Arthropods on Terminalia argentea (Combretaceae) Fertilized with Sewage Sludge

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Arthropods on *Terminalia argentea* (Combretaceae) fertilized with sewage sludge

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Abstract

We tested the effects of dehydrated sewage sludge fertilization on leaf mass production and ground cover of *Terminalia argentea* Mart. & Zucc. (Combretaceae) trees. In addition, we determined the abundance, diversity, and species richness of phytophagous Hemiptera, Sternorrhyncha predators (Hemiptera), and protocooperating ants visiting those plants. Numbers of leaves per branch, branches per tree, percentage of ground cover, abundance, species richness of phytophagous Hemiptera and Sternorrhyncha predators, including diversity of protocooperating ants, were greater on *T. argentea* trees fertilized with dehydrated sewage compared with unfertilized trees. We conclude that the addition of the dehydrated sludge aided the recovery of the severely disturbed area by increased fertilization of *T. argentea* trees used in the restorative process and increased the associated arthropod fauna.

Key Words: insect diversity; ground cover; plant fertilization; restoration ecology

Resumo

Testamos os efeitos da adubação com lodo de esgoto desidratado na produção de massa foliar e cobertura do solo por árvores de *Terminalia argentea* Mart. & Zucc. (Combretaceae). Além disso, determinamos abundância, diversidade e riqueza de espécies de Hemiptera fitófago, predadores de Sternorrhyncha (Hemiptera) e formigas protocooperantes visitando aquelas plantas. O número de folhas para galho, galhos para árvore, porcentagem de cobertura do solo, e a abundância e riqueza de espécies de Hemiptera fitófago e predadores de Sternorrhyncha incluindo diversidade de formigas protocooperantes foram maiores em árvores de *T. argentea* fertilizadas com lodo de esgoto desidratado comparado com árvores não adubadas. Concluímos que a adição do lodo de esgoto desidratado auxiliou na recuperação da área severamente perturbada pelo aumento da fertilização das árvores de *T. argentea* utilizadas no processo restaurador e aumento da fauna de artrópodes associados.

Palavras Chave: diversidade de insetos; cobertura do solo; fertilização de plantas; ecologia da restauração

Sewage sludge, a residue rich in organic matter, has potential as a fertilizer or substrate in seedling production. This material often is used in forest plantations and in the restoration of severely disturbed ecological areas, reducing the risk of toxic elements entering the human food chain (Kimberley et al. 2004). This material, when applied to agriculture and forests, is a sustainable, low-cost alternative to reduce the use of chemical fertilization and environmental pollution (Caldeira et al. 2014, 2018; Silva et al. 2016). For example, the heavy metal content in soil where maize grains, *Zea mays* L. (Poaceae), and cowpea, *Vigna unguiculata* (L.) Walp. (Fabaceae), had been fertilized with dehydrated sewage sludge did not differ with areas not fertilized in this manner (Nogueira et al. 2007).

Terminalia argentea Mart. & Zucc. (Combretaceae) is a secondary growth deciduous tree that can attain heights up to 8 m, and is native to southeastern and central western Brazil. This tree species is widely used in landscaping, wood and coal production, civil construction, and for severely disturbed ecosystem recovery (Lorenzi 2002). Exudates, continuously released at different trunk heights by this tree, attract beneficial insects such as *Trigona branneri* (Cockerell) and *Mesembrinella bicolor* (Fabricius) (both Hymenoptera: Apidae) (Boff et al. 2008).

Insect diversity, especially Hemiptera and Hymenoptera species (González & Ruiz 2000; Skern et al. 2010), are considered indicators of habitat recovery because these organisms respond quickly to environmental change (Fernandes et al. 2010). Sewage sludge is rich in macro- (e.g., N, P, K) and micronutrients (Cu and Zn) (Mass 2010) that increase humus content in the soil, favoring plant and consequently insect development. Moreover, abundance and diversity of herbivorous insects and natural enemies generally are greater on large trees with their numerous and longer branches that provide greater canopy height and width (Ferrier & Price 2004; Espírito-Santo et al. 2007; Leite et al. 2017).

The aim of our study was to test the effects of dehydrated sewage sludge fertilization on leaf mass production and ground cover of *T. argentea* trees during a 24 mo period. Additionally, the abundance,

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diversity, and species richness of phytophagous Hemiptera, Sternorrhyncha predators, and protocooperating ants was determined in this ecosystem. The hypotheses tested were: (i) *T. argentea* plants fertilized with dehydrated sewage sludge have larger canopies, and therefore produce more ground cover; and (ii) the abundance of phytophagous Hemiptera, Sternorrhyncha predators, and protocooperating ants are greater on trees fertilized with sludge.

Materials and Methods

EXPERIMENTAL SITE

The study was conducted in a severely disturbed area in the Instituto de Ciências Agrárias of the Universidade Federal de Minas Gerais in Montes Claros, Minas Gerais State, Brazil (16.860555°S, 44.916666°W; 943 masl) from Mar 2015 to Feb 2017. The study area has been severely disturbed with pronounced soil loss. The climate of the area is tropical and dry; total annual rainfall is about 1,000 to 1,300 mm, with dry winters and mean yearly temperature of ≥ 18 °C according to Köppen's classification (Alvares et al. 2013). The soil type is litolic neosoil (Santana et al. 2016), and its chemical and physical characteristics have been described previously by Silva et al. (2020).

EXPERIMENTAL DESIGN

Terminalia argentea seedlings were produced from seeds of trees grown at the Instituto de Ciências Agrárias of the Universidade Federal de Minas Gerais. Seedlings were planted during Mar 2014 in plastic bags (8 × 12 cm) (Bom Cultivo Company, Rio do Sul, Santa Catarina, Brazil) in a nursery with a mixed substrate containing 30% organic compost, 30% clay soil, 30% sand, and 10% reactive natural phosphate (160 g per seedling). Organic fertilizer consisted of 2:1 parts of garden pruning debris (≤ 5 cm) and dried manure obtained from Nelore cattle (*Bos taurus indicus* L.; Bovidae). The soil pH in the planting holes on site (40 × 40 × 40 cm) was corrected with dolomitic limestone (90% relative total neutralization power = 187 g per hole), increasing base saturation to 50% (Kopittke & Menzies 2007). Natural phosphate (80 g per hole), fritted trace elements (10 g per hole), and marble rock dust (1 kg per hole) were added according to prior soil analysis. Plants were transplanted on flat terrain at the recovery site during Sep 2014. One 30 cm tall *T. argentea* seedling was planted per hole with a 2 m spacing between them. Six parallel transects of 8 trees each were planted with 2 m between each line (total 48). Twenty liters of dehydrated sewage were added to the planting hole of 4 plants, whereas 4 were not fertilized and were used as controls. Assignment of fertilized and unfertilized plants within transects was completely random. Seedlings were watered twice per wk until the beginning of the rainy season (Nov 2015). Each plant was pruned with a sterilized razor when its branches reached 5 cm long. Additional branches were removed including those up to one-third of crown height, leaving only the best one intact. The pruned parts of each plant remained on the ground between their respective planting lines. Numbers of leaves per branch, branches per tree, and percentage of ground cover by leaf litter, grass, and herbaceous plants in the canopy projection of each tree was evaluated visually once per mo (24 observations).

DEHYDRATED SEWAGE SLUDGE

Dehydrated sewage sludge (5% moisture content) was collected at the Estação de Tratamento de Esgoto treatment plant in the municipality of Juramento, Minas Gerais State, Brazil, about 40 km away from the *T. argentea* experimental site. The Minas Gerais Sanitation Company operates the Estação de Tratamento de Esgoto – Companhia de Saneamento de Minas Gerais S.A., treating 217 m^3 sewage per day. The efficiency of the system for organic matter removal is greater than 90%. Treatment of the sludge is carried out by solarization in coarse sand tanks for 3 mo in the Estação de Tratamento de Esgoto that reduces the thermotolerant coliforms to levels acceptable by the National Council for the Environment, Conselho Nacional do Meio Ambiente (Resolution N° 375) of the Ministry of the Environment, Ministério do Meio Ambiente of Brazil for its use in agriculture; which is < than 10^3 of the most likely number per g of total solids. The main chemical and biological characteristics of this dehydrated sewage sludge have been described previously by Nogueira et al. (2007).

INSECT ABUNDANCE, DIVERSITY, AND RICHNESS ON *TERMINALIA ARGENTEA*

Insects were counted visually on each tree during biweekly visits between 7:00 AM and 11:00 AM from Mar 2015 to Feb 2017. At that time, 12 adaxial and abaxial leaf surfaces per tree were observed in the apical, middle, and basal canopy at each cardinal direction. At the time of observations, trees were 6 mo of age. Insects were not removed from trees during counting, although some specimens were collected for identification. At least 3 specimens per species were captured with an aspirator for this purpose. These insects were stored in glass flasks with 70% ethanol or mounted, separated by morphospecies, and sent for identification by specialists. The total sampling effort consisted of observing 27,648 leaves during the study.

STATISTICS

Ecological indices (abundance, diversity, and species richness) were calculated per identified insect from trees fertilized with or without dehydrated sewage sludge using the software BioDiversity Professional, vers. 2 (McAleece et al. 1997). Abundance and species richness are the total number of individuals and species in a sampling unit, respectively (Scheiner 2003). Diversity was calculated using the Hill formula (first order): N1 = exp (H'), where H' is the Shannon-Weaver diversity index (Hill 1973). Mean data of leaves per branch, branches per tree, percentage of ground cover, abundance, diversity, and species richness of phytophagous Hemiptera, Sternorrhyncha predators, and protocooperating ants were subjected to the non-parametric Wilcoxon signed-rank test ($P < 0.05$) (Wilcoxon 1945) because the data did not have a normal distribution. This analysis used the software program Sistema para Análises Estatísticas e Genéticas, vers. 9.1 (Saeg 2007) (Supplier: Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil).

Results

TERMINALIA ARGENTEA PRODUCTION

The numbers of leaves per branch, branches per tree, and the percentage of ground cover (i.e., leaf litter, grasses, and herbaceous plants) were significantly greater for *T. argentea* trees fertilized with dehydrated sewage sludge than those without. The sewage sludge almost doubled leaf production per branch with a 6-fold increase in the number of branches per *T. argentea* and twice as much ground cover (Table 1).

INSECT ABUNDANCE, DIVERSITY, AND SPECIES RICHNESS

Mean abundance and species richness of phytophagous Hemiptera, Sternorrhyncha predators (Hemiptera), including the diversity of protocooperating ant species, were statistically greater ($P < 0.05$) on trees fertilized with sewage sludge than without fertilization (Table 1). Also, phytophagous *Aphis spiraecola* (Patch) (Hymenoptera: Aphidi-

Table 1. Mean ± SE leaves per branch, branches per tree, percentage of ground cover, abundance, diversity, and species richness of phytophagous Hemiptera, Sternorrhyncha predators, and protocooperant ants per *Terminalia argentea* fertilized with dehydrated sewage sludge or unfertilized (control) in a severely disturbed area of the Instituto de Ciências Agrárias of the Universidade Federal de Minas Gerais in Montes Claros, Minas Gerais State, Brazil.

	Sewage sludge		Wilcoxon test	
	Fertilized	Control	VT*	*P*
Leaves per branch	11.47 ± 0.34	6.38 ± 0.23	5.9	< 0.01
Branches per tree	28.88 ± 0.51	4.48 ± 0.22	5.9	< 0.01
Ground cover (%)	11.98 ± 0.63	5.39 ± 0.31	4.2	< 0.01
Abundance of phytophagous Hemiptera	21.04 ± 8.46	2.79 ± 1.17	3.1	< 0.01
Diversity of phytophagous Hemiptera	1.60 ± 0.52	0.80 ± 0.22	0.7	0.26
Species richness of phytophagous Hemiptera	1.83 ± 0.28	0.58 ± 0.16	3.4	< 0.01
Abundance of Sternorrhyncha predators	2.96 ± 0.47	0.33 ± 0.11	5.4	< 0.01
Diversity of Sternorrhyncha predators	0.52 ± 0.19	0.42 ± 0.13	0.1	0.46
Species richness of Sternorrhyncha predators	1.38 ± 0.11	0.29 ± 0.09	5.2	< 0.01
Abundance of protocooperant ants	28.67 ± 3.98	5.08 ± 0.18	5.2	< 0.01
Diversity of protocooperant ants	6.57 ± 0.71	3.78 ± 0.48	3.0	< 0.01
Species richness of protocooperant ants	3.96 ± 0.20	2.08 ± 0.23	4.7	< 0.01

n = 24 per treatment; *VT = value of the test.

dae), *Mahanarva fimbriolata* Stål (Hemiptera: Cercopidae), Fulgoridae, Membracidae, and Pentatomidae were more abundant on fertilized trees. Sternorrhyncha predators *Chrysoperla* sp. (Neuroptera: Chrysopidae), *Cycloneda sanguinea* L. (Coleoptera: Coccinelidae), Dolichopodidae (Diptera) and *Photinus* sp. (Coleoptera: Lampyridae) similarly were more abundant on trees treated with sewage, as well as protocooperating ants *Brachymyrmex* sp. and *Camponotus* sp. (both Hymenoptera: Formicidae) ($P < 0.05$) compared with unfertilized trees. Abundance of protocooperating ants *Ectatoma* sp., *Pheidole* sp., and *Pseudomyrmex termitarius* Smith (all Hymenoptera: Formicidae) was significantly greater ($P < 0.05$) on fertilized trees compared with unfertilized trees (Table 2).

Discussion

We found the greatest leaf mass production and ground cover occurred on *T. argentea* trees fertilized with dehydrated sewage sludge

Table 2. Mean ± SE phytophagous Hemiptera and natural enemies per *Terminalia argentea* tree fertilized or unfertilized (control) with dehydrated sewage sludge in a severely disturbed area of the Instituto de Ciências Agrárias of the Universidade Federal de Minas Gerais in Montes Claros, Minas Gerais State, Brazil.

			Sewage sludge		Wilcoxon test	
Order	Family	Species	Fertilized	Control	VT*	*P*
Hemiptera	Aleyrodidae	Non-identified	0.54 ± 0.42	1.92 ± 1.07	0.9	0.18
	Aphididae	*Aphis spiraecola*	16.71 ± 8.46	0.13 ± 0.12	2.4	< 0.01
	Cercopidae	*Mahanarva fimbriolata*	0.21 ± 0.13	0	1.8	< 0.04
	Cicadellidae	*Balclutha hebe*	0	0.08 ± 0.05	1.4	0.08
		Cicadellinae	0.08 ± 0.08	0	1.0	0.16
		Erythrogonia sexguttata	0	0.08 ± 0.05	1.4	0.08
	Coreidae	*Leptoglossus* sp.	0.04 ± 0.04	0	1.0	0.16
	Cicadidae	*Quesada gigas*	0.13 ± 0.06	0.04 ± 0.04	1.0	0.15
	Fulgoridae	Non-identified	2.17 ± 0.61	0.50 ± 0.30	3.3	< 0.01
	Membracidae	Non-identified	0.13 ± 0.06	0	1.8	< 0.04
		Membracis sp.	0.04 ± 0.04	0	1.0	0.16
	Pentatomidae	Non-identified	0.58 ± 0.20	0.04 ± 0.04	2.4	< 0.01
	Pseudococcidae	*Phenacoccus* sp.	0.42 ± 0.41	0	1.0	0.16
Coleoptera	Coccinelidae	*Cycloneda sanguinea*	0.21 ± 0.13	0	1.8	< 0.04
	Lampyridae	*Photinus* sp.	0.33 ± 0.15	0	2.3	< 0.01
Diptera	Dolichopodidae	Non-identified	2.38 ± 0.46	0.29 ± 0.11	4.5	< 0.01
	Syrphidae	*Syrphus* sp.	0.17 ± 0.07	0.04 ± 0.04	1.4	0.08
Hymenoptera	Braconidae	Non-identified	0.08 ± 0.05	0	1.4	0.08
	Formicidae	*Brachymyrmex* sp.	5.13 ± 1.05	0.54 ± 0.18	4.7	< 0.01
		Camponotus sp.	10.63 ± 1.76	2.17 ± 0.49	4.0	< 0.01
		Cephalotes sp.	0.04 ± 0.04	0	1.0	0.16
		Ectatoma sp.	2.29 ± 0.85	0.13 ± 0.06	3.3	< 0.01
		Pheidole sp.	6.38 ± 2.01	1.25 ± 0.33	3.1	< 0.01
		Pseudomyrmex termitarius	4.21 ± 0.99	1.00 ± 0.46	3.2	< 0.01
Neuroptera	Chrysopidae	*Chrysoperla* sp.	0.21 ± 0.08	0	2.3	< 0.01

n = 24 per treatment; *VT = value of the Wilcoxon test.

compared with unfertilized trees. Also, higher abundance and species richness of phytophagous Hemiptera, Sternorrhyncha predators, and protocooperating ants was noted; this may be due to the high number of leaves produced per tree by the fertilization with dehydrated sewage sludge, because it is rich in macro- and micronutrients (Nogueira et al. 2007; Jakubus 2016), thus improving nutritional plant quality (Taiz et al. 2017) for arthropod colonization. Our results support the hypothesis that trees with higher leaf mass production and greater associated free amino acids would attract more sucking insects, and thus consequently Sternorrhyncha predators and protocooperating ants. For example, Leite et al. (2011) and Taiz et al. (2017) reported that increased development of phytophagous Hemiptera occurred on plants with higher rates of nitrogen fertilization or lower levels of potassium; this may be due to the greater amino acid content of the phloem. Additionally, Ferrier & Price (2004), Espírito-Santo et al. (2007), and Leite et al. (2016, 2017) stated that higher diversity and abundance of herbivorous insects and their natural enemies occurred on larger size trees compared with smaller trees.

We believe that the greatest abundance and species richness of arthropods on *T. argentea* fertilized trees improves the quality of using this species in the restoration of severely disturbed areas. This agrees with the reports of increased macrofauna richness associated with sewage sludge fertilization in severely disturbed areas of the Cerrado in Brazil (Kitamura et al. 2008) and in Oxford, Ohio, USA (Larsen et al. 1996). However, it should be noted that the use of exotic species in restoration, such as the neem, *Azadirachta indica* A. Juss. (Meliaceae), can negatively affect invertebrate fauna.

We observed a greater percentage of ground cover (i.e., leaf litter, grasses, and herbaceous plants) directly under the crown of fertilized *T. argentea* trees. This may be due to the greater number of leaves (> 1.8 times) and branches (> 6.6 times) produced from these trees compared with unfertilized controls. Ground cover is important for reducing laminar soil erosion and increasing fertility (Franco et al. 2002) in environmentally damaged habitats. Our results support the hypothesis that fertilization with dehydrated sewage sludge improves tree growth and ground cover, assisting in the restoration of ecologically damaged areas. As previously stated, dehydrated sewage sludge is rich in organic matter, containing macronutrients such as nitrogen and phosphorus as well as the micronutrients copper and zinc that favor tree development (Jakubus 2016). The recovery of severely disturbed areas is usually slow (Ferreira et al. 2007), but fertilization with dehydrated sewage sludge is promising (Lorenzi 2002) and may allow for shorter recovery periods. The positive impact of dehydrated sewage sludge on tree development has been reported for several species, including *Acacia mangium* Willd. (Fabaceae), *Senna spectabilis* (DC.) Irwin & Barneby (Fabaceae), *Araucaria angustifolia* (Bertol.) Kuntze (Araucariaceae), *Cedrela fissilis* Vell. (Meliaceae), *Eucalyptus grandis* W. Hill ex Maiden (Myrtaceae), and *Lafoensia pacari* St.-Hil. (Lythraceae) (Kimberley et al. 2004; Silva et al. 2011, 2020; Abreu et al. 2017). We conclude that the addition of the dehydrated sewage sludge aided the recovery of the severely disturbed area by increased fertilization of *T. argentea* trees used in the restorative process while increasing arthropod abundance and diversity in the ecosystem.

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