therefore, either the physical presence of micro-organisms, or their catalytic effects, influence, at least partially, the palatability of leaves to terrestrial amphipods. The results presented in table 2 suggest that hydrolytic effects not only change leaf composition, but increase palatability, even in the absence of living microbial colonies. Hydrolysis by HCl can only represent one of the processes enhancing palatability which micro-organisms bring about during the decomposition of leaf material (Dickinson & Pugh 1974), and this does not totally remove the possibility that the presence of microbial colonies also influences the attractiveness of leaves. Barlocher & Kendrick (1975) demonstrated that HCl hydrolysis greatly increased the palatability of leaves and filter paper to the aquatic amphipod, *Gammarus pseudolimnaeus*.

This study has failed to demonstrate any differences between the dietary preferences of *Talitrus vulgaris* and *T. angulosus*. Since *T. angulosus* is found in the fermentation layer of the soil-litter system (Friend & Richardson 1977), it might have been expected to show a preference for leaves at a later stage of decomposition. The absence of any such preference does nothing to alter the suggestion (Friend & Richardson 1977, Friend 1980) that it is competition for habitat space which has brought about the niche separation between the two species.

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