

Impact of Subterranean Termites (Blattodea: Isoptera: Rhinotermitidae) on Tallgrass Prairie Forage Grass Growth

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Abstract - Previous studies investigated subterranean termite ecology on The Nature Conservancy's Joseph H. Williams Tallgrass Prairie Preserve near Pawhuska, northeastern Oklahoma. However, these studies did not examine the impact of termites on growth of prairie grasses. This study evaluated the impact of termites on leaf and stem mass of forage grasses as well as root mass over a 12-month growing season. Four field sites were established, each consisting of four blocks of land with each block sub-divided into two plots. Using baiting systems, termites were eliminated from one plot in each block, establishing termite-free plots. From plots with actively foraging termites (non-baited 'control' plots) or without termites (baited), 64 groups of mixed grass leaf and stem samples, and 64 groups of mixed grass root samples were collected and differences in their mass compared at 12 months after termite elimination from baited plots. Leaf and stem measurements showed that above-ground mass within plots containing actively foraging termites was significantly greater compared with plots with no termites. However, root mass was similar among plots with and without termites. Results indicate subterranean termite foraging activity within a native tallgrass prairie is beneficial to prairie forage grass growth.

Introduction

Termites are important soil-dwelling reducer-decomposers of cellulosic and woody materials. As soil engineers they influence their ecosystem by building nests and foraging galleries above and below ground. Termites use their saliva, excrement, soil, and organic material (leaves, grass, and woody material) to form mounds, mud tubes, galleries, carton nests and nest chambers (Jouquet et al. 2011). Nests consist of enclosed galleries and chambers with a few openings to the outside (Noirot and Darlington 2000). Subterranean termites in the genus *Reticulitermes* Holmgren connect below-ground galleries to above-ground mud tubes (Abe 1987, Mizumoto and Matsuura 2013, Shellman-Reeve 1997). Mud tubes and galleries in soil constructed by worker termites provide protected passageways for foraging above and below ground. Soil transported to build these passageways alters soil properties and affects water infiltration rates as well as diversity of animals, plants, and soil microbes (Jouquet et al. 2011, Mo and Stroosnijder 1996).

Termites also play a significant role in breaking down dead cellulosic material and altering chemicals in the soil. They adjust their feeding behavior depending on food quality, seasonal changes, and nutritional needs of an expanding colony (Collins 1984; Eggleton et al. 1996, 1997; Jones et al. 1998). Soil obtained from termite workings and mounds contains fine loam particles and exchangeable cations Ca^{++} , K^{+} , Mg^{++} , and Na^{+} , with greater concen-

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trations of organic matter (OM) compared with termite-free soils (Folgarait 1998, Jouquet et al. 2006, Lobry de Bruyn and Conacher 1990, Myer and Forschler 2018). Increased nutrients and improved water infiltration rates caused by termite foraging aid plant growth and success of plant communities in arid, semi-arid, and tropical environments (Blomqvist et al. 2000, Dean et al. 1997, King 1977, Woodell and King 1991). Nutrients added to soils worked by termites elicited increased growth of grasses as well as tomato plants and orchids (Flores-Palacios and Ortiz-Pulido 2005, Garba et al. 2011, Jouquet et al. 2005, Mo and Brussaard 1999).

In this study, the objective was to evaluate the impact of subterranean termite foraging and feeding on mass of forage grasses on The Nature Conservancy's Joseph H. Williams Tallgrass Prairie Preserve (TGPP) in northeast Oklahoma. Changes in above-ground, mixed grass leaf and stem mass, and changes in mixed root mass between field plots without subterranean termite activity (baited plots) were compared with field plots with known continuous termite activity (non-baited 'control' plots) at the end of a 12-month growing season. Study plots contained native grasses that were the only available food resources for foraging termites. No forb, woody shrub or tree components were present in the grass samples collected. Only mixed samples of the four predominant TGPP grasses were evaluated during this 12-month study. Because termite feeding on plant roots stimulates plant growth, termite foraging in the soil would cause an increase in forage for prairie animals including *Bison bison* [L 1758] (American bison), demonstrating that these small soil-dwelling insects can be beneficial for large animals (Garba et al. 2011, Jouquet et al. 2005, Mo and Brussaard 1999). Using standard termite keys and DNA analysis, termites foraging in the grass study plots were identified as *Reticulitermes flavipes* Kollar and *Reticulitermes tibialis* Banks (Brown et al. 2005, Gleason and Koehler 1980, Schefrahn and Su 1994, Smith et al. 2010).

Materials and Methods

Tallgrass Prairie Preserve

The TGPP in Osage County is located 18.5-km north of Pawhuska in northeast Oklahoma (Brown et al. 2009). It encompasses 15,659 ha consisting mainly of native tallgrass prairie grasses interspersed with occasional woody plants and is home to ~2,500 bison. The four predominant grasses on the TGPP are *Andropogon gerardii* Vitman (Big Bluestem), *Schizachyrium scoparium* Michx. (Little Bluestem), *Panicum virgatum* L (Switchgrass), and *Sorghastrum nutans* [L] Nash (Indiangrass) (Brown et al. 2009; Hurt 1999; Smith et al. 2022a, 2022b). Groups of approximately equal mixtures leaves and stems, and approximately equal mixtures of roots of these four grasses in each sample as collected intact from field sites were weighed and compared for this study.

Field sites and plots

Four rectangular field sites, each measuring 130.0 x 50.0 m, were established within bison-excluded native grass areas. Each site was partitioned to contain four rectangular blocks of land, each measuring 50.0 x 10.0 m. Each block was partitioned to contain two plots, each measuring 10.0 x 10.0 m with one plot located at each end of its block and marked with a 0.5-m-tall wooden stake. Termite colonies on the TGPP have a maximum linear foraging distance of 19.0 m (Brown et al. 2008). To ensure termites from one plot were not overlapping into adjacent plots, each plot was separated by 30.0-m buffer zone (Fig. 1).

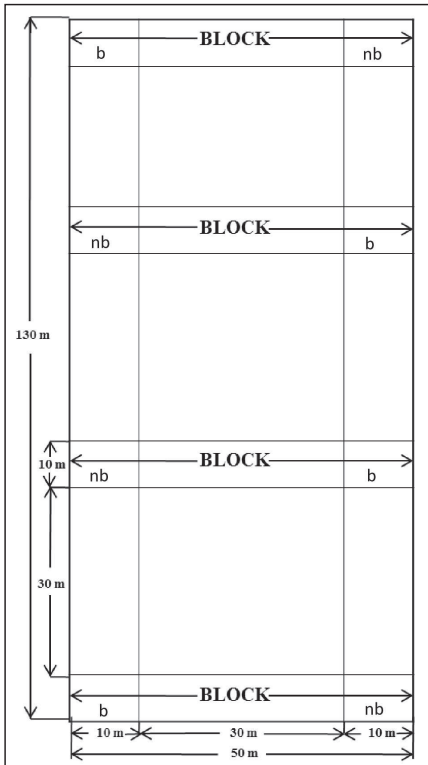


Figure 1. Each of four field sites consisted of four blocks of land, each containing two 10- x 10-m square plots separated by a 30-m buffer zone. One plot per block was randomly chosen for bait emplacement and the opposite plot designated a non-baited control. b = baited; nb = not baited.

Bait systems

One of the two plots in each block was randomly selected for termite elimination using a bait system. Baited plots in field sites 1 and 4 each contained ten Sentricon® Stations, each with a Recruit® IV Baitube® device containing the insect growth regulator (IGR) noviflururon (Dow AgroSciences LLC, Indianapolis, IN). Baited plots in field sites 2 and 3 each contained ten Advance® termite bait stations containing the IGR diflubenzuron (BASF, Research Triangle Park, NC; Fig. 2). Both these well-established baiting systems have proven

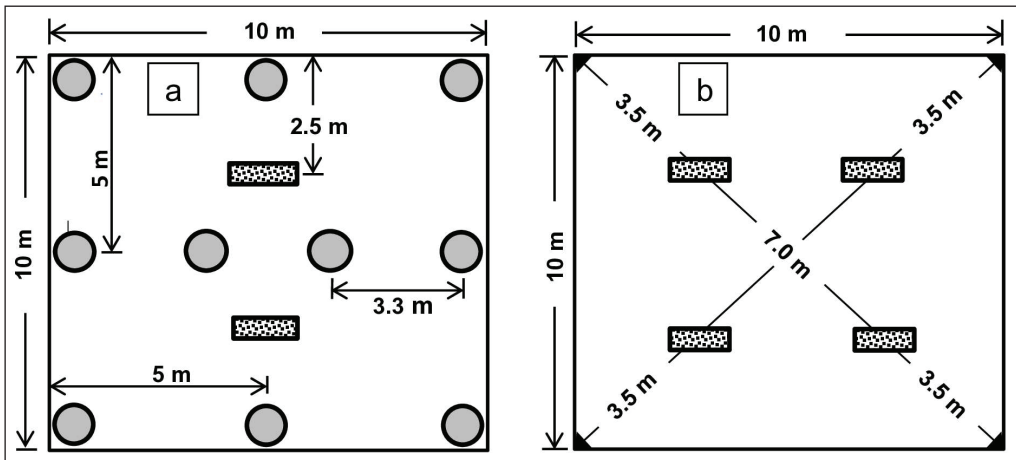


Figure 2. a) Baited plot with two soil-surface termite-monitoring ground-boards and ten cylindrical active-bait stations. b) Non-baited control plot with four soil-surface termite-monitoring ground-boards.

effective for eliminating termite colonies (Keefer et al. 2015, Kistner and Sbragia 2001, Su et al. 1991). Each bait station was covered with a 2.4-L inverted metal bucket stabilized with a standard building brick placed on top. The bucket protected the plastic bait station from melting during natural TGPP wildfires and range management prescribed burns, gave protection from inclement weather, and marked a station location in tall grass. Bait stations were regularly checked for termite activity. Noviflumeron and diflubenzuron have no herbicidal activity noted on their labels or in the toxicity sections of their safety data sheets and are not known to directly affect plant growth (DowAgrosciences 2011, 2015; EPA 1997; OSHA 2022). However, termite baiting systems would be expected to indirectly affect grass growth by eliminating termites that would otherwise be feeding on grass stems, leaves, and roots.

Monitoring devices

Pine (*Pinus* spp. L) soil-surface ground boards (GB) measuring 30.5 x 15.2 x 2.5 cm were used to monitor termite activity. Vegetation was removed where each ground-board was placed directly on bare mineral soil surface. A standard building brick was placed on each GB to hold it in place and ensure firm contact with the soil surface (Brown et al. 2004). In baited plots, two GBs were centrally placed 3.3 m apart to monitor for unlikely return termite activity and ten bait stations were installed to eliminate termites (Fig. 2a). In non-baited control plots, one GB was placed at each corner of a 4.7-m square centered in each plot (Fig. 2b). GBs remained in control plots only until termite activity was confirmed and then were removed to avoid providing an alternative food source that could affect the amount of feeding on grass stems, leaves, and roots. However, GBs remained in baited plots to confirm continued absence of termites.

Plant mass

A total of 128 separate samples were collected during late summer 12 months after elimination of termites from baited plots. A total of 64 mixed grass leaf and stem samples and 64 mixed grass root samples were collected from all 32 field plots for the four field sites. The collection event consisted of 16 above-ground leaf and stem samples and 16 root samples from both termite-free and termite-active plots in each of the four field sites. Two leaf plus stem samples and two root samples were collected from random locations in each plot, resulting in 32 samples for each of the four field sites. Leaf and stem samples were collected within a circular sampling ring (enclosing 0.25-m² inside area) within a different quadrant of each plot to preclude sampling any location twice. Sampled ‘spots’ were marked to avoid future sampling overlap. All above-ground grass mass within the sampling ring was excised at the soil surface using sharp steel-bladed shears, placed into a labeled container, and brought to the laboratory to be dried and weighed.

Using a 10.2-cm inside diameter x 20.3-cm-tall (1,658.8 cm³ volume) cylindrical soil auger, root samples were extracted from each plot quadrant, placed in a labeled container, and brought to the laboratory for processing. Each root sample was thoroughly rinsed with water to remove soil, blotted with a paper towel to remove excess water, and returned to its open container for oven drying. All leaf and stem samples, and root samples were oven-dried at 80°C for 24 hours and dry masses obtained.

Data analyses

Analyses were performed with SAS Version 9.4 (SAS 2016). Analysis of variance (PROC MIXED) was used to assess the effect of treatment and time. A repeated measures model was utilized in a randomized block design with site as block and time as the repeated

factor. An autoregressive period-1 covariance structure was used to model the intra-site variation, with means and standard errors reported.

Results and Discussion

There were significant differences in mixed leaf plus stem mass between baited and non-baited plots, suggesting that foraging termites have a positive effect on plant growth ($\alpha = 0.0262$; Fig. 3). Average dry mass of mixed grass leaves plus stems collected from plots with actively foraging termites was 15.6% greater compared with mixed grass leaves plus stems samples collected from termite-free plots, demonstrating the benefit termites had on increasing forage grass mass on the TGPP (Smith et al. 2022a, 2022b). Both bait systems were effective in eliminating termite activity in their plots.

Research conducted on termite mounds in other countries reported increased plant growth due to termite tunneling and mound building. Additional studies show that increases in leaf and stem growth are influenced by termites foraging within the root zone, stimulating a growth response beneficial to plants (Duponnois et al. 2005; Folgarait et al. 2002; Holt and Lepage 2000; Jouquet et al. 2004, 2006; Lobry de Bruyn and Conacher 1990). Termite soil excavation and construction activities increase chemical nutrients and OM and improve water infiltration into soil (Brown et al. 2009, Garba et al. 2011, Jouquet et al. 2005, Mo and Stroosnijder 1996). Throughout this study, Osage County experienced “severe” to “extreme” drought conditions (USDM 2012, 2013), which would cause termites to tunnel deeper into the soil seeking greater moisture conditions. Deeper termite tunneling activity can increase water infiltration to deeper roots, as well as deliver soil nutrients to drought-stressed plants (Jouquet et al. 2011, Mo and Stroosnijder 1996).

There was no significant difference in root mass between baited and control plots ($\alpha = 0.2342$; Fig. 4). However, average mixed grasses dry root mass extracted from plots with

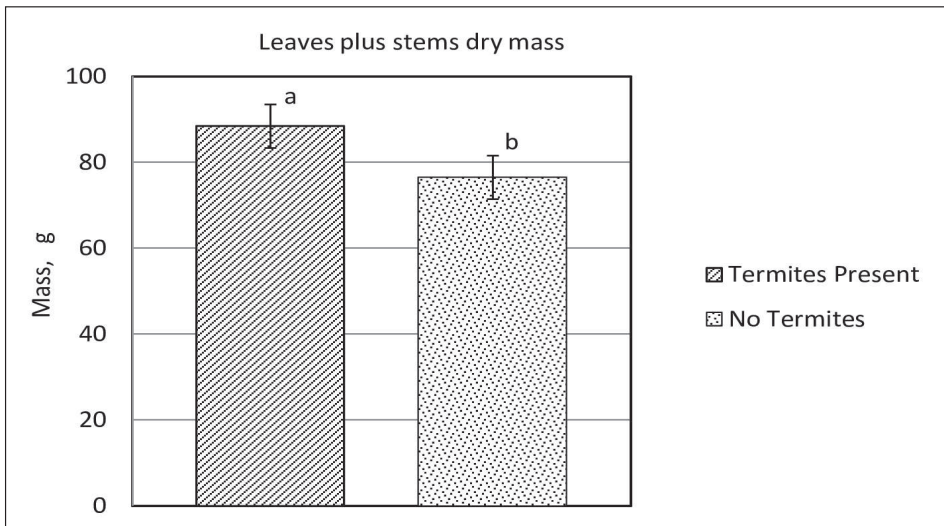


Figure 3. Above-ground mixed leaves plus stems dry mass comparison between non-baited control plots with active termite foraging and termite-free baited plots at the 12-month evaluation. Mean \pm SE ($\alpha = 0.0262$).

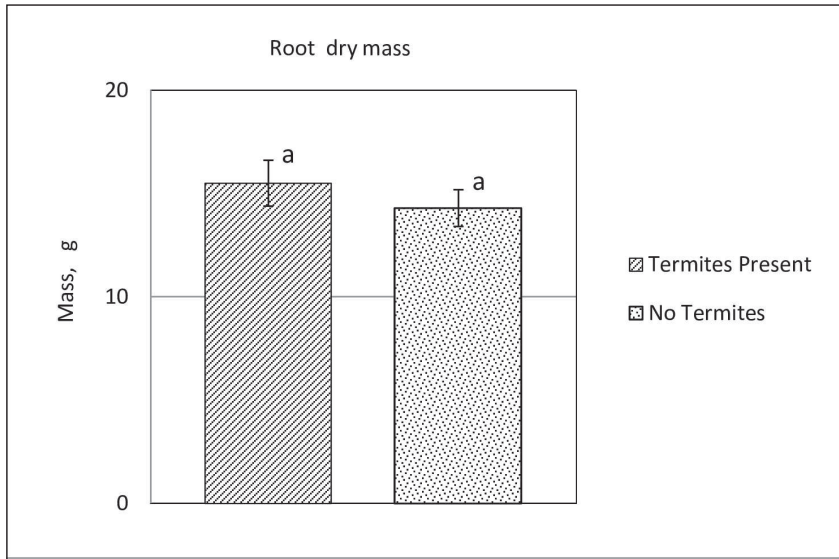


Figure 4. Mixed roots dry mass comparison between non-baited control plots with active termite foraging and termite-free baited plots at the 12-month evaluation. Mean \pm SE ($\alpha = 0.2342$).

actively foraging termites was 8.4% greater compared with root samples extracted from termite-free plots, indicating a positive effect due to termite presence. Root mass may have showed no significant difference between baited and control plots because any increased root growth could have been counter-balanced by removal of root tissue by termites feeding on deeper roots due to the drought. Because this study was only 12 months in duration, a longer-term study may produce a different result as termites continue to forage on grass roots, possibly causing root mass differences between treatments over more time.

In a concurrent laboratory study that compared termite feeding preferences between the four most predominant TGPP grasses, *R. tibialis* preferred roots over leaves and stems of some grasses in both choice and no-choice tests (Smith 2016). Termites were also observed feeding inside stems of Big Bluestem and Indiangrass but with the exterior sheath of the grasses remaining intact. This interesting behavior provided a protected high humidity environment for foraging termites. Additional research is needed to evaluate the extent and diversity of these foraging and feeding behaviors.

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A.L. Smith, M.P. Smith, B.M. Kard, and M.E. Payton

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