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Soil invertebrates in Australian rain gardens and their potential roles in storage and processing of nitrogen

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23 **Highlights**

- 24 ▪ Rain garden soil invertebrate communities differed spatially but not temporally
- 25 ▪ Oligochaetes and springtails accounted for 80% of total soil invertebrate abundance
- 26 ▪ Earthworm abundance was positively correlated to soil organic matter content
- 27 ▪ Estimated soil invertebrate N storage and gas emissions were low but estimated ingestion
- 28 rates were high
- 29 ▪ Intersite differences in invertebrate functional effects may be temporally stable

30 **Keywords**

31 Biofilter, bioretention system, low impact development, LID, Lumbricidae, WSUD

32 **Abstract**

33 Research on rain gardens generally focuses on hydrology, geochemistry, and vegetation. The
34 role of soil invertebrates has largely been overlooked, despite their well-known impacts on soil
35 nutrient storage, removal, and processing. Surveys of three rain gardens in Melbourne, Australia,
36 revealed a soil invertebrate community structure that differed significantly among sites but was
37 stable across sampling dates (July 2013 and April 2014). Megadrilacea (earthworms),
38 Enchytraeidae (potworms), and Collembola (springtails) were abundant in all sites, and
39 accounted for a median of 80% of total soil invertebrate abundance. Earthworms were positively
40 correlated to soil organic matter content, but the abundances of other taxonomic groups were not
41 strongly related to organic matter content, plant cover, or root biomass across sites. While less
42 than 5% of total soil N was estimated to be stored in the body tissues of these three taxa, and
43 estimated N gas emissions from earthworms (N₂O and N₂) were low, ingestion and processing of
44 soil was high (e.g., up to 417% of the upper 5 cm of soil ingested by earthworms annually in one

site), suggesting that the contribution of these organisms to N cycling in rain gardens may be substantial. Thus, invertebrate communities represent an overlooked feature of rain garden design that can play an important role in the structure and function of these systems.

1. Introduction

Rain gardens (also known as biofilters and bioretention systems) are small, terrestrial natural treatment systems designed to filter pollutants from stormwater using porous filter media planted with one or more species of vegetation (Ambrose and Winfrey 2015; Askarizadeh and others 2015). Soon after rain garden construction, soil invertebrate communities develop (Ayers 2009) that may impact important rain garden functions such as infiltration and nutrient retention or removal (Levin and Mehring 2015; Mehring and Levin 2015). Though it has yet to be tested in the context of rain gardens, substantial amounts of carbon (C), nitrogen (N) and phosphorous (P) may be stored in soil invertebrate biomass (Teuben and Verhoef 1992), which temporarily immobilizes nutrients and prevents them from being leached. Soil invertebrates may also impact nutrient retention indirectly through increasing nutrient uptake by plants. The effects of earthworm activity on plant uptake of N are particularly dramatic, with some species reported to enhance uptake > 200% in vertical-flow wetlands (Xu and others 2013). Springtails, millipedes (Diplopoda), and isopods also have the potential to enhance plant uptake of N due to high levels of plant-available N in their fecal material (Anderson and others 1983; Teuben and Roelofsma 1990).

Soil invertebrates may play an important role in nutrient removal in rain gardens as well as nutrient immobilization/retention. Earthworms, for instance, have the potential to increase nitrate removal via denitrification because anoxic conditions within their guts favor production of dinitrogen (N₂) and nitrous oxide (N₂O) by ingested soil-derived microbes (Horn and others

2006), even when surrounding soil conditions are aerobic. Indeed earthworm casts themselves may be denitrification hotspots, prolonging the effects of "worm facilitated" N removal long after excretion (Parkin and Berry 1994).

Despite their notable effects on soil biogeochemistry, few studies to date have quantified soil invertebrates in rain gardens. Those studies that have, emphasize above-ground invertebrates (Kazemi and others 2009a; Kazemi and others 2009b), leaving within-soil diversity (and biogeochemical roles) in rain gardens largely unknown. Here we present results from one of the first studies of below-ground invertebrate communities in rain gardens. Our study took place in July 2013 and April 2014 in Melbourne, Australia, and was designed to (1) assess spatial and temporal patterns in invertebrate community structure within rain gardens (2) identify drivers of invertebrate abundance within raingardens (e.g., vegetation type, vegetation cover, and soil organic matter content), and (3) determine if soil invertebrates are likely to contribute substantially to nutrient retention/removal within rain gardens (based on the nutrient content in their biomass, soil processing capacity, and estimates of their contribution to denitrification).

2. Methods

2.1 Study sites – Three rain gardens near Melbourne, Victoria (Australia) were sampled in the winter of 2013 (July) and autumn of 2014 (March-April): (1) “Hereford Road” (HR) is a 100-m² rain garden that treats runoff from a 0.93-ha peri-urban catchment in the town of Mt. Evelyn; (2) “Wicks Reserve” (WR) is a 1900 m² rain garden that treats runoff from a 11.43-ha peri-urban catchment in the town of The Basin; and (3) “Lynbrook Boulevard” (LB) is a bioretention swale that treats runoff from a 2.0-ha suburban development known as Lynbrook Estate. More

information on these rain gardens can be found in Supplementary Material (Supplementary Material, Table S2).

2.2 Field Sampling Methods – Samples were collected at points evenly-spaced along a transect, from the inlet (where stormwater flows into the rain garden) to the outlet (where excess water leaves the rain garden). At least four points were sampled in each site per sampling season, with 9, 10, and 13 points sampled in total at LB, WR, and HR, respectively. At each sampling location, a 0.25m x 0.25m quadrat was used to determine the percent cover of three commonly-planted vegetation types: grasses, sedges, and rushes. Filter media samples (top 5 cm of soil media) were collected from within the quadrat using a plexiglass corer 5 cm in diameter. Filter media cores were fixed in 10% phosphate-buffered formalin within 8 hours of collection, and shipped to the University of California, San Diego (UCSD) for analysis of invertebrate community structure and filter media organic matter content. Additional sub-samples of filter media were collected and composited into glass sample jars for future nitrogen content analysis. Composite samples came from two depths (< 2 cm and ~ 10 cm). All composites were frozen within 8 hours of collection and stored at -20°C prior to analysis.

2.3 Laboratory Methods - Formalin-preserved filter media were rinsed over nested sieves to separate invertebrates, organic matter, roots, and inorganic matter into three size fractions: 1) > 2 mm, 2) 0.3 - 2 mm, and 3) 0.045–0.3 mm. Soil size fractions $\geq$ 0.3 mm were sorted under a Wild M5A stereomicroscope at 12x magnification in order to remove all invertebrates, which were then classified according to order, suborder, or family, and enumerated. The most abundant invertebrates (Oligochaetes, including Megadrilacea [earthworms] and Enchytraeidae [potworms]; and Collembola [springtails]) were dried and individual weights were measured in

order to estimate biomass per site, body tissue N content, and ingestion rates. Tissue N content and ingestion rates by oligochaetes were estimated using conversion factors (see Appendix S2). Following removal of invertebrates and roots, each filter media size fraction was dried to a constant weight at 60°C, combusted at 500°C, and re-weighed in order to estimate soil organic matter content as ash-free dry mass (AFDM). Frozen composite filter media samples (2 depths) were sent to a NATA accredited laboratory (<http://www.nata.asn.au/>) for analysis of total N using standard methods and quality assurance procedures (APHA 2012).

2.4 Statistical Analysis - Overall invertebrate community composition was explored using multivariate analyses (MDS, ANOSIM, SIMPER) run using PRIMER 6 (Primer-E 2006, Plymouth Marine Laboratory, Clarke 1993, Clarke & Warwick 1994) on fourth-root transformed, unstandardized data (data provided in Supplementary Material, Table S1). Using the same software, patterns of species richness among rain gardens were compared by using a sample-based rarefaction procedure ('DIVERSE', 'Rarefaction'), where number of individual invertebrates collected are set as samples and estimated species richness is calculated for a given sample size. Taxonomic richness was estimated repeatedly for increasing sample sizes in each site, at increments of 5 individuals, until the total number of invertebrates collected within a given site was reached. If an asymptote in expected taxonomic richness is not reached for a given site, it suggests that actual taxonomic richness is higher than that estimated from the sampling effort.

Two-factor Analysis of Variance (ANOVA) was used to examine differences in abundance of earthworms, potworms, springtails, and Acari (mites) among sites and between

sampling dates (seasons). Linear regression was used to test for correlations between abundance of these taxonomic groups and plant cover, filter media organic matter, and root biomass.

The contribution of invertebrate communities to nine important rain garden functions (plant growth, water infiltration, plant pathogen removal, denitrification, nutrient uptake in plants, nutrient storage in soil, heavy metal uptake by plants, coarse organic matter shredding, and decomposition) was estimated as follows. Briefly, the average abundance (from this study) of each taxonomic group capable of performing a function (Supplementary Material, Table S1) was multiplied by a biomass correction factor based on body size. These values were summed and then multiplied by the number of contributing taxa present in our samples, positively weighting richness. The resultant scores were binned logarithmically so that function scores spanning multiple orders of magnitude could be compared (see supplementary material, Appendix S1 for calculations).

A Monte Carlo framework (Mehring and others [2015]) was used to further evaluate a subset of the above-noted invertebrate functions in rain gardens concerning nitrogen. Specifically, we quantify (a) the percentage of total soil N in the tissues of earthworms, potworms, and springtails, (b) the amount of soil media ingested by dominant soil invertebrates (earthworms and potworms) in a single year, and (c) areal rates ($\text{m}^{-2} \text{h}^{-1}$) of N_2O and N_2 emission from earthworms in each rain garden (see Appendix S2 for calculations). This approach required 1) our measurements of taxon-specific abundance and biomass from each rain garden; 2) parameter ranges from literature for the tissue N content of earthworms (Schmidt and others 1999), potworms (Pandian and Marian 1985), and springtails (Chahartaghi and others 2005); 3) soil ingestion rates by earthworms (Curry and Schmidt 2007) and potworms (McBrayer and Reichle 1971); and 4) N_2O and N_2 emission by earthworms (Depkat-Jakob and others 2012;

Horn and others 2006; Karsten and Drake 1997; Matthies and others 1999). Note that our range of earthworm soil ingestion rates is limited to values from sandy soils and mull. Because Monte Carlo techniques were employed, our final estimates of invertebrate tissue N content, soil ingestion, and NO₂ and N₂ emission for each rain garden reflect both variability in literature parameters and our empirical measurements of abundance and biomass. These analyses were conducted using R software (R Development Core Team 2008).

3. Results

3.1 Spatial and Temporal Patterns in Invertebrate Community Structure - Analysis of 32 soil cores revealed diverse communities of soil invertebrates (Table S1). The highest invertebrate abundance was at WR (Fig. 1A), the highest biomass was at HR, reflecting the prevalence of large earthworms (Fig. 1B), and the highest taxonomic richness was found at LB, the oldest site (Figure 1C). Multivariate analyses of invertebrate assemblages show significant community differences among rain gardens (Fig. 1D, Table 1, ANOSIM, all $p < 0.05$), but not sampling dates, the exception being HR (Table 1, ANOSIM, $p = 0.014$). Community structure at LB diverged most strongly from the structure observed at other rain garden sites (SIMPER, > 68% dissimilar to other sites in Winter 2013 and Autumn 2014) (Fig. 1D, Table 1).

The most abundant invertebrates sampled across all rain gardens were potworms, earthworms, springtails, and mites (Table S1, Fig. 1A), which accounted for a median of 80% of total invertebrate abundance. However, significant inter-site differences in taxonomic group abundance were observed. For instance, although oligochaetes in the family Naididae were among the most abundant organisms at WR, they were not detected at other sites (Table S1). Potworm abundance was also significantly higher at WR, while earthworm abundance was

higher at HR (Tukey HSD, all $p < 0.05$). Neither earthworm nor potworm abundances changed significantly between sampling dates, and relationships among rain gardens were stable over time (no site $\times$ date interaction). Springtail abundance, however, was significantly higher in Autumn 2014 than in Winter 2013 ($F_{1,26} = 5.47$, $p < 0.05$), with no significant differences among sites. Mite abundance was the most stable, and did not vary significantly among sites or over time.

3.2 Correlates of Invertebrate Abundance Within Rain Gardens - Across all sites, earthworm abundance was positively correlated with fine organic matter (e.g., the percentage of total soil dry weight composed of organic particles 0.045-2 mm in size) ($t_{1,30} = 2.91$, $R^2 = 0.22$, $p < 0.01$). Mite, springtail, and potworm abundance across sites could not be explained by variations in soil organic matter content, plant cover, or root biomass. However, a site-specific positive correlation between potworm abundance and root biomass was observed at LB ($t_{1,7} = 2.95$, $R^2 = 0.55$, $p < 0.05$), and WR ($t_{1,8} = 2.23$, $R^2 = 0.39$, $p = 0.056$).

3.3 Invertebrates and Rain Garden Function – Potential invertebrate community contributions to rain garden function were highest in HR (Fig. 2), followed by WR and LB. Soil invertebrates in HR had high function scores (in decreasing order) for decomposition, coarse organic matter (e.g. leaf litter, wood) fragmentation, nutrient uptake into plants, plant growth, and water infiltration. Scores for similar functions were elevated in WR and LB, but to a lesser degree. An exception is coarse organic matter fragmentation, which had relatively low scores at WR and LB, due to limited abundance (or absence) of several litter transforming taxa (sensu Lavelle 1997). Total soil N concentrations in HR, LB, and WR were 5100, 2900, and 460 mg kg⁻¹ at < 2 cm depth, and significantly lower (1400, 1000, and 140 mg kg⁻¹) at 10 cm depth (paired $t_2 = 4.42$, $p <$

0.05). Our estimates suggest that <5% of depth-averaged total soil N was contained within the body tissues of earthworms, potworms, and springtails > 0.3 mm in size. The contribution of body tissues to soil N was particularly low at LB (< 0.05%).

Median estimates of areal N₂O and N₂ emissions from earthworms were low (max of 0.12 µg N₂O-N m⁻² h⁻¹ and 0.23 µg N₂-N m⁻² h⁻¹), and decreased as follows: HR > WR > LB (Fig. 3A). The median estimate of soil ingestion (expressed as the percent total soil mass ingested annually by earthworms and potworms in the upper 5 cm of soil) was also higher at HR than at WR (43%, range: <1 - 417% y⁻¹ vs 4%, range: <1 - 28% y⁻¹), and negligible at LB (<1%, range: 0-3% y⁻¹) (Fig. 3B). Note that estimates as high as 137% y⁻¹ fell within the interquartile range at HR. Because preservation in 10% formalin can cause reductions in dry mass of invertebrates (Leuven and others 1985), our estimates of biomass, and therefore N storage, N₂ and N₂O emission, and soil ingestion rates are likely to be conservative..

4. Discussion

The findings presented here suggest that invertebrate community structure may differ substantially among rain gardens within a region, and that these differences are fairly stable over short (~9 month) time scales. Apart from a positive correlation between earthworm abundance and soil organic matter content (22% of the variation explained), we were unable to isolate factors correlated with the abundance of the most common soil taxa across sites. However, typical annual rainfall was higher at WR and HR than at LB (1100, 1000 and 750 mm, respectively) and the presence of aquatic organisms (Chironomidae, Ostracoda, Naididae) at WR, and desiccation-intolerant terrestrial amphipods (Maraldo and others 2009; Spicer and others 1987) at HR, suggests that soil moisture may contribute to inter-site variability in

invertebrate abundance. In addition, particle size distributions, which were originally dominated by gravel at LB, may have influenced early development of community structure.

The contribution of invertebrates to specific rain garden functions varied significantly by site (Fig. 2)—a pattern that can be explained mostly by site-to-site variability in the abundance of earthworms, which are well documented engineers of ecosystem services and function in soils (Blouin and others 2013). Although $< 5\%$ of total filter media N was estimated to be stored in invertebrate tissues per rain garden, invertebrates may stimulate N storage in alternative reservoirs. For instance, soil invertebrate populations turn over quite rapidly (e.g. 1-3 $\times$ per year for earthworms [Parmelee and Crossley 1988]), and upon decomposition release bioavailable N that is readily assimilated by plants (Whalen and others 1999). Indeed, flux of N through earthworm biomass has been estimated to contribute up to 38% of plant N assimilation in some agricultural systems (Parmelee and Crossley 1988). Future studies should focus on quantifying invertebrate secondary production and population turnover in rain gardens, as these unknowns are required to accurately assess the contribution of invertebrate-facilitated storage to total N retention in these systems.

Denitrification within the anoxic guts of earthworms accounts for $< 2.2\%$ of average N_2O emissions from rain gardens designed with and without submerged zones (Grover and others 2013). However, because our estimates of invertebrate-facilitated denitrification do not account for enhanced denitrification in the walls of earthworm burrows (Parkin and Berry 1999) or excreted soils (Parkin and Berry 1994), they are conservative, and may substantially underestimate invertebrate contribution to total N removal. Indeed, our estimates of soil ingestion by earthworms and potworms in rain gardens suggest that the total soil processing capacity is large (up to 4 $\times$ the available near-surface soil mass in a given year [Fig. 3B]).

Because the potential for enhanced denitrification by earthworms scales with the mass of ingested soil, their impacts on N removal in rain gardens may dramatically exceed that implied by our within-worm estimates of N₂O and N₂ emission.

In summary, the findings presented here suggest that rain garden soil invertebrate communities are diverse, stable over short timeframes (~ 9 months), vary among sites, and likely impact many important rain garden functions. Rigorous assessment of soil invertebrate population dynamics in rain gardens is necessary to clarify these functional roles and their importance for contaminant treatment efficacy. Perhaps the most important take-home points from our study are that: (1) invertebrate assemblages are site-specific, suggesting that engineering design, age, and other site-associated characteristics strongly influence their abundance and diversity; and (2) invertebrate functional roles in rain gardens are also site-specific, with generally better outcomes expected for systems with higher abundance of ecosystem engineers (such as earthworms). These findings suggest that, with improved understanding of the factors that favor colonization of some taxa over others, rain garden design and maintenance could be “tuned” to favor invertebrates that confer important functional traits which, in turn, may improve the utility of these systems for protecting aquatic ecosystems and improving human and ecosystem water security.

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271 **Table 1.** Comparisons of soil invertebrate (≥ 0.3 mm) assemblages of Hereford Road (HR), Wicks Reserve (WR), and Lynbrook
 272 Boulevard (LB) rain gardens sampled in July 2013 (Winter) and March-April 2014 (Autumn). Probabilities resulting from pair-wise
 273 analysis of similarity (ANOSIM) tests for invertebrate community similarities between sites are given above the diagonal (*n.s.* = *p* >
 274 0.05). Values on the diagonal are percent similarity within site (SIMPER); values below the diagonal are percent dissimilarity between
 275 sites (SIMPER). Significance was set at $\alpha = 0.05$.

SITE / SEASON	WR Winter	WR Autumn	HR Winter	HR Autumn	LB Winter	LB Autumn
WR Winter	65.92%	n.s.	0.001	0.029	0.008	0.029
WR Autumn	43.96%	51.39%	0.005	0.010	0.002	0.005
HR Winter	59.66%	52.44%	59.52%	0.014	0.001	0.001
HR Autumn	64.32%	60.51%	53.17%	45.50%	0.016	0.029
LB Winter	68.09%	74.70%	75.24%	70.23%	38.64%	n.s.
LB Autumn	72.90%	74.59%	75.37%	70.59%	57.79%	46.04%

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278 **Figure legends.**

279 **Figure 1.** (A) Average Megadrilacea (earthworm), Enchytraeidae (potworm), Collembola
280 (springtail), and Acari (mite) abundance and (B) earthworm, potworm, and springtail biomass
281 per site. (C) Individual-based species accumulation curves and (D) multidimensional scaling
282 (MDS) plot of soil invertebrate (≥ 0.3 mm) assemblages in Hereford Road (HR), Wicks Reserve
283 (WR), and Lynbrook Boulevard (LB) rain gardens sampled in July 2013 (Winter) and March-
284 April 2014 (Autumn).

285 **Figure 2.** Potential soil invertebrate community contributions to nine selected rain garden
286 functions in Hereford Road (HR), Wicks Reserve (WR), and Lynbrook Boulevard (LB). Each
287 function is represented by 1/9 of the pie chart and is computed for each site using the average
288 abundance of each taxonomic group known to contribute to the given function (supplementary
289 material, Table S1) multiplied by a body size correction factor. Resulting products per function
290 and site were summed and either multiplied by the number of taxa known to positively affect the
291 function of interest (“richness-weighted”, left column) or not weighted by richness (right
292 column), and sorted into logarithmic bins (see supplementary material, Appendix S1 for
293 calculations). The resulting values combine species richness, biomass, and abundance into a
294 score for potential invertebrate community effect on rain garden function. Higher values within a
295 column indicate a greater potential enhancement of the given function of interest among sites.
296 Note that scales differ between columns.

297 **Figure 3.** (A) Box and whisker plots of estimated daily N_2O and N_2 emissions by earthworms.
298 Error bars represent 95% quantiles. (B) Box and whisker plots of estimated percentage (by dry
299 weight) of top 5 cm of rain garden soil media ingested by earthworms and potworms annually.

300 Error bars represent 95% quantiles. Calculations are provided in supplementary material,
301 Appendix S2.
302

303 **Figure 1.**

305 **Figure 2.**
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318 **Figure 3.**

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320 **References**

- 321 Ambrose, R.F.; Winfrey, B.K. Comparison of stormwater biofiltration systems in Southeast
322 Australia and Southern California. *WIREs Water*. 2:131–146; 2015
- 323 Anderson, J.M.; Ineson, P.; Huish, S.A. Nitrogen and cation mobilization by soil fauna feeding
324 on leaf litter and soil organic matter from deciduous woodlands. *Soil Biology &*
325 *Biochemistry*. 15:463-467; 1983
- 326 APHA. Standard methods for the examination of water and wastewater: American Public Health
327 Association Publication, APHA, AWWA, WEF; 2012
- 328 Askarizadeh, A.; Rippey, M.A.; Fletcher, T.D.; Feldman, D.; Peng, J.; Bowler, P.; Mehring, A.;
329 Winfrey, B.; AghaKouchak, A.; Jiang, S.C.; Sanders, B.F.; Levin, L.; Taylor, S.; Grant,
330 S.B. From rain tanks to catchments: Use of low-impact development to prevent and cure
331 the urban stream syndrome. *Environmental Science & Technology*. 49:11264–11280;
332 2015
- 333 Ayers, E.M. Pedogenesis in rain gardens: The role of earthworms and other organisms in long-
334 term soil development. *Biological Resources Engineering*. College Park, Maryland:
335 University of Maryland; 2009
- 336 Blouin, M.; Hodson, M.E.; Delgado, E.A.; Baker, G.; Brussaard, L.; Butt, K.R.; Dai, J.;
337 Dendooven, L.; Peres, G.; Tondoh, J.E.; Cluzeau, D.; Brun, J.-J. A review of earthworm
338 impact on soil function and ecosystem services. *European Journal of Soil Science*.
339 64:161–182; 2013
- 340 Chahartaghi, M.; Langel, R.; Scheu, S.; Ruess, L. Feeding guilds in Collembola based on
341 nitrogen stable isotope ratios. *Soil Biology & Biochemistry*. 37:1718–1725; 2005

342 Clarke, K.R.; Warwick, R.M. Change in marine communities: an approach to statistical analysis
 343 and interpretation. Plymouth, UK: Natural Environmental Research Council and
 344 Plymouth Marine Laboratory; 1994

345 Curry, J.P.; Schmidt, O. The feeding ecology of earthworms – A review. *Pedobiologia*. 50:463-
 346 477; 2007

347 Depkat-Jakob, P.S.; Brown, G.G.; Tsai, S.M.; Horn, M.A.; Drake, H.L. Emission of nitrous
 348 oxide and dinitrogen by diverse earthworm families from Brazil and resolution of
 349 associated denitrifying and nitrate-dissimilating taxa. *FEMS Microbiology Ecology*.
 350 83:375-391; 2012

351 Grover, S.P.P.; Cohan, A.; Chan, H.S.; Livesley, S.J.; Beringer, J.; Daly, E. Occasional large
 352 emissions of nitrous oxide and methane observed in stormwater biofiltration systems.
 353 *Science of The Total Environment*. 465:64–71; 2013

354 Horn, M.A.; Mertel, R.; Gehre, M.; Kästner, M.; Drake, H.L. In vivo emission of dinitrogen by
 355 earthworms via denitrifying bacteria in the gut. *Applied and Environmental*
 356 *Microbiology*. 72:1013-1018; 2006

357 Karsten, G.R.; Drake, H.L. Denitrifying bacteria in the earthworm gastrointestinal tract and in
 358 vivo emission of nitrous oxide (N₂O) by earthworms. *Applied and Environmental*
 359 *Microbiology*. 63:1878-1882; 1997

360 Kazemi, F.; Beecham, S.; Gibbs, J. Streetscale bioretention basins in Melbourne and their effect
 361 on local biodiversity. *Ecological Engineering*. 35:1454–1465; 2009a

362 Kazemi, F.; Beecham, S.; Gibbs, J.; Clay, R. Factors affecting terrestrial invertebrate diversity in
 363 bioretention basins in an Australian urban environment. *Landscape and Urban Planning*.
 364 92:304-313; 2009b

365 Lavelle, P. Faunal activities and soil processes: adaptive strategies that determine ecosystem
 366 function. in: Begon M., Fitter A.H., eds. *Advances in Ecological Research*. San Diego,
 367 CA: Academic Press; 1997

368 Leuven, R.S.E.W.; Brock, T.C.M.; Druten, H.A.M.v. Effects of preservation on dry- and ash-free
 369 dry weight biomass of some common aquatic macro-invertebrates. *Hydrobiologia*.
 370 127:151-159; 1985

371 Levin, L.A.; Mehring, A.S. Optimization of bioretention systems through application of
 372 ecological theory. *WIREs Water*. 2:259–270; 2015

373 Maraldo, K.; Ravn, H.W.; Slotsbo, S.; Holmstrup, M. Responses to acute and chronic desiccation
 374 stress in *Enchytraeus* (Oligochaeta: Enchytraeidae). *Journal of Comparative physiology*
 375 B. 179:113–123; 2009

376 Matthies, C.; Griebhammer, A.; Schmittroth, M.; Drake, H.L. Evidence for involvement of gut-
 377 associated denitrifying bacteria in emission of nitrous oxide (N₂O) by earthworms
 378 obtained from garden and forest soils. *Applied and Environmental Microbiology*.
 379 65:3599–3604; 1999

380 McBrayer, J.F.; Reichle, D.E. Trophic structure and feeding rates of forest soil invertebrate
 381 populations. *Oikos*. 22:381-388; 1971

382 Mehring, A.S.; Kuehn, K.A.; Thompson, A.; Pringle, C.M.; Rosemond, A.D.; First, M.R.;
 383 Lowrance, R.R.; Vellidis, G. Leaf litter nutrient uptake in an intermittent blackwater
 384 river: influence of tree species and associated biotic and abiotic drivers. *Functional*
 385 *Ecology*. 29:849-860; 2015

386 Mehring, A.S.; Levin, L.A. Potential roles of soil fauna in improving the efficiency of rain
 387 gardens used as natural stormwater treatment systems. *Journal of Applied Ecology*.
 388 52:1445–1454; 2015

389 Pandian, T.J.; Marian, M.P. Nitrogen content of food as an index of absorption efficiency in
 390 fishes. *Marine Biology*. 85:301-311; 1985

391 Parkin, T.B.; Berry, E.C. Nitrogen transformations associated with earthworm casts. *Soil*
 392 *Biology and Biochemistry*. 26:1233-1238; 1994

393 Parkin, T.B.; Berry, E.C. Microbial nitrogen transformations in earthworm burrows. *Soil Biology*
 394 *and Biochemistry*. 31:1765-1771; 1999

395 Parmelee, R.W.; Crossley, D.A. Earthworm production and role in the nitrogen cycle of a no-
 396 tillage agroecosystem on the Georgia piedmont. *Pedobiologia*. 32:355-361; 1988

397 Schmidt, O.; Scrimgeour, C.M.; Curry, J.P. Carbon and nitrogen stable isotope ratios in body
 398 tissue and mucus of feeding and fasting earthworms (*Lumbricus festivus*). *Oecologia*.
 399 118:9-15; 1999

400 Spicer, J.I.; Moore, P.G.; Taylor, A.C. The physiological ecology of land invasion by the
 401 Talitridae (Crustacea: Amphipoda). *Proceedings of the Royal Society B*. 232:95-124;
 402 1987

403 Teuben, A.; Roelofsma, T.A.P.J. Dynamic interactions between functional groups of soil
 404 arthropods and microorganisms during decomposition of coniferous litter in microcosm
 405 experiments. *Biology and Fertility of Soils*. 9:145-151; 1990

406 Teuben, A.; Verhoef, H.A. Direct contribution by soil arthropods to nutrient availability through
 407 body and faecal nutrient content. *Biology and Fertility of Soils*. 14:71-75; 1992

408 Whalen, J.K.; Parmelee, R.W.; McCartney, D.A.; Vanarsdale, J.L. Movement of N from
409 decomposing earthworm tissue to soil, microbial and plant N pools. *Soil Biology &*
410 *Biochemistry*. 31:487-492; 1999

411 Xu, D.; Li, Y.; Howard, A. Influence of earthworm *Eisenia fetida* on removal efficiency of N and
412 P in vertical flow constructed wetland. *Environmental Science and Pollution Research*
413 *International*. 20:5922-5929; 2013

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