# Field evaluation of the « Bait-lamina test » to determine the soil microfauna feeding activity

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**Key words:** soil microfauna feeding activity assessment, forage plants, methodology, soil biological activity, soil animals, field experiment.

#### Abstract

The soil microfauna regulate nutrient cycling through predation on soil microorganisms but also through comminution of organic residues in soils. The feeding activity of the soil microfauna has a large impact on nutrient cycling and soil function, but is rarely considered because it is difficult to assess. The Baitlamina test was proposed as a practical mean to assess microfauna feeding activity. The test consists of vertically inserting 16-hole-bearing plastic sticks stuffed with a plant material preparation into the soil. We tested if the plant material used to prepare the bait would be differentially utilized by microfaunal population present under different plant species or mixtures of plants. We evaluated the Bait-lamina test in a 5-year old field experiment with five levels of plant communities (monocultures of Russian wild rye, switchgrass, green needlegrass, or western wheatgrass, and a grass mixture) distributed in four complete blocks, using six levels of bait flavour (Russian wild rye, switchgrass, green needlegrass, western wheatgrass, alfalfa, and wheat bran). Bait-lamina strips were equally spaced at five locations between plant rows. We found that the bait flavour had no significant (P = 0.22) effect on feeding, although the concentration of crude protein in the plant material used in the baits varied (P = 0.006) from 2.9% in switchgrass to 5.9% in Russian wild rye. We found low feeding activity in our field plots over the period of the test (13 June to 17 August 2005), with only 2.7% of the lamina (hole stuffing) showing signs of feeding. The data nevertheless revealed that microfauna feeding was more important close to the soil surface (0.5 mm deep), and in Russian wild rye plot mid-rows as compared to green needlegrass or switchgrass plots. Closer to plant rows, however, differences were not significant. We conclude that bait prepared with any plant material used in this study can be used to compare microfauna feeding in different plant stands. We recommend the use of a large number of replicated strips in agricultural field experiments where the microfauna may be scarce.

#### Introduction

The soil is a complex environment where plant, microorganism and microfaunal interactions largely determine the chemical environment of the Earth. Understanding soil processes may require the consideration of both soil microorganisms and microfauna. A « bait-lamina test » has been proposed as

a simple way to assess microfaunal activity in soil (von Törne, 1990). The test consists of vertically inserting 16-hole-bearing plastic strips stuffed with a plant material preparation into the soil. After a period of time, the bait-lamina strips are removed and examined for evidence of feeding. Hellig et al. (1998) have shown high feeding activity of Collembola and enchytraeids using bait-lamina strips in controlled experiments, and a reduced impact of microorganisms on the disappearance of the lamina, they also demonstrated the preference of the organisms for bait made with nettle tissues to bait made with wheat bran, as proposed by von Törne (1990).

We evaluated the bait lamina test in an agricultural soil growing different forage grasses in 5-year old field plots. In particular, we wanted to know if the plant material used to prepare the bait would be differentially utilized by the microfaunal population established under different plant communities.

# Methods

## Site description

The experiment was conducted on a Brown Chernozem (Haverhill clay loam) located at the South Farm of the Semiarid Prairie Agricultural Research Centre in Swift Current Saskatchewan, Canada (latitude: 50° 17' N; longitude: 107° 41' W). The region receives 361 mm of annual precipitation and has a yearly mean temperature of 3.6°C, with mean monthly temperatures ranging from -13.2°C in January to 18.6°C in June (54-year averages). Precipitation in 2005 were close to normal (285.4 mm from Jan to August, as compared to a 54-year average of 282.1 mm) with amounts above normal in June (123.2 mm, normal 73.2 mm) and below normal in July (21.4 mm, normal 52.0 mm). Air temperature from June to September averaged 15.8°C i.e., 1.4 °C below normal.



Fig. 1. Experimental set up in one plot. One row of bati-famina strips (as shown in the circle) is an experimental unit. Each of six rows contains one different bati flavour. Sings in other rows were used to determine the most appropriate removal time.

# Experimental design

The Bait-lamina method was evaluated in 5-year old field plots (4 m by 8m) planted with monocultures of Russian wild rye (*Elymus junceus* Fisch.), switchgrass (*Panicum virgatum* L.), green needlegrass (*Stipa viridula* Trin.), or western wheatgrass (*Agropyron smithii* Rydb.), and a mixture of western wheatgrass, green needlegrass, little bluestem (*Schizachyrium scoparium* Michx.) and switch grass. Each plant community treatment was arranged in four complete blocks. Six bait flavour treatments were factorially arranged and randomized into each plot. Five bait-lamina strip replicates constituted one experimental unit. These five bait-laminae were inserted in the soil on 13 June 2005 using a modified chefs knife. They were placed between two plant rows, evenly spaced along an imaginary line laid perpendicular to plant rows (Fig. 1). Four supplementary bait-lamina strips of each flavour were also inserted in the soil of each plot. They were harvested at different time intervals to monitor feeding and to determine the incubation time most appropriate for the test,

under our field conditions. Experimental bait-lamina were removed from the soil on 17 August 2005, just prior to forage harvest. The number of lamina showing feeding activity were counted and the location of the hole on the bait-lamina strips recorded.

# Preparation of the bait-laminae

Bait materials were prepared by mixing ground dry plant tissues with agar-agar (Anachemia Canada Inc., Montreal Qc), cellulose powder and bentonite (Sigme-Aldrich Inc., St.Louis MO) in a 1/6.5/1.5/1

proportion calculated on a weight basis. Several applications of the bait preparation were required to load the laminae, as the material would shrink. The recipe was modified by excluding bentonite in later applications. The plant tissue from the 2004 harvest was used to prepare the bait. This material was analysed for: N by the Kjeldahl method (McGill and Figueiredo, 1993), P according to Murphy and Riley (1962), and crude protein by multiplying Kjeldahl N by 6.25. The percentage of N, P and crude protein in the plant materials is presented in Table 1. Alfalfa and wheat bran analyses are not available.

Table 1. Analysis of the plant material used to make the bait, which was material harvested from the test plot the previous year. Means are significantly different according to an ANOVA protected (P <0.007) LSD test at P = 0.05; n = 20.

	N%	Р%	Crude protein%	
Russian wild rye	0.95 a	0.078 b	5.9 a	
Western wheatgrass	0.88 ab	0.088 ab	5.5 ab	
Green needlegrass	0.72 bc	0.095 a	4.5 bc	
Switchgrass	0.47 d	0.090 ab	2.9 d	
Mix	0.79 b	0.085 ab	5.0 b	
Average	0.76	0.087	4.8	

Table 2. Percentage of bait strips holes fed upon in at different distance from plant rows, in plots with different plant species. Means are significantly different according to LSD P = 0.05; n = 20.

	Position in the inter-row						
	1	2	3	4	5	Average	
Russian wild rye	3.0	3.9	4.3 a	4.0	1.9	3.4	
Western wheatgrass	1.4	3.8	2.3 ab	2.8	2.6	2.6	
Green needlegrass	0.8	2.6	1.4 b	2.1	2.6	1.9	
Switchgrass	3.3	2.3	1.4 b	3.2	1.4	2.3	
Mix	2.9	3.0	3.9 ab	4.6	2.3	3.3	
Average	2.1	2.9	2.7	3.5	2.1		

Mix: western wheatgrass + green needlegrass + little bluestem + switchgrass

Inc).

#### **Results and discussion**

Microfauna feeding activity was extremely low with 2.7% of lamina showing signs of feeding after 62 days in the soil. This level of feeding is comparable to that observed by von Törne

#### Statistical analysis

Very few laminae showed signs of feeding and the data was summed and parsed to analyse different effects. The effect of plant community treatments on microfauna feeding was assessed using one-way ANOVA conducted on the data parsed by inter-row positions and summed over the 16 depths of each bait-lamina strips. . Positions 3,4 and 5 required a ln+1 transformation of the data prior to analysis, which was done using JMP v. 3.2.6 (SAS Institute). Kruskal-Wallis tests were conducted to assess bait flavour effects on microfauna feeding within each plant communities treatments as well as the overall bait flavour effect. It was also used to assess the impact of soil depth on feeding across all treatments. The non-parametric Kruskal-Wallis analyses were conducted using Systat v. 10 (SPSS



Fig 2. Variation in feeding activity with depth across all treatments. P < 0.0005; N = 20.

(1990) after 12h of incubation in German forest soils. Feeding activity was higher closer to the soil surface (Fig. 1) suggesting that organisms walking on the soil surface were important factor of residue comminution in our agricultural soil system.



Fig. 3. Percentage of holes eaten in different bait flavour treatment within different plant community treatments. WWG, western wheatgrass; SG, switchgrass; RWR, Russian wild rye; GNG, green needlegrass; Mix, WWG + GNG + little bluestem + SG. N = 4. We found no effect (P < 0.22) of bait flavour on feeding activity across plant community treatments (data not shown) or any significant bait flavour by plant community interaction (Fig. 2). The data was variable, however, and there was a trend for greater feeding on switchgrass flavoured bait in Russian wild rye plots (P = 1.5). In contrast to our observation, Hellig et al. (1998) report a preference of two collembola and two enchytraeids for bait made with nettle powder over bait made with wheat bran. Feeding on wheat bran bait was variable in our study, with trends showing that this bait flavour was the most fed upon in the mixed plant community and the least fed upon in the green needlegrass community. Wheat bran bait flavour does not seem to be the most appropriate bait material.

Feeding on the baits was more important (P < 0.06) directly in between plant rows (position 3, Table 2) in stands of Russian wild rye (RWR) suggesting that the microfaunal community was larger in RWR midrows than in green needlegrass and switchgrass midrows (Table 2). The increased feeding in midrows of RWR may also be a response to the lateral rooting pattern of this plant. Russian wild rye was the better source of protein of all the grasses (Table 1).

In conclusion, the bait flavour did not influence microfauna feeding activity significantly and did not interact with plant community treatments, suggesting that lamina of all flavours are eaten similarly in the different plant communities. We detected more feeding close to the soil surface and in Russian wild rye plots using the Bait-lamina test in forage stands grown in a semiarid climate. Variability was high in our data, however, and the number of replicates per plot should be increased to improve the precision of the test.

## Acknowledgement

Thanks to D. Wiebe for his perseverance in loading the bait-lamina strips and in data acquisition.

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