The importance of coprophagous macrodetritivores for the maintenance of vegetation heterogeneity in an African savannah

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in review Ecosystems
ABSTRACT

Grazing ecosystems are often characterized by dynamic vegetation structure mosaics of short grazing lawns and tall grass vegetation that are important for the biodiversity and functioning of these ecosystems. Grazing-induced trampling, causing soil compaction and reduced water infiltration, has been shown to be an important mechanism for lawn grass formation. However, insights in reverse bioturbation mechanisms were mostly lacking, especially how tall vegetation persists under continuous grazing by herbivores. In this study we explore if defecation by large herbivores in combination with different groups of coprophagous macrodetritivores can locally convert compacted grazing lawn patches back to tall bunch grasslands with a more loose soil. Across a rainfall gradient in an African savannah, we separated the potential roles in this process between dung beetles versus earthworms and termites. We placed different mesh sizes under dung piles and studied the consequences for soil, vegetation and hydrological properties. We found that soil water infiltration rate, soil organic matter content, electrical conductivity, bunch grass cover and bunch grass biomass were significantly promoted by dung addition, irrespective of position along the rainfall gradient. In addition the presence of tunneling dung beetles significantly increased water infiltration rate and biomass of bunch grasses, pointing at a new mechanism whereby macrodetritivores affect the structure and diversity of plant communities. We conclude that coprophagous macrodetritivores interact with large herbivores in contributing to the maintenance of structural heterogeneity in the vegetation of grazing ecosystems, with a special role played by soil-tunneling dung beetles.
INTRODUCTION

Grazing ecosystems are often characterized by dynamic vegetation structure mosaics (Frank et al., 1998, Schrama et al., 2013) of short horizontally growing, stoloniferous lawn grasses (Hempson et al., 2014) and tall tussock forming bunch grasses (Anderson et al., 2013), that are important in the biodiversity and functioning of these ecosystems (Pickett et al., 2003, Knapp et al., 1999, Cromsigt and Olff, 2008). In migratory systems the maintenance of heterogeneity in vegetation structure is attributed to seasonally intensive grazing alternating with rest periods, where herbivores concentrate grazing on preferred resources and move on through the landscape, leaving less preferred patches intact (Knapp et al., 1999, McNaughton, 1979). Moreover, resident grazing herds may promote structural heterogeneity when total grazing pressure is maintained at intermediate levels, thus patchiness is maintained by the aggregation of herbivores on higher forage quality patches (Oom et al., 2010, Stuth et al., 1997, Cromsigt et al., 2009). Expansive patchiness of tall grasses alternating with grazing lawns provides an essential buffer against high herbivore density oscillations, since tall vegetation may be used as a lower quality forage reserve (Owen-Smith, 2004, Ruifrok et al., 2015) as well as increasing habitat availability and potential for biodiversity (Hagenah et al., 2009, van der Plas Howison et al., 2013) which may be especially important in more confined natural areas (Chapin et al., 2000).

The main mechanisms that have been put forward for grazing lawn formation are; co-evolution of plants that tolerate or resist grazing (Milchunas et al., 1988, McNaughton, 1984) in the presence of large herbivores which optimize foraging strategies (McNaughton, 1979, Westoby, 1974) and positive nutrient feedbacks (Ruess and McNaughton, 1987, Cromsigt and Olff, 2008). In addition, the growing emphasis on the effects of global change has inspired additional explanations for vegetation patchiness. In particular scale-dependent differences in water availability at the landscape scale, where water runs off from dry bare soil and seeps into the soil under perennially vegetated patches (Rietkerk and Van de Koppel, 1997), resulting in an uneven distribution of water, thus promoting a patchy mosaic of dry bare patches alternating with moist vegetated patches. Although mosaics of tall and lawn grasses are frequently described as a typical feature of grazed ecosystems with an important role of large herbivores (Frank et al., 1998, Stock et al., 2010), the maintenance of tall vegetation has traditionally been ascribed to a range of determinants of temporal patch avoidance, either through, predation risk (Hopcraft et al., 2010), avoidance of dung due to pathogen risk (Hart, 1990, Cid and Brizuela, 1998) or seasonal migration (Fryxell, 1991, Fryxell and Sinclair, 1988). However, recent work shows important differences in soil physical properties with significantly higher water infiltration rates and increased soil macroporosity in tall vegetated bunch patches compared to adjacent short vegetated lawn patches, within the same spatial context (Veldhuis et al., 2014, Howison et al., 2015, van Klink et al., 2015). Coupled to this we find higher abundance of soil macrodetritivores within the tall vegetation (Howison et al., 2015). This suggests that mechanisms other than temporal patch avoidance, such as the bioturbating activities of soil macrodetritivores, may be important in creating differences in soil properties within tall vegetation (Howison et al., 2015, Schrama et al., 2015). In time (between 5 – 10 years) we
observe spatial dynamics of tall and short patches converging into each other, i.e. tall vegetation invades lawn or lawn invades tall vegetation. Therefore we expect positive feedbacks generated by large grazing herbivores which promote lawn to alternate with positive feedbacks generated by soil bioturbation.

Soil macrodetritivores play a crucial role in soil formation processes (Wilkinson et al., 2009). As far back as Darwin (1881) and more recently (Meysman et al., 2006), scientists recognize the importance of bioturbation by soil organisms in long-term modifications in local soil conditions through nesting and foraging behavior, thereby often promoting their own resources and conditions (van Breemen, 1993). Estimates for the magnitude of impact soil organisms have, through mounding, mixing and burial range from 3 to 53 ton.ha$^{-1}$.yr$^{-1}$ of processed soil for the temperate regions and between 730 to 1100 ton.ha$^{-1}$.yr$^{-1}$ in the humid tropics (Wilkinson et al., 2009). Predominantly in agricultural literature we find studies that quantify the benefits to natural soil processes by bioturbating soil macrodetritivores (Stuth et al., 1997, Lal, 1988) and consequences for above ground biomass production (Curry and Boyle, 1987, van Breemen, 1993). However, to our knowledge, no clear linkages have been made between the bioturbating activities of soil biota and patch conversion from lawn to tall grass vegetation patches through soil amelioration, within grazing ecosystems literature.

In this study we test the hypothesis that macrodetrivore aggregation in dung patches can cause compacted, dry lawn grass-dominated grazing lawns to revert to bioturbated,
wetter, tall bunch grass-dominated patches (Fig. 5.1). If this is true, this may explain why small-scale vegetation structural heterogeneity in grazing ecosystems can persist even under continuous grazing pressure throughout the growing season. For this, we experimentally tried to switch herbivore-compacted grazing lawns to macrodetrivore, bioturbation-dominated tall bunch grass patches by dung addition (Fig. 5.1), with experimental separation of the importance of different groups of coprophagous macrodetrivores (i.e. earthworms, termites and dung beetles). The study was situated along a steep rainfall gradient to test for effects contingent on water availability. For understanding the mechanisms involved, we measured the importance of the incorporation of dung into the soil profile thus enhancing soil organic matter content and nutrient availability (Mittal, 1993), and the promotion of water infiltration through tunneling, foraging and nesting activities (Lal, 1988), separating between dung beetle and earthworm/termite effects. We measured the consequences for above ground biomass for tall bunch grasses, lawn grasses, forbs, leaf litter production and bare ground cover to quantify the strength of patch conversion.

**METHODS**

**Research area**
This research was performed at Hluhluwe-iMfolozi Park (HiP) between the coordinates 28°00” to 28°26”S and 31°41” to 32°09”E in northern KwaZulu Natal, South Africa. HiP is a highly heterogeneous but relatively small (900 km²) nature conservation area within the savannah biome of Southern Africa, situated in the transition zone between the foothills of the kwaHlabisa mountains to the coastal lowlands (Stock et al., 2010, Mucina and Rutherford, 2006). Elevation decreases from 500 to 40 m.a.s. in a northwest-southeast plane, which is strongly associated with a decreasing rainfall gradient from 920 to 490 mm yr⁻¹, falling mostly between October and April. The areas host a high plant and animal diversity (including several endemics, (Smith et al., 2008) and high wildlife abundance (Stock et al., 2010, Kleynhans et al., 2011, Cromsigt et al., 2009). Air temperatures range from a mean minimum temperature of 13°C to a mean maximum temperature of 35°C (Balfour and Howison, 2001). The vegetation comprises a patchy mosaic of tall bunch grasses (*Themeda triandra, Sporobolus pyramidalis, Eragrostis curvula*) and short lawn grasses (*Sporobolus nitens, Paspalum notatum, Digitaria longiflora, Cynodon dactylon*). The most commonly occurring large herbivore species are white rhinoceros (*Ceratotherium simum*), Cape buffalo (*Syncerus caffer*), impala (*Aepyceros melampus*), nyala (*Tragelaphus angasi*), Burchell’s zebra (*Equus quagga*), waterbuck, warthog (*Phacochoerus africanus*) and wildebeest (*Connochaetes taurinus*). Coprophagous macrodetrivores include different species of earthworms (Plisko, 2012), termites (Freyman et al., 2008) and dung beetles (Coleoptera: Scarabaeidae), which can be divided into: paracoprids, which tunnel in soil beneath the dung pile; endocoprids, which reside and brood their young directly within the dung pile; and telecoprids, which famously roll their dung balls away from the dung pile and bury them at some distance (Nichols et al., 2008, Chao et al., 2013, Stronkhorst and Stronkhorst, 1997).
Treatments

In a full factorial block design we compared the changes that occurred in two dung addition treatments to a control site within every replicate, which would account for any changes that occurred due to external environmental factors (e.g., seasonal progression). For the two dung addition treatments we placed 20 kg dung piles (1 x 1 x 0.25 m, a typical defecation) on top of either a double sheet of fine metal mesh (0.1 x 0.1 cm – to prevent penetration of tunneling dung beetles and similar large macrodetritivores from the dung into the soil, or coarse mesh (2 x 2 cm – allowing the activity of all types of macrodetritivores (Fig. 5.2 A-F)). This design allowed the separation between the effects of activities of macrodetritivores that arrive in the dung pile from below ground (termites and earthworms) and macrodetritivores arriving from above ground (dung beetles). The dung was collected as fresh as possible in the early morning, i.e., had to have been deposited by a white rhinoceros within the previous night and was mixed thoroughly in a large 80 L bin before placing on the mesh. All plots including that of the control sites were located in mixed bunch-lawn vegetation where the ratio ranged between 40:60 to 60:40 and no obvious signs of soil macrodetritivore activities were visible. This excluded (delays in) plant species colonization from the experimental design as a potential explanation for understanding vegetation responses. Vegetation plots were located directly adjacent to the dung addition plots, oriented along the main downslope gradient. Vegetation responses were therefore not explained by the direct killing of vegetation by the shading of the dung, while any increased infiltration of water at the dung patch was expected to also extent to affect the undisturbed vegetation directly next to it, downhill. The experiment was done in a randomized block design at 5 sites of 3 replicates per treatment (control, coarse mesh, fine mesh), arranged in 3 blocks containing all 3 treatments. Spacing between blocks within a site was from 50 and 100 m. The 5 sites (maximally 30 km and minimally 2 km apart) were positioned across a rainfall gradient from 500 – 700 mm.yr\(^{-1}\) to allow for generality of the conclusions (i.e., not contingent to a specific rainfall or set of local soil conditions). The experiment lasted for 12 weeks, in the period between December 2013 and March 2014. Figure 5.2 provides an overview of the layout of the different experimental treatments.

Macrodetritivore distribution

Two plots of 40 x 40 cm per dung pile were used to score earthworm casts and termite sheeting, which are characteristic signs of their presence (Henrot and Brussaard, 1997, Kihara et al., 2014, Ndiaye et al., 2004) and used as relative estimates for their activities between sites. The dung piles were carefully lifted and one plot was located central (maximum moisture) and one randomly at the edge (transitioning from wet to more dry). Plots were divided into 10 cm\(^2\) subplots (4 x 4 subplots) and in each subplot the cover of termite, earthworm or dung beetle holes was scored as: 0 (not present), + (<25), 25–50, 51–75, 76–100% coverage. To quantify the abundance and distribution of dung beetles we also conducted a dung colonization study following the approach of Stronkhorst and Stronkhorst (1997). In order to not disturb the patch conversion experiment by macrodetritivore sampling we collected additional dung and placed 3 x 10 kg dung piles on a sheet of coarse mesh within 20 m of each replicate. Dung was placed at 7 am in the morning and
equal portions were removed and placed in a 10L plastic basin and searched for beetles (dung-dwellers) after 3, 5, 8 and 26 hours of dung incubation. All dung beetles encountered were counted, identified and the pronotum width measured to estimate body size. At 8 and 26 hours of dung incubation we applied a water flotation method (Edwards, 1991) below the dung piles to sample soil-tunneling dung beetles. Body mass from protonum width calibration curves per dung beetle species (≈10 individuals per species, dried for 200 hours at 70°C) were used to estimate the biomass of each individual, using linear regression.

Ground cover and vegetation biomass measurements
Permanent 40 x 80 cm vegetation monitoring plots were set up as controls and on the downslope side of the dung pile. Ground cover by vegetation was assessed by dividing the plot into 36 smaller 10 cm² subplots (4 x 8 subplots). Each subplot was subdivided into quarters and in each subplot the lawn vegetation (stolons and leaf cover), bunch grass vegetation (basal cover), forbs, litter, and bare soil were given a score of: 0 (not present), + (<25), 25–50, 51–75, 76–100% soil cover. Vegetation height for lawn was estimated by dropping a small disc pasture meter (weighing 68.5 g, Ø 11.5 cm) sliding along a plastic pole (Ø 2cm) from 50 cm height above the ground, repeated 3 times within the vegetation plot. For the bunch-grass vegetation we recorded the number of tussocks, and within each tussock counted the number of individual ramets and measured the length of the tallest leaf from 10 randomly selected ramets. These non-destructive measurements were used to
calculate real vegetation biomass using a series of calibration plots. The vegetation cover and height was recorded in these plots the same way as in the vegetation plots. The vegetation of each plot was then clipped separately for the lawn and bunch grasses, dried at 70°C until constant weight (24 hours), and weighed. Linear regressions were then fitted to calculate the dry biomass from estimated abundance for each group. Vegetation measures were conducted at the start and end of the experiment.

**Soil measurements**

Soil water infiltration rate (mm.sec⁻¹) was measured using a double-ring infiltrometer (Bower, 1986). Soil organic matter content (%) was estimated using the loss on ignition method by ashing the samples for 16 hours at 420°C (Stock et al., 2010). Soil electrical conductivity (µS.cm⁻³) was measured as a proxy for soil salinity (Mills et al., 2009). Soil air porosity (%) was measured by maximally compressing 10 x 5 cm Ø soil cores using a technique following Jafarzadeh (2006). Lastly, we estimated soil moisture content (%) and bulk density (g.cm⁻³) using 100 cm⁻³ volumetric samples dried at 105°C for 48 hours (Terzaghi, 1996). Soil measurements were conducted at the start and end of the experiment to control for environmental effects.

**Dung analysis**

Dung electrical conductivity (µS.cm⁻³) was measured as a proxy for dissolved salts content (Mills et al., 2009) by washing a mixed sub-sample (50 g) of fresh dung at the beginning and end of the experiment with 150 ml of demineralized water. A second set of dung samples (50 g) were weighted and dried at 70°C for 48 hours (until constant weight). Differences in dung moisture content were calculated between the start and end of the experiment.

**Data analysis**

We used redundancy analysis (RDA) as available in the vegan R package (Oksanen et al, 2013) to reduce the dimensionality of the dataset, and explore the multivariate relationships between predictor and response variables. The RDA calculations were based on correlation instead of covariance matrices in order to standardize variables of varying scales and magnitudes. PERMANOVA (permutational multivariate ANOVA available from the vegan R package (Oksanen, 2011), with 999 permutations) was used to test for treatment and rainfall effects for the combined response of vegetation cover (bunch, lawn, forb) and soil properties (water infiltration, organic matter, electrical conductivity, air porosity, moisture, bulk density) to the experimental treatments. Dung colonization by different groups of coprophagous macrodetritivores along the rainfall gradient were analyzed using Linear Mixed Models (LMM) (Pinheiro et al., 2013) with block as a random effect. Using general LMMs we compared vegetation and ground cover measures (litter, bare ground), the change in bunch and lawn vegetation biomass (change = end BM – start BM), and soil properties in relation to the two dung addition treatments (placed on fine and coarse mesh), using rainfall and their interactions with block as a random effect. Since no interaction effects were found across the rainfall gradient for all response measures (Table 5.1)
we further tested for differences between treatments by contrasting pairs of treatments (Bates et al., 2014) with block nested within site as random effects. Response ratios were calculated following Hedges et al. (1999). All statistical analyses were conducted using the statistical software R, version 3.1.1 (RCoreTeam, 2015).

RESULTS

The RDA analysis revealed two main axes of variation within the data. Axis 1 mostly represents variation within the data along the rainfall gradient (55%, indicated as contours, Fig. 5.3) and axis 2 represents the variation mostly due to the dung addition treatments in comparison to the control (40%, indicated as ellipses, Fig. 5.3). The PERMANOVA, showed that the relationships between plant cover (bunch, lawn and forb) and soil properties (water infiltration rate, organic matter content, electrical conductivity, air porosity, soil moisture content, bulk density) for both rainfall (PERMANOVA: F(1,44) = 6.41, \(P < 0.01\)) and dung addition treatments (PERMANOVA: F(2,44) = 14.86, \(P < 0.001\)) were highly significant.

Macrodetritivore distribution

The analysis of the variation in the colonization of the dung by coprophagous macrodetritivores revealed that earthworms increased in abundance with increasing rainfall (LMM:

Figure 5.3 Biplot showing the scores an RDA of response measures (ground cover and soil properties), in relation to constrained treatments (shown as centroid ellipses: control, fine and coarse mesh treatments) and unconstrained predictors (shown as contours: rainfall). Abbreviations: Lawn, Bunch, Litter, Bare, Forb = ground cover scores; Infil = water infiltration; EC = electrical conductivity (as proxy for salinity); OM = soil organic matter content; Porosity = soil air porosity; Moist = soil moisture content; BulkD = soil bulk density.
The dung addition treatments significantly favored the expansion of bunch grass at the cost of non-bunch cover (lawn + bare ground + forb cover), as indicated by the uniformly higher ratio of bunch to non-bunch cover (Table 5.1) across the rainfall gradient. The change in bunch grass shoot biomass between the start and end of the experiment was significantly higher for the dung addition treatments. In addition, bunch grass shoot biomass increased towards higher rainfall and a significant difference was detected between the different mesh treatments, where bunch grass next to the coarse mesh treatment was higher than next to the fine mesh (see differences in means and response ratios, Table 5.1). The change in lawn grass biomass was not different between the control and treatments and was uniform along the rainfall gradient (Table 5.1).

**Figure 5.4** Changes in (proxies for) the abundance of different coprophagous macrodetritivores along the rainfall gradient in the dung colonization experiment. A) Colonization of termites and earthworms, measured as % surface cover of their sheeting or casting beneath a dung pile. B) The cumulative mean biomass (dry weight) of soil-tunnelling versus dung-dwelling dung beetles (Coleoptera: Scarabaeidae). Points represent mean ±SE.

F(2,57) = 14.44, P < 0.001) while termites instead decreased in abundance (LMM: F(2,57) = 5.81, P < 0.05) (Fig. 5.4A). The colonization by tunneling dung beetles instead was not related to the rainfall gradient (LMM: F(1,13) = 1.71, P = 0.21) while dung-dwelling dung beetles increased significantly in abundance with increasing rainfall (LMM: F(1,13) = 7.64, P < 0.05) (Fig. 5.4B). The different mesh size treatments were successful in separating effects of dung beetles from that of earthworms and termites since only very few holes (average <1, max = 3) indicative of soil-tunneling dung beetles were found beneath the double fine mesh layer compared to the coarse mesh (average = 22, max = 128), (Paired t-test: T(1,13) = 2.41, P < 0.05).

**Ground cover and vegetation biomass measurements**

Bunch grass cover was significantly higher in both of the dung addition treatments, which were not different from each other, whereas lawn grass was not affected by the dung addition treatments (Table 5.1). The dung addition treatments significantly favored the expansion of bunch grass at the cost of non-bunch cover (lawn + bare ground + forb cover), as indicated by the uniformly higher ratio of bunch to non-bunch cover (Table 5.1) across the rainfall gradient. The change in bunch grass shoot biomass between the start and end of the experiment was significantly higher for the dung addition treatments. In addition, bunch grass shoot biomass increased towards higher rainfall and a significant difference was detected between the different mesh treatments, where bunch grass next to the coarse mesh treatment was higher than next to the fine mesh (see differences in means and response ratios, Table 5.1). The change in lawn grass biomass was not different between the control and treatments and was uniform along the rainfall gradient (Table 5.1).
Table 5.1 The effects of the dung addition treatments placed along a rainfall gradient on vegetation and soil properties, allowing the separation of the impacts of dung beetles versus earthworms and termites. Means with different letters are significantly different (using contrasted pairs of treatments [Bates et al., 2014]). Effect sizes were calculated following Hedges et al. (1999). Results with significant effects are shown in bold and variables significantly affected by the dung addition treatment are highlighted. Asterisks denote significant F values (*P < 0.05; **P < 0.01; ***P < 0.001).

<table>
<thead>
<tr>
<th>Measure</th>
<th>Treatment F-Value</th>
<th>Rainfall F-Value</th>
<th>Interaction (Treatment* Rainfall) F-Value</th>
<th>Means</th>
<th>Effect Size</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Control</td>
<td>Fine mesh</td>
</tr>
<tr>
<td>Bunch Cover</td>
<td>8.65 ***</td>
<td>7.04 *</td>
<td>0.94 n.s.</td>
<td>17.24 a</td>
<td>28.83 b</td>
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<td>Lawn Cover</td>
<td>1.64 n.s.</td>
<td>5.07 *</td>
<td>0.04 n.s.</td>
<td>33.18 a</td>
<td>29.64 a</td>
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<tr>
<td>Non Bunch Cover (Lawn + Bare + Forb)</td>
<td>3.59 *</td>
<td>6.98 **</td>
<td>1.74 n.s.</td>
<td>65.65 a</td>
<td>54.40 b</td>
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<tr>
<td>Bunch/Non-bunch Cover Ratio</td>
<td>3.03 *</td>
<td>1.75 n.s.</td>
<td>0.87 n.s.</td>
<td>0.29 a</td>
<td>0.83 b</td>
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<tr>
<td>Litter Cover</td>
<td>0.38 n.s.</td>
<td>1.24 n.s.</td>
<td>1.19 n.s.</td>
<td>11.88 a</td>
<td>11.51 a</td>
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Vegetation and ground cover

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<th>Treatment F-Value</th>
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<th>Interaction (Treatment* Rainfall) F-Value</th>
<th>Means</th>
<th>Effect Size</th>
</tr>
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<td></td>
<td></td>
<td></td>
<td></td>
<td>Control</td>
<td>Fine mesh</td>
</tr>
<tr>
<td>Change in biomass Bunch</td>
<td>9.01 ***</td>
<td>8.74 **</td>
<td>0.10 n.s.</td>
<td>10.72 a</td>
<td>23.94 b</td>
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<td>Change in biomass Lawn</td>
<td>0.69 n.s.</td>
<td>1.38 n.s.</td>
<td>1.84 n.s.</td>
<td>12.07 a</td>
<td>19.85 a</td>
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Shoot biomass

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<th>Rainfall F-Value</th>
<th>Interaction (Treatment* Rainfall) F-Value</th>
<th>Means</th>
<th>Effect Size</th>
</tr>
</thead>
<tbody>
<tr>
<td>Water infiltration</td>
<td>10.08 ***</td>
<td>1.75 n.s.</td>
<td>1.41 n.s.</td>
<td>1.00 a</td>
<td>3.31 b</td>
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<tr>
<td>Organic matter</td>
<td>8.33 **</td>
<td>47.36 ***</td>
<td>0.55 n.s.</td>
<td>4.65 a</td>
<td>5.20 b</td>
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<tr>
<td>Salinity</td>
<td>33.91 ***</td>
<td>13.26 ***</td>
<td>1.06 n.s.</td>
<td>54.25 a</td>
<td>121.87 b</td>
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Soil properties

<table>
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<th>Interaction (Treatment* Rainfall) F-Value</th>
<th>Means</th>
<th>Effect Size</th>
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<tbody>
<tr>
<td>Air porosity</td>
<td>1.56 n.s.</td>
<td>7.82 **</td>
<td>0.04 n.s.</td>
<td>11.47 a</td>
<td>14.33 a</td>
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<td>Soil moisture</td>
<td>0.89 n.s.</td>
<td>31.92 ***</td>
<td>0.36 n.s.</td>
<td>11.85 a</td>
<td>12.77 a</td>
</tr>
<tr>
<td>Bulk density</td>
<td>0.91 n.s.</td>
<td>1.37 n.s.</td>
<td>0.25 n.s.</td>
<td>1.34 a</td>
<td>1.30 a</td>
</tr>
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</table>
Soil measurements
Water infiltration rate, soil organic matter content and electrical conductivity were significantly positively affected by the dung addition treatments; a significant difference was detected between treatments where water infiltration in the coarse mesh treatment was highest. However, there were no significant differences between treatments for soil organic matter content and electrical conductivity (Table 5.1). Soil air porosity, soil moisture content and bulk density were unaffected by the dung addition treatments. Soil organic matter content, air porosity and soil moisture content were all significantly positively correlated and soil electrical conductivity negatively correlated to increasing rainfall, whereas water infiltration rate and bulk density were unrelated to the rainfall gradient (Table 5.1).

Dung analysis
Electrical conductivity of the remaining dung at the end was significantly lower compared to the start of the experiment (Paired t-test: $T_{(1,4)} = 11.67, P < 0.001$). Moisture content of the remaining dung was wetter at the end compared to the start of the experiment (Paired t-test: $T_{(1,4)} = 7.33, P < 0.01$).

DISCUSSION
A remarkable first finding is that the effect of adding dung on vegetation and soil variables were equal in magnitude at all sites along the rainfall gradient in our study area. Specifically, the responses of the tall bunch grasses to the experimental manipulations were uniform across the rainfall gradient. In contrast, earthworms and termites replaced each other along the rainfall gradient, where earthworms were more prevalent towards higher mean annual rainfall and termites were more prevalent towards the drier regions, suggesting a functional complementarity at the landscape scale that is in agreement with other such studies (Lal, 1988, Henrot and Brussaard, 1997). Dung beetles are attracted by aromatic cues and, being mobile, opportunistically occupy dung piles within hours of its deposition (Stronkhorst and Stronkhorst, 1997). Soil-tunneling dung beetles were equally spread along the rainfall gradient, however dung-dwelling dung beetles increased in abundance toward higher rainfall.

We demonstrated that basal cover of tall bunch grasses in this savannah ecosystem, can significantly increase within 12 weeks by attracting coprophagous soil biota which colonize the dung pile, either entering the dung pile from below (earthworms and termites) or from above (dung beetles). We showed that particularly the cover ratio of bunch:non-bunch and biomass of bunch grasses profit from important alterations to soil properties. Macrodetritivores had two main effects on soil properties as shown by our results. First, they increased soil organic matter content, which positively enhances nutrient availability (Ruess and McNaughton, 1987) through incorporation of dung into the soil. Secondly, they increased water availability through the creation of more permeable soil, since we found significantly higher infiltration rates under the dung piles, which both due to accessing the
dung from below the pile and the burying of dung balls by soil-tunneling dung beetles. We did not find differences in soil moisture, however our measure was a once off instantaneous measure and does not capture the strong temporal variability (e.g. depending on rainfall of the previous day). In our interpretation of the results we put higher emphasis on the differences in infiltration rate, as actual soil moisture differences are subject to much stronger temporal variability (e.g. depending on rainfall the previous day). Increased soil permeability allows surface water, running off from adjacent compacted vegetation patches, to soak into these patches therefore improving growing conditions. Lawn cover and lawn dry mass were unaffected by the dung addition treatments, although we did not control for biomass removal by grazing herbivores, (caging the plots would have led to unwanted interactions). However, grazing by herbivores would lead to a reduction in both biomass and cover, as herbivores generally prefer the higher quality lawn grasses. A more likely explanation is therefore that the better growing conditions (more water and nutrients) allowed the taller, thicker rooted bunch grasses (van der PlasZeinstra et al., 2013) to expand in basal cover and above ground biomass. Therefore, although bunch grasses were not able to displace lawn grasses in 12 weeks, bunch grasses were able to invade the available bare ground (faster than the lawn species) and displace herbaceous forb species.

A clear result was that soil electrical conductivity was significantly higher in the vicinity of the dung pile, likely caused from salts in the dung leaching into the ground. Higher electrical conductivity (as a proxy for salinity) is physiologically stressful to plants (Parida and Das, 2005) as it can impair the water balance of the plant in ecosystems with a clear dry season. However, it has been shown that nutrient addition is generally far more important to plant production than salinity (Poorter and Nagel, 2000). Soil bulk density and air filled porosity tended to increase in the presence of dung and soil macrodetritivores, however, the effect of treatment was not significant. The contrast between our findings and expectations from other studies could be because most studies test the decreasing effect of soil macrodetritivores on bulk density in the excavated soil (Mittal, 1993, Joschko et al., 1989). In this case, the soil remaining within the undisturbed soil profile remains at the same average density (in our case, per 10 cm²) albeit with increased water infiltration rate and organic matter content due to increased presence of millimeter-scale biovoids. Quantifying these effects would require more advanced quantification of 3D soil physical structure than we did in our study.

Previous studies have shown that increased nutrient addition as ureum or NH₄NO₃ in the presence of herbivores promotes lawn formation in savannahs (Ruess and McNaughton, 1987), specifically above a certain threshold patch size (Cromsigt and Olff, 2008). Such studies mostly mimic nutrient returns by herbivores as urine (Veldhuis et al., 2014) or fecal deposits that comprise small pellets and are easily scattered. Our study suggests that large dung piles may have an opposite effect in grazing ecosystems. In this case, the residual plant material remains on top of the soil surface for an extended period of time. Very large herbivores such as buffalo, white rhino and elephant (Stronkhorst and Stronkhorst, 1997, Hobbs, 1996) defecate in exponentially larger quantities compared to herbivores < 500kg weight (Hobbs, 1996). The social and territorial behavior in males of smaller grazers as zebra, wildebeest and impala to defecate on the same pile, leading to
middens can lead to similar effects. Territorial middens created by megaherbivores (>1000kg) such as male white rhinoceros are surrounded by grazing lawns created by frequent use, intense grazing pressure and increased nutrients (Owen-Smith, 1988), on the other hand female rhinoceros do not concentrate their dung in middens and defecate throughout the landscape (Estes, 1991). Our observed natural effects of dung on termites and associated soil properties likely point at similar mechanism known from African agro-ecosystems. Rural farmers in Niger have been shown to utilize termite activities to restore degraded and compacted soil by placing vegetative residue on the soil surface. This promotes feeding tunnel formation of termites and thus ameliorates soil conditions by improving water infiltration, incorporation of organic materials into the soil profile and vegetation establishment (Lal, 1988). Similarly, organic mulch (woody debris + straw) has been shown to work well in the Sahel in improving water infiltration in degraded (crusted) soils due to its stimulating effect on termites (Mando and Miedema, 1997).

We conclude that our study supports the hypothesis that macrodetritivore aggregation in and under dung piles can lead to patch conversion from short lawn grass dominated to tall bunch grasses in small-scale grazing mosaics (Fig. 5.1). Removal of the keystone species from African savannahs, as the white rhinoceros (as now ongoing in our study area due to heavy poaching) that strongly interact with macrodetritivores could hence induce large ecological changes, with cascading impacts on local vegetation heterogeneity, biodiversity, and associated ecosystem functions and services (Coleman and Williams, 2002). Across a wider range of ecosystems, similar types of interactions between macrodetritivores and soil are now even considered to be at least as important as the classical trophic interactions studies (Reise, 2002, Meysman et al., 2006).

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