Persistence as a target in restoration: What can we learn from the storage effect?

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<u>Executive Summary</u>

- This project was funded by City of Boulder Open Space & Mountain Parks (OSMP) in March 2018 ($18,554, including fence installation). This report describes the status and plan for completion.
- **Project Objectives:** The study proposed to establish a combined manipulation of rainfall (ambient, dry [rainfall shelter], wet [water addition]) and grazing (fall, spring, none) in OSMP's grasslands to evaluate plant responses to these factors and address two key questions:
    - **Q1)** Do species "see" different environmental conditions as favorable (presenting opportunities for regeneration) or unfavorable (requiring storage to persist)? Do traits explain the variation among species?
    - **Q2)** Can practical management strategies structured around regeneration events and storage capacities enhance native diversity at the community level?
- **Summary of Progress:** In 2018, we established the proposed grazing and rainfall manipulations and began collecting baseline data on plant responses. Baseline data include mapped plant communities (to follow recruitment, growth, and survival of individuals over time) and plant production data at the community-level (standing plant biomass and plant height).
- **Summary of Preliminary Results:**
    - The rainfall manipulation from 22 May 2018 to 15 Sep 2018 intercepted 145mm of rainfall, which was redistributed to water addition plots across 7 water addition events.
    - Rainfall manipulation had measurable effects on soil volumetric water content across the plot surface (i.e. from the center of plots to the edge). Observed effects on plant production during peak growing season (August) were smaller and non-significant in the first year of the manipulation.
    - The first fall graze treatment is in progress. Grazed blocks were opened to cattle for 3 weeks in October, then re-opened at the end of November 2018 (cattle occupation anticipated through December 2018). We have collected mid-graze data on plant production prior to re-opening the grazed blocks in November.
        - In November, we found small but non-significant reductions in plant biomass (19%) and height (22%) in grazed blocks relative to the non-grazed blocks following the 3 week graze period. For both metrics, plant height and biomass were reduced more in **external** graze areas (~40m from site fence, exposed to approx. 6 weeks of grazing at time of sampling).
        - These preliminary results informed the decision to re-open the grazed blocks to cattle in late November. They will remain open until cattle leave the site, and data will be re-collected in January 2019.
- **Scope of Report:** We cannot yet address the key research questions above. However, this report details project methodology to date and presents preliminary results related to the effects of rainfall and grazing. Contents include:
    - Proposal Abstract
    - Methods
    - Preliminary Results
    - Plan for Completion

Proposal Abstract

Understanding plant community dynamics over time is a critical but challenging goal for ecological restoration, particularly with growing consensus that communities must be managed for resilience to anticipated environmental changes. We need to better understand how tools that managers can deploy – such as the timing of livestock grazing – can be used to meet management goals in the face of increasing threats from aspects less under their control (e.g., exotic plant invasions and extreme climate events). Storage effect theory posits that population persistence depends on an ability to regenerate (produce seed, recruit) during favorable conditions while storing in a buffered stage (adults or seedbank) during unfavorable conditions. If species' regeneration and storage responses to fluctuating conditions differ predictably, community diversity could be maintained by managing for species that thrive in different conditions. In this project, I propose to measure demographic responses of species in a mixed prairie over time (3+ years) and in response to direct manipulations of grazing timing and precipitation. I will (1) use species' traits (e.g., seed, leaf, root, and growth attributes) to predict how changing conditions affect regeneration, storage, and, ultimately, persistence; and (2) ask whether targeted seed addition and grazing treatments are viable tools to reduce regeneration opportunities for exotic species and increase chances of native regeneration—thus enhancing community diversity—under a range of conditions.

Methods

Study Site

The study was established on a 100 ha fenced parcel of public land owned by the City of Boulder Open Space and Mountain Parks (OSMP, Boulder, CO, USA; Fig. 1). The Tracy Collins property has no pubic trails or access, but is regularly leased to tenant ranchers for cattle grazing. The grazing regime at this site has varied over the last decades (records extending back to 1990s), and has included spring (May-Jun; sometimes extending through September) and/or fall (Oct-Dec) grazing by non-calving heifers (as part of cow-calf operations). Vegetation at the site is classified as xeric tallgrass prairie, and contains a diverse mix of perennial warm season grasses (e.g., *Andropogon gerardii, Bouteloua curtipendula , Chondrosum gracile, Schizachyrium scoparium*), perennial cool season grasses (e.g., *Elymus elymoides, Hesperostipa comata, Koeleria macanthra, Poa compressa*), a variety of forbs, and a few annuals (e.g., *Bromus japonicas, Vulpia octoflora, Plantago patagonica*). This system receives less precipitation than most tallgrass prairie systems; average annual precipitation is approximately 487mm, with nearly half (214mm) occurring during the summer growing season (1 May – 31 Aug) (1894-2017, NOAA ESRL Historic Climate Data).

The study is located on the Rocky Flats soil surface (alluvium from the Rocky Mountain range deposited 1-2 Ma years ago (Madole 1991), and soils are classified as Nederland very cobbly sandy loam (NRCS Soil Survey). By spring 2019, we should be able to report site-specific soils information (soils collected <100m from plots), including soil texture, N, and C (organic and inorganic pools) to a depth of 20cm (personal communication, T. Seastedt collections for GMAP soil surveying efforts). To capture within-site soil heterogeneity, we will also assess soil texture to a depth of 10cm from each rainfall control plot (n=24, see below for experimental design) by January 2019. Precipitation and other meteorological data are obtained from a weather station <1km from the study site (1 minute data resolutionJager and Andreas 2018).

Experimental Design

To carry out cattle grazing and rainfall manipulations, we established a fenced grazing exclosure (160m x 40m) divided into six consecutive blocks with interior barbed wire fences (each block approx. 27m x 40m) (Fig. 1). To assess effects of grazing timing, we assigned each block to one of three grazing treatments (2 blocks per treatment): no grazing, spring grazing (within the period of Apr-Jun), or fall grazing (within the period of Sep-Dec). To implement these cattle grazing treatments, the exclosure was constructed with removable exterior walls (comprised of 16' x 4' cattle panels), allowing us to control cattle access to individual blocks (Fig. 2).

Within each grazing block, twelve plots (3m x 2.3m) were laid out on a grid, with a 5m buffer between plots in the dominant windward direction (to avoid rain blocking effects of shelters) and a 3m buffer between plots in the non-windward direction (Fig 3). We randomly assigned plots to one of three treatments: dry (covered by rainout shelters), wet (receiving additional water captured in dry treatment), or ambient (no alteration). Within each plot, we have installed a permanent $1m^2$ subplot for annual vegetation mapping and seedbank sampling (see 'Vegetation Sampling'). These subplots are located at least 50cm from all plot edges and marked with nails in opposite corners. The rest of the 3m x 2.3m plots remain open for plant biomass samples and other types of experiments.

Rainfall Treatment.

For the dry treatment, rainfall shelters (3m x 2.3m coverage area) were arranged so that the downward slope faces due west (the primary direction of incoming precipitation) (Fig. 4a). Shelter roofs were constructed of clear acrylic strips bent into a V-shape (2.4m x .11m) and spaced to exclude 66% of precipitation. Roofs were angled at approx. 12.5 deg, and runoff was captured by gutters and drained into storage bins (55 gal) for irrigation of wet plots (described below). Because this is a short-statured grassland, shelters stood at a minimum height of 50cm. Shelter roofs were installed on 22 May 2018, initiating the experiment.

For the wet treatment, water was added back within 48 hours of a rain event resulting in the accumulation of at least 5mm of rain. Water addition started in the early morning (typically 7-8am) and the starting block was staggered to minimize biased evaporative effects of mid-day water addition (typically complete by 11am-12pm). Water was added via an overhead sprinkler system (four 90deg sprinkler heads on 30cm risers, one per plot corner) hooked up to a battery-powered transfer pump (Wayne PC1 12V transfer pump) (Fig. 4b). Sprinkler systems were only in place during water addition events. In the situation that a storage bin failed to capture water (e.g., via a gutter leak or infrastructural damage), rainwater was added to bring bin totals within approx. 2 standard deviations of the average captured amount.

To assess treatment efficacy, we monitored soil volumetric water content (VWC , %) periodically in 36 randomly selected plots (n=4 per rainfall x grazing treatment combination). To do this, we installed two permanent 12.7cm nails flush with the soil surface in the middle of each plot to be used as paired 'probes' for repeat sampling with a handheld two-prong TDR soil moisture sensor (Campbell Scientific Hydrosense II). Preliminary results suggest that this nail-based system is effective at capturing relative differences in soil moisture (see 'Preliminary Results'). We will perform a more thorough field-calibration in spring 2019 to try to map changes in measured VWC to known increases in soil water content.

To quantify potential edge effects (i.e. less rainfall exclusion at shelter edges, or less water added along 'wet' plot edges), half of the 36 plots also had nails installed at a plot corner and at the edge of the buffer zone (50cm in from each plot side). These allowed us to follow soil moisture effects across the plot surface over the growing season. We also directly quantified edge effects on the rainfall received underneath shelters during a rain event in mid-June. To do this, we randomly selected three rainfall shelters from across the site and installed water collection cups systematically underneath and outside of rainfall shelters prior to the precipitation event (Fig. 5a). Collection cups were located at plot corners, 25cm and 50cm inside the shelter edge, and in the shelter center (Fig. 5b). The amount of rainfall captured outside of the shelter was used to quantify the percent rainfall reduction at various locations underneath the shelters.

To quantify rainfall shelter effects on light and temperature, we sampled these metrics at mid-day (11:45 –2:30pm) on a clear sunny day in late July 2018. Measurements were taken in half of the plots per block (2 plots per rainfall treatment, matching those plots with soil moisture nails installed). In this timeframe, data could only be collected for four of the six blocks (n=24, or 8 plots per rainfall treatment). Because grazing had not yet been implemented, this sample size should be useful for a preliminary understanding of rainfall treatment effects, but we intend to repeat sampling in full in 2019. We sampled temperature (deg C) at the center of each plot, 5cm above the ground, with a digital temperature probe while protecting the sensor from direct light and wind. We sampled light with a quantum meter (micromols m^{-2} s^{-1}; Apogee Instruments) at five sampling points across each plot, and use the plot average in analyses.

Graze treatment

In the first year of the experiment, the fall grazing treatment was applied for 3 weeks in October (6 Oct - 28 Oct 2018) and for an additional month in December (29 Nov to TBD). We anticipate that the first spring grazing treatment will be applied in May-June 2019, and will coordinate closely with OSMP to ensure that this treatment can be implemented. We anticipate that all infrastructure within the grazed blocks (see Rainfall Treatment) will be removed during grazing periods. Because the fall 2018 graze treatment occurred after the growing season, rainfall treatment infrastructure was already removed and all plots received the same exposure to climatic conditions. When the spring graze overlaps with the rainfall treatment implementation window (Beginning 1 May), we will open grazing blocks only during weeks with relatively little rainfall expected. During this time, we anticipate removing all infrastructure from grazed blocks, and removing only the rainfall shelter roofs from all other blocks. While we may miss a few small rain events, this will ensure that all plots receive similar climatic exposure regardless of grazing treatment.

To quantify the effects of graze treatments on total plant production, we will sample vegetation in grazed blocks, ungrazed blocks, and adjacent external grazed areas (~40m from the site fence, one block-length away). We are initially sampling both plant residual dry matter (0.05m^2 standing plant biomass harvests at each plot) and vegetation height (estimated at 10 random points stratified across the 0.05cm^2 sampling area) at each control plot within the site (n=24 total; we will not yet attempt to account for rainfall interactions) and at one sampling plot north and south of each block (n=12 external sampling points). We will use results from the first fall and spring grazes to define future vegetation height and residual dry matter targets, and develop general rules for how long to open exclosures (see Preliminary Results for data available thus far).

Vegetation Sampling

In addition to collecting community-level plant height and standing biomass samples following graze periods (see 'Grazing Treatment'), we also collected this data in August 2018 to assess the initial effects of rainfall on plant production. This timing was meant to occur after the bulk of growing precipitation has fallen but before the fall graze. We followed similar protocols as specified for the grazing treatment, clipping this-year's standing plant biomass in 10cm x 50m frames (0.05m2) in each plot (n=72 across all grazing x rainfall treatment combinations). Frames were placed adjacent to thepermanent 1m^2 subplots. In the future, the exact location of biomass sampling frames will be rotated to avoid repeat biomass sampling.

To follow plant recruitment, growth, and survival, we mapped all individuals within a 0.5m2 portion of each 1m^2 subplot. Using a gridded quadrat, all individuals were mapped as shapes (capturing aerial cover). Shoots of the same species were assumed to be separate individuals if separated by at least 2.5cm aboveground. If individuals of a particular species could not be distinguished by this rule and/or too abundant to be mapped as individuals, we mapped total species coverage in the subplot and counted flowering heads in a known area to generate estimates of genet density and total cover (*Poa compressa* in all plots; *Bromus japonicus*, *Vulpia octoflora* in some plots). Plot mapping occurred mid-summer (12 Jun – 29 Jun 2018), between anticipated spring and fall graze periods. To avoid bias based on sampling date, we staggered and randomized the order of plot mapping within the sampling period across blocks in the first year, and will follow this same sampling order in future years.

Baseline seedbank samples were collected on 5 June 2018. For each plot, we pooled three 5cm (diam) x 2cm (depth) samples taken haphazardly along three sides of each plot (within 20cm of edge). We repeated seedbank sampling in the fall as a post-growing season metric of reproductive success (Nov 2018). We followed similar protocols, but pooled three samples taken from the non-mapped portion of permanent 1m^2 subplots (an adjacent 0.5m^2 area). We anticipate utilizing these non-mapped halves of permanent plots for seedbank samples and other minor destructive sampling that should not affect recruitment and survival in the mapped portion of plots.

Analyses

We tested for an effect of rainfall treatment and sampling location (plot middle, buffer zone, or corner) on soil VWC in a mixed effects model with rainfall treatment, sampling location and their interaction as fixed effects and plot nested within block as a random effect (random intercepts). Expecting different edge effects in wet and dry plots (drier and wetter edges, respectively), we used multiple comparisons (emmeans package in R, with Tukey adjustment) to assess sampling location effects within rainfall treatments.

We tested for an effect of rainfall treatment on standing plant biomass in August using linear fixed effects models. Because no grazing had been implemented at this point, graze treatment was not included in the model. We compared a full model with rainfall treatment as a fixed effect and block as a random effect (random intercepts) to a null model including only a random block effect only. We then used a log-likelihood ratio test and AIC values to ask whether the full model outperformed the null (i.e. significant effect of rainfall treatment).

We used a similar approach to test grazing effects on standing plant biomass and plant height after the first 3 week fall graze period in October (note that external sampling sites had received 6 weeks of grazing by the vegetation sampling date). For each vegetation metric, we specified a full model including graze treatment (i.e. fall graze (within site), no graze (within site), or external graze) as a fixed effect and block as a random effect (note that external sampling sites were blocked as 'south' or 'north' of site). We then compared full models to a null model including only block as a random effect. When the full model out-performed the null model, we used multiple contrasts (emmeans package in R, with Tukey adjustment) to evaluate group differences.

Finally, we used simple linear regression to assess whether plant height could serve as a good proxy of treatment effects on plant biomass in the future. We use only plant biomass and height data from the November 2018 sampling date (mid-fall-graze).

Preliminary Results

Rainfall Treatment – Environmental and edge effects

Rainout shelters appear to slightly increase ground-level temperatures (4% increase, Fig. 6A) and decrease light levels (6% decrease; Fig. 6B) relative to control plots. The target rainfall reduction was 66% (i.e. dry plots receive 34% of ambient rainfall). Rainfall sampling under the shelters suggested that we reached these targets with a slight edge effect, where rainfall was reduced to 48% of ambient (on average) at 25cm inside the shelter edge and to 36% of ambient at 50cm inside the shelter edge (Fig 5B). Similarly, soil moisture data suggest slightly wetter soils at plot edges in the dry treatment (Fig. 7A), and slightly drier soils at plot edges in the wet treatment (Fig. 7B). However, sampling location effects (plot middle, buffer, or edge) were not significant within or across treatments.

Rain events & Soil moisture

The study site received approximately 480mm of precipitation in 2018 (through early December), with nearly 150mm falling during the rainfall manipulation period (22 May – 15 Sep; Figure 8). We expect to capture nearly twice as much rainfall during the manipulation period in 2019 with earlier installation of rainfall shelters (1 May). Captured rainfall was redistributed to wet plots across 7 water addition events (spanning 20 June to 7 Sept). Across sampling dates, we detected significantly lower VWC in dry plots relative to control and wet treatments ($p<.05$), but no difference between wet and control treatments (Fig. 7C, Fig. 8).

Vegetation responses

We observed a slight reduction of August standing plant biomass in dry treatments relative to control (16%) or wet (10%) treatments, but the effect of rainfall treatment was not significant (Fig. 9; full model AIC=347.87; null model AIC=347.11; Chi-sq p=.1938).

The first fall graze treatment is still in operation (Oct – Dec 2018). Initial results suggest that in three weeks (during the October graze), cattle reduced standing vegetation biomass by 19% in the grazed blocks relative to the non-grazed blocks, and by 55% outside of the grazing exclosure relative to ungrazed blocks (Fig. 10 A&B). The effect of grazing treatment on biomass was significant (Chi-sq p = .03027), but small sample sizes in this mid-way graze analysis failed to reveal significant group differences. A simple linear model (no random block effect) suggests that a significant effect of graze treatment is driven largely by lower values external to the site rather than significant differences between fall graze and no graze blocks (analysis not shown in this report). Cattle also reduced vegetation height by 22% in the grazed blocks and by 25% outside of the grazing exclosure (Fig. 10 C&D), but the overall effect of graze treatment was weaker (Chi-sq p=.09831) and multiple comparisons revealed no significant group differences.

Based on these initial grazing observations, we decided to re-open the fall graze blocks, and will report updated numbers once the 2018 fall graze is complete (anticipated in December/January). Results do suggest that in the future, plant height measurements could serve as a non-destructive proxy for plant production (i.e. standing plant biomass; $p<0.001$, $R2=0.5394$; Fig. 11). We believe this relationship could be improved with greater height sampling effort, and plan to explore the possibility during the final assessment of the fall graze treatment in January.

Plan for Completion (through 2019)

By February 2019

- Fall graze treatment completed and follow-up plant production data (biomass and height) are collected.
- Preliminary soil analyses are completed and compiled, including texture (Julie) and soil chemistry (T. Seastedt group).
- Plot maps from 2018 are digitized.
- Begin 2018 seedbank sample grow-outs in the greenhouse

May 2019

- Spring seedbank samples are collected before May 1.

- Rainfall shelters are re-constructed by May 1. I am currently seeking additional external funds to order extra materials in case of damage.
- Spring graze treatment has begun, or dates are agreed upon (prior to mid-June is preferable so that plot mapping can begin).
- Vegetative trait database completed for majority of Droughtnet species (5-10 more species must be grown out in spring of 2019 to fill in dominant species at this site).
- Meeting with City of Boulder Open Space to discuss possible management actions that could be tested within the Droughtnet study site. In the original proposal, this involved seeding treatments structured around plant traits. However, this approach could (should?) be reconsidered in light of unsuccessful seeding efforts that have occurred in a separate project across OSMP grasslands (Larson 2017 Funded Research Project). This meeting should occur by May in case seed collection is necessary to implement treatments.

<u>June-July 2019</u>

- Plot mapping is completed for Year 2.
- Repeated or new environmental sampling is conducted as needed (temperature, light, soil compaction, soil water infiltration, etc.)

<u>August 2019</u>

- Peak growing season biomass is collected for Year 2 (will capture spring graze AND rainfall treatment effects)

<u>September 2019</u>

- Rainfall shelters are removed

<u>October-December 2018</u>

- Fall seedbank sampling is completed for Year 2.
- Fall graze treatment is implemented for Year 2 if possible.

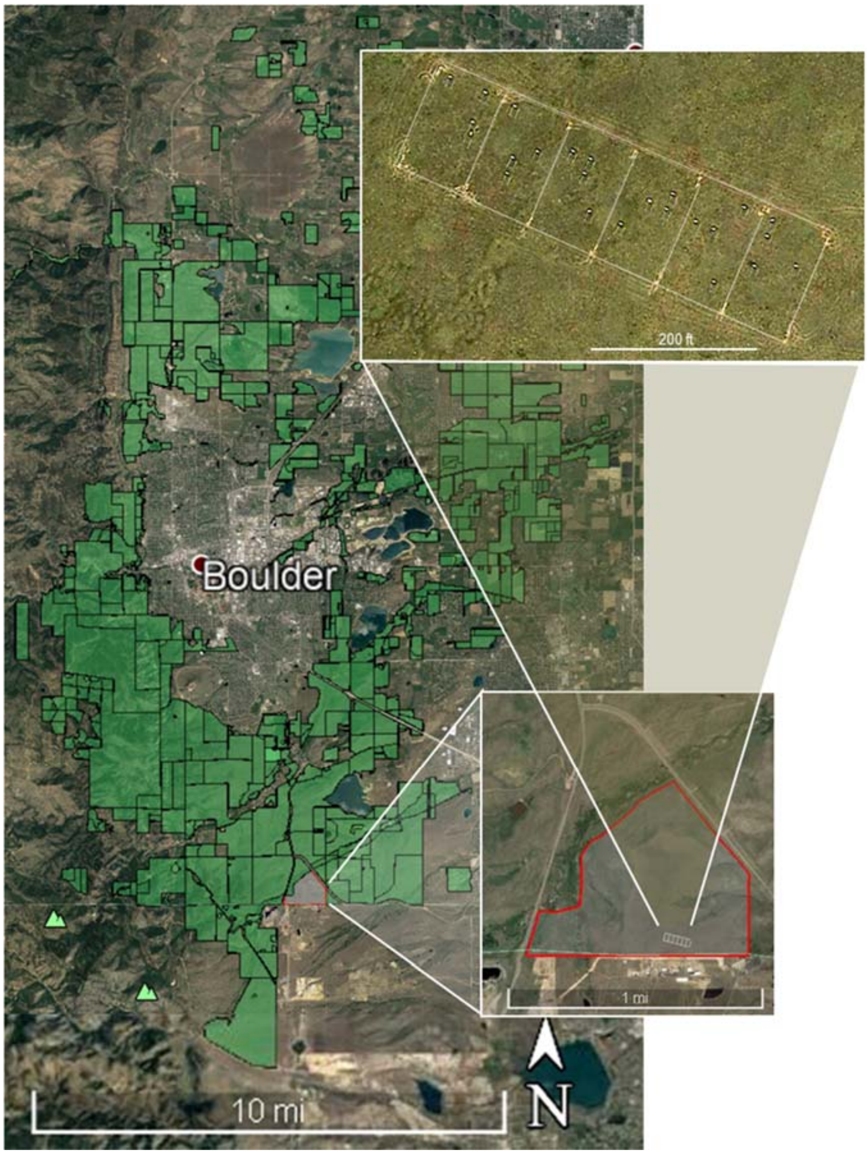


Fig. 1. Map of study site within the larger context of City of Boulder Open Space and Mountain Parks lands (green polygons). The lower inset shows the Tracy Collins property (100ha fenced parcel) where the site is located. The upper inset shows a closer view of the study site within the property. Four rainfall shelters per grazing block can be seen in this aerial image (aligning with the plot map in Fig. 3).

Fig. 2. Grazing blocks were constructed with permanent interior fences (barbed wire supported by H braces; black arrow) and removable exterior fences (cattle panels; white arrow).

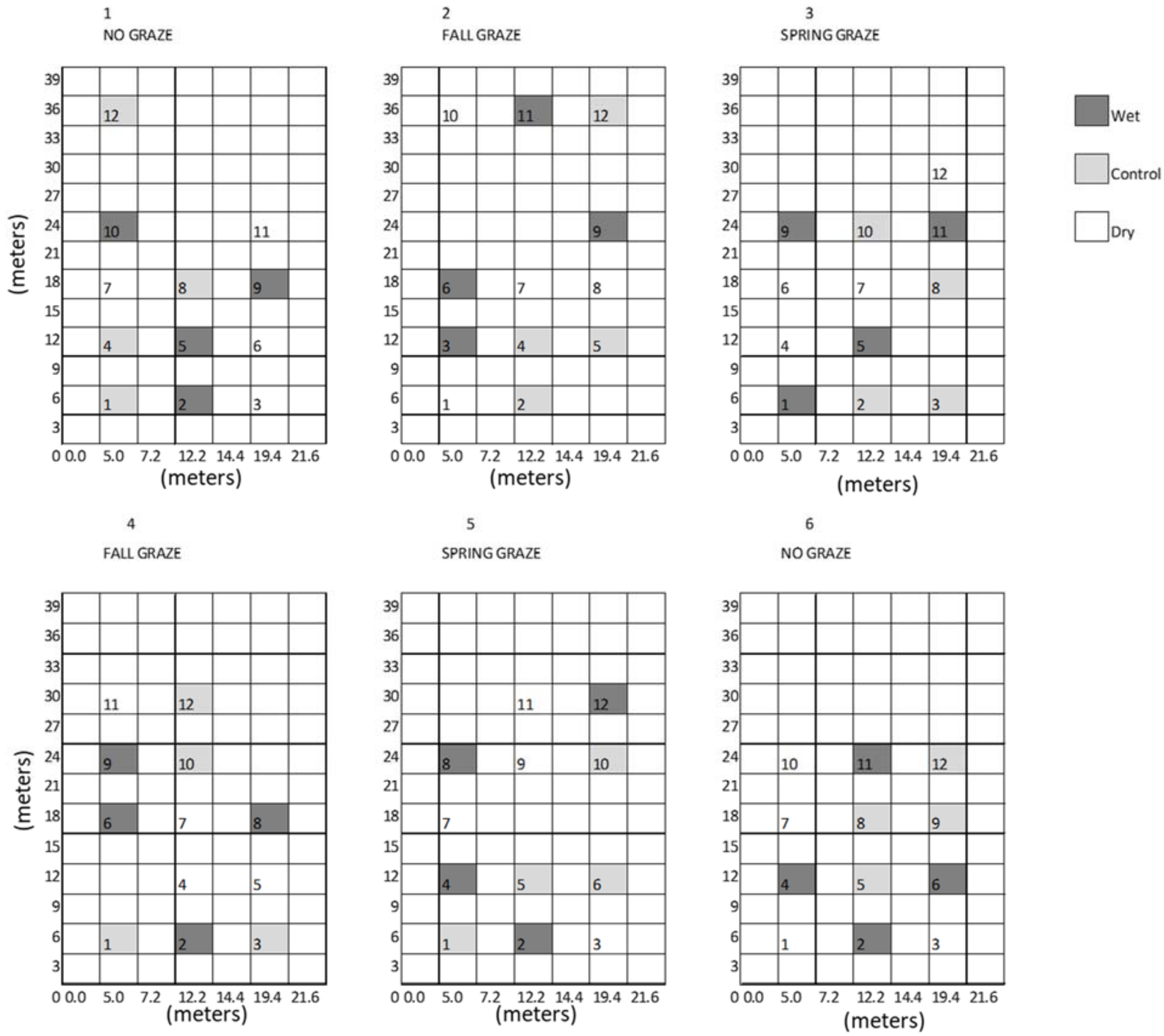
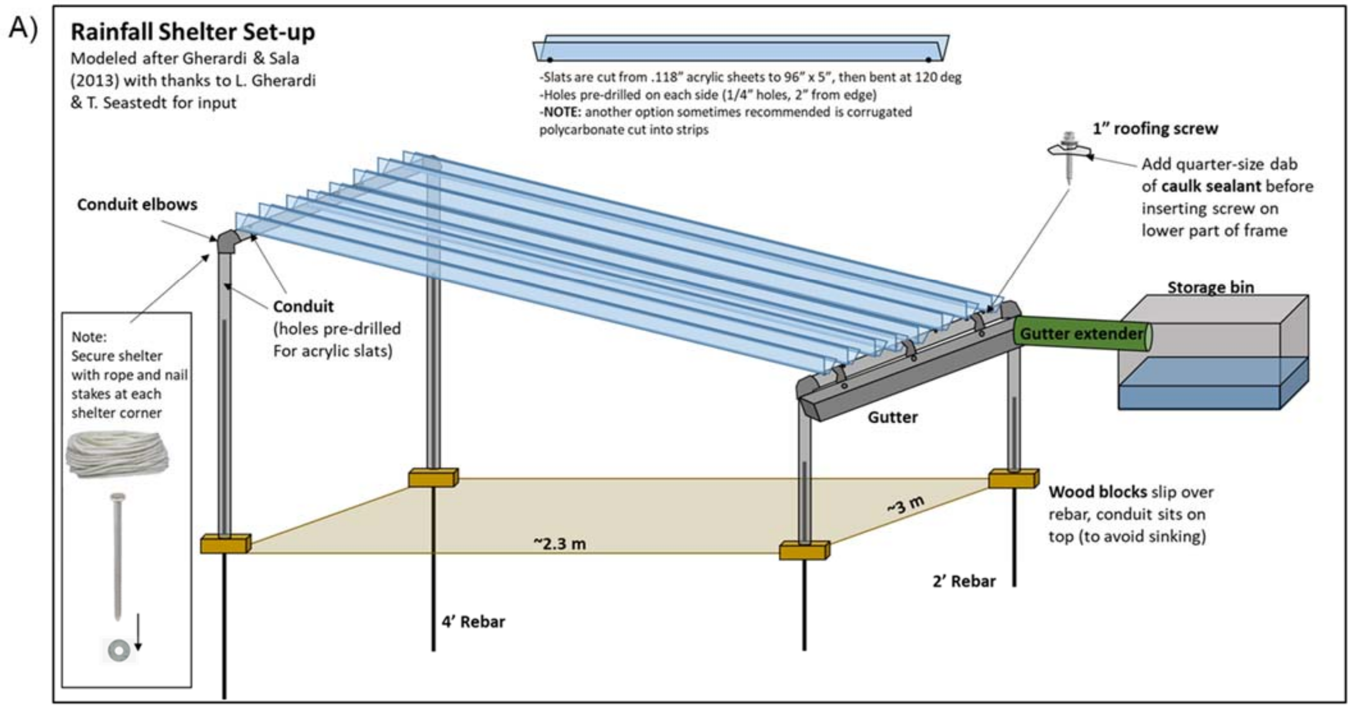


Fig. 3. Site map showing the spatial arrangement of plots (numbered cells, n=72) within blocks (large squares (n=6, with 12 plots per block). Grazing treatments are applied at the block-level (assignments shown above each block) and rainfall treatments are applied at the plot level (indicated by shading). Within each block, plots were randomly assigned to one of three rainfall treatments, with the constraint that each dry plot must be next to its own water plot (for ease of water addition). The plot map is spatially explicit, and shows where certain areas of blocks have been skipped over to avoid mounds or other anomalies.



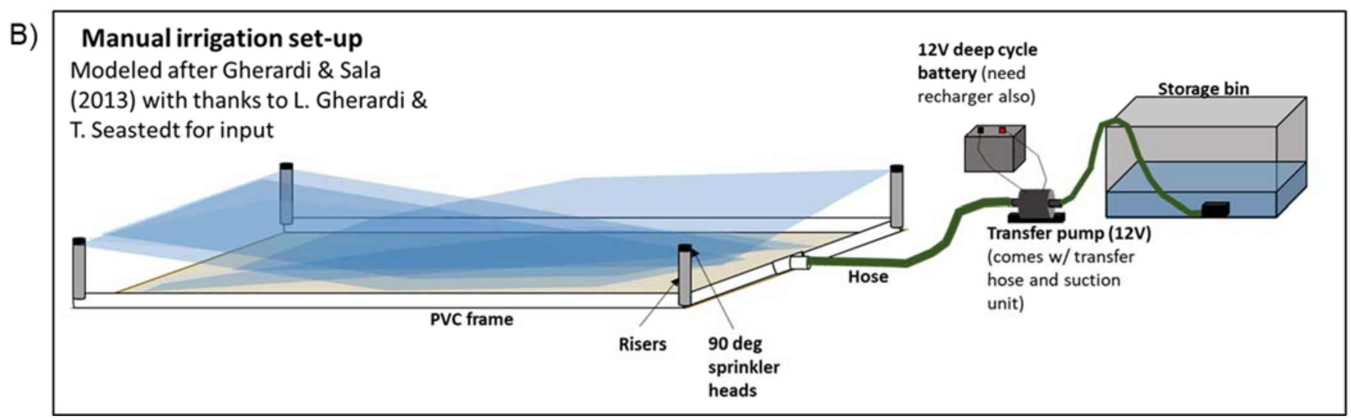


Fig. 4A & B. Construction specs for A) rainfall shelters and B) manual irrigation system.

A)

B)

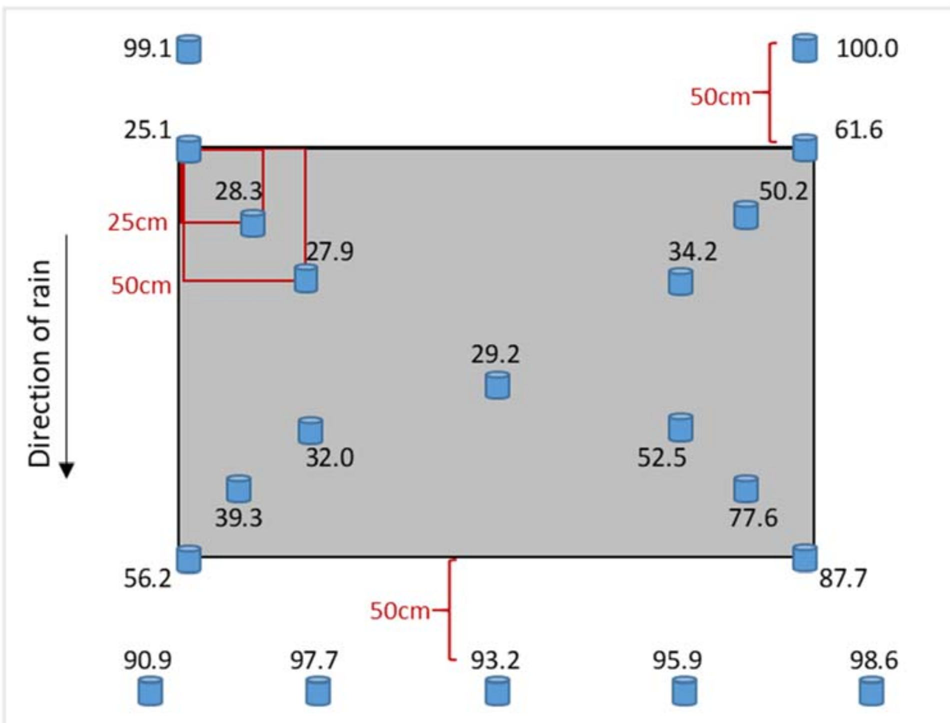


Fig. 5A & B. Sampling design for rainfall shelter edge effects on precipitation. A) Sampling cups were placed at ground level prior to a June 2018 rain event to quantify shelter rainfall interception and detect edge effects. B) Percentage of water captured in each sampling cup relative to the maximum amount of rainfall captured in external sampling cups (target reduction = 34% of average). Values shown for each sampling cup are averaged across three shelter replicates.

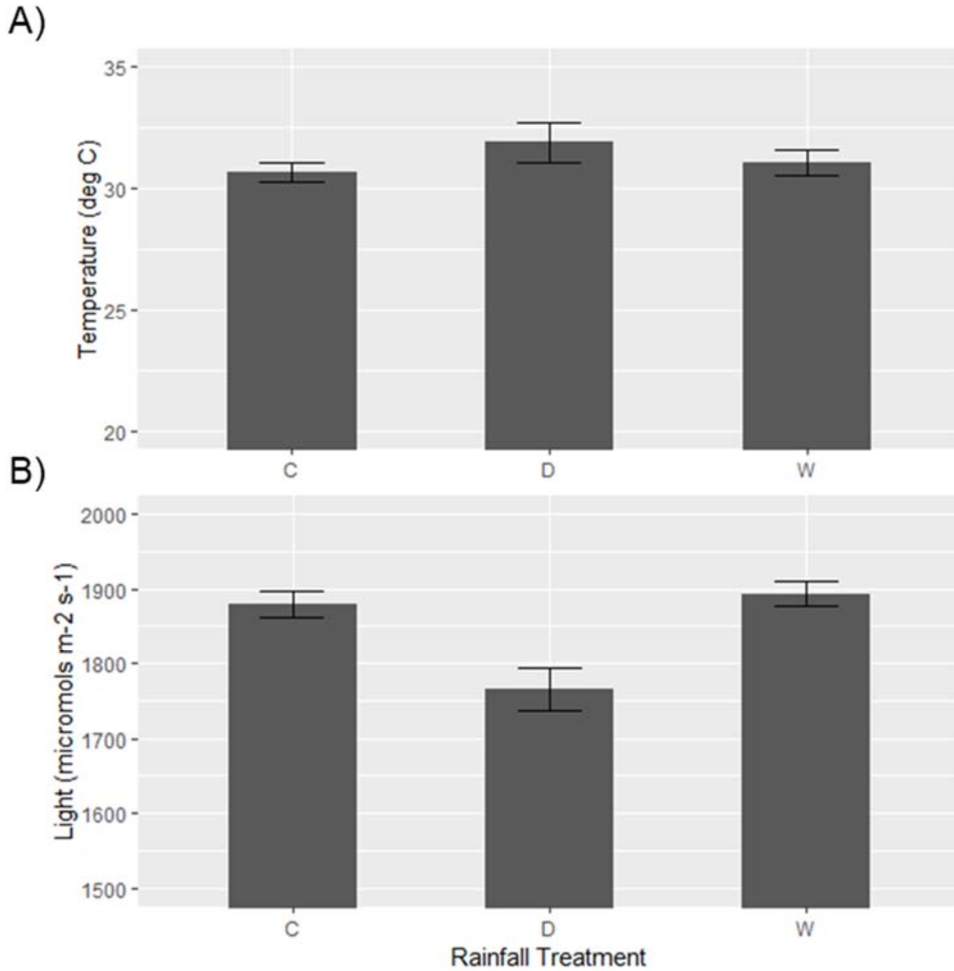


Fig. 6A & B. Rainfall shelter effects on A) temperature and B) light on a clear day in late July 2018. Effects in drought plots (D) are shown relative to ambient rainfall (C) and wet (W) plots, both of which lack rainfall shelters.

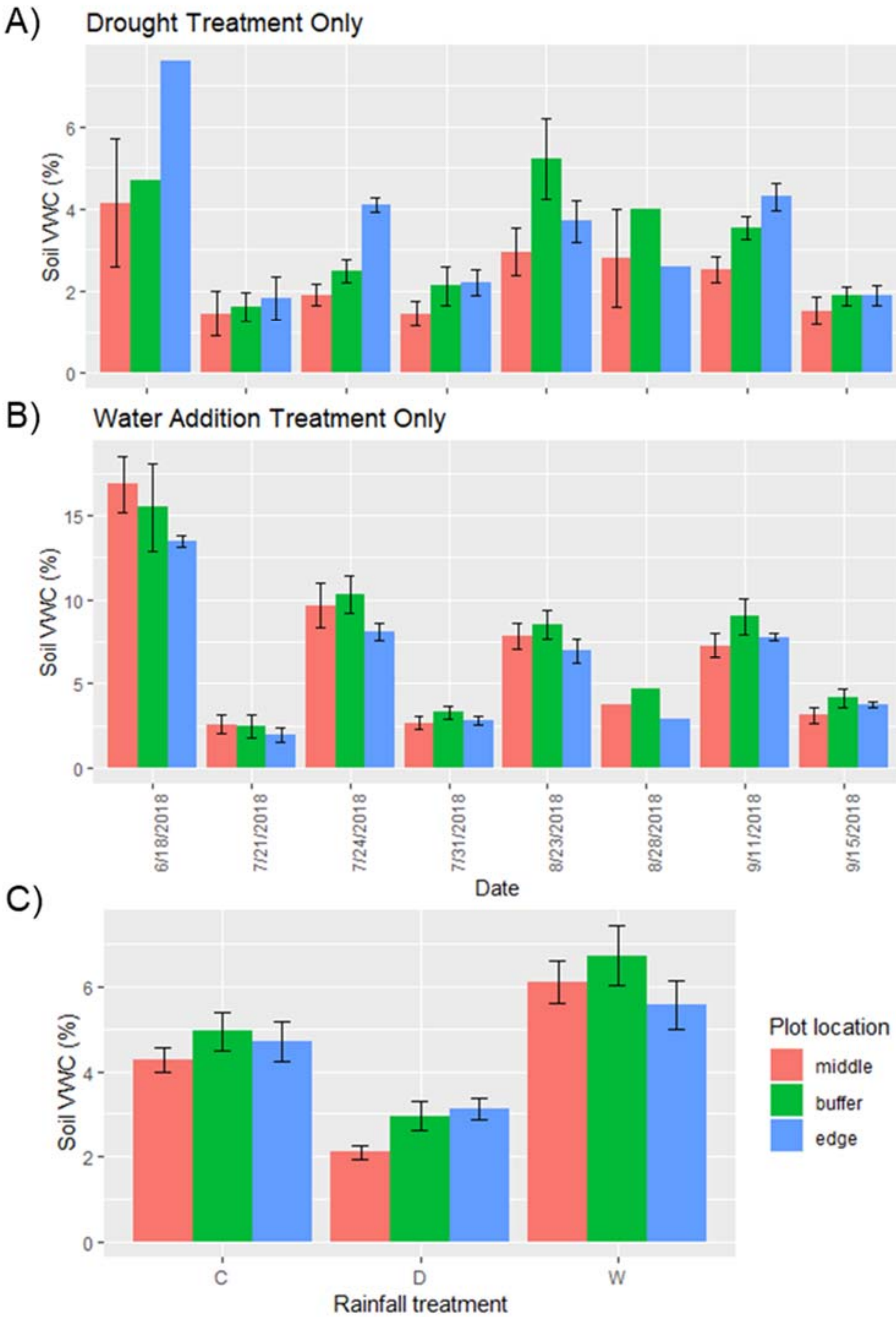


Fig. 7. Effects rainfall treatment and within-plot location (i.e. edge effects) on soil volumetric water content (VWC) across 8 sampling dates. Edge effects result in slightly wetter soils at plot edges in the dry treatment (A) and slightly drier soils at plot edges in the wet treatment (B), although these differences were not significant. (C) Across sampling dates and sampling locations, strong differences in VWC were detected across rainfall treatments, outweighing any edge effects. In the plot middle, VWC in the dry treatment different significantly from both the control (p=.0223) and wet (p<0.001) treatment, although control and wet plots were not significantly different (p =.1360).

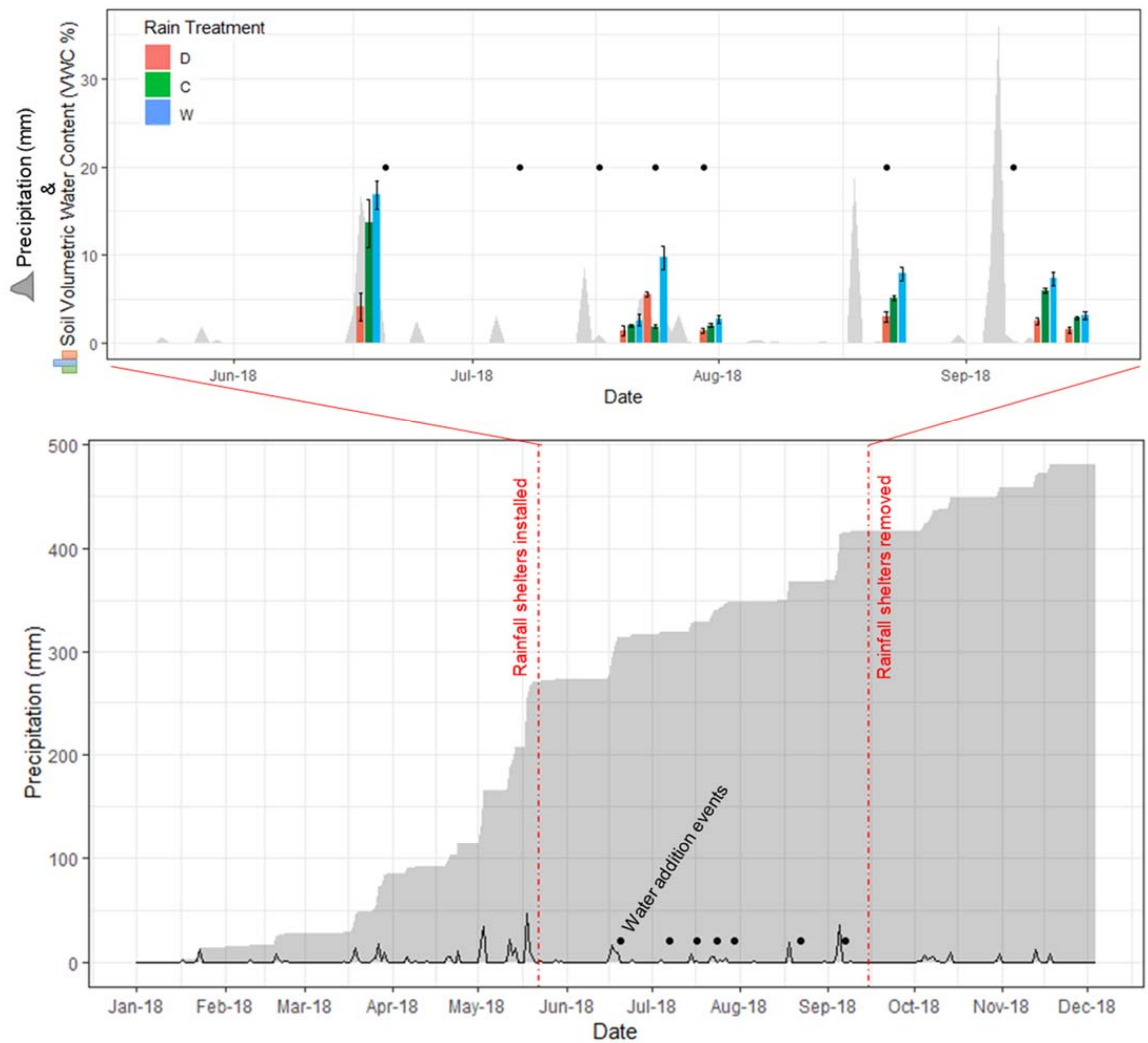


Fig. 8. Lower panel. Cumulative precipitation (gray shading) and individual rain events (black line) at the study site across 2018. Rainfall shelters were installed on 22 May and removed on 15 September (red lines). A large portion of May rainfall events were missed due to constraints with first year installation timing (approx. 150mm). **Upper panel.** 145mm of precipitation fell during the rainfall manipulation period, which was re-distributed to wet plots across 7 water addition events (black circles). Across sampling days, mid-plot soil volumetric water content (colored bars) tended to be lower in dry plots and higher in wet plots (see also Fig. 7).

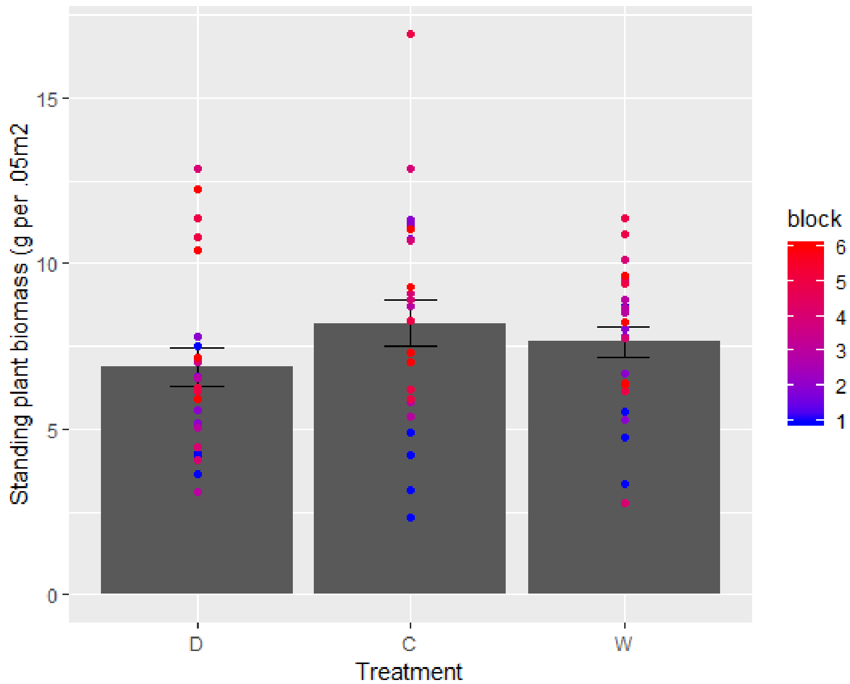


Fig. 9. Effect of rainfall treatment on standing plant biomass (current year's growth) during peak growing season (early Aug. 2018.) Samples from individual plots are overlaid as points and colored by block to show trends in biomass that are independently associated with block (a steady decline moving east across the site, from block 6 to block 1). Note that biomass samples were collected one month prior to rainfall shelter removal, so full rainfall effects during 2018 may not be captured completely.

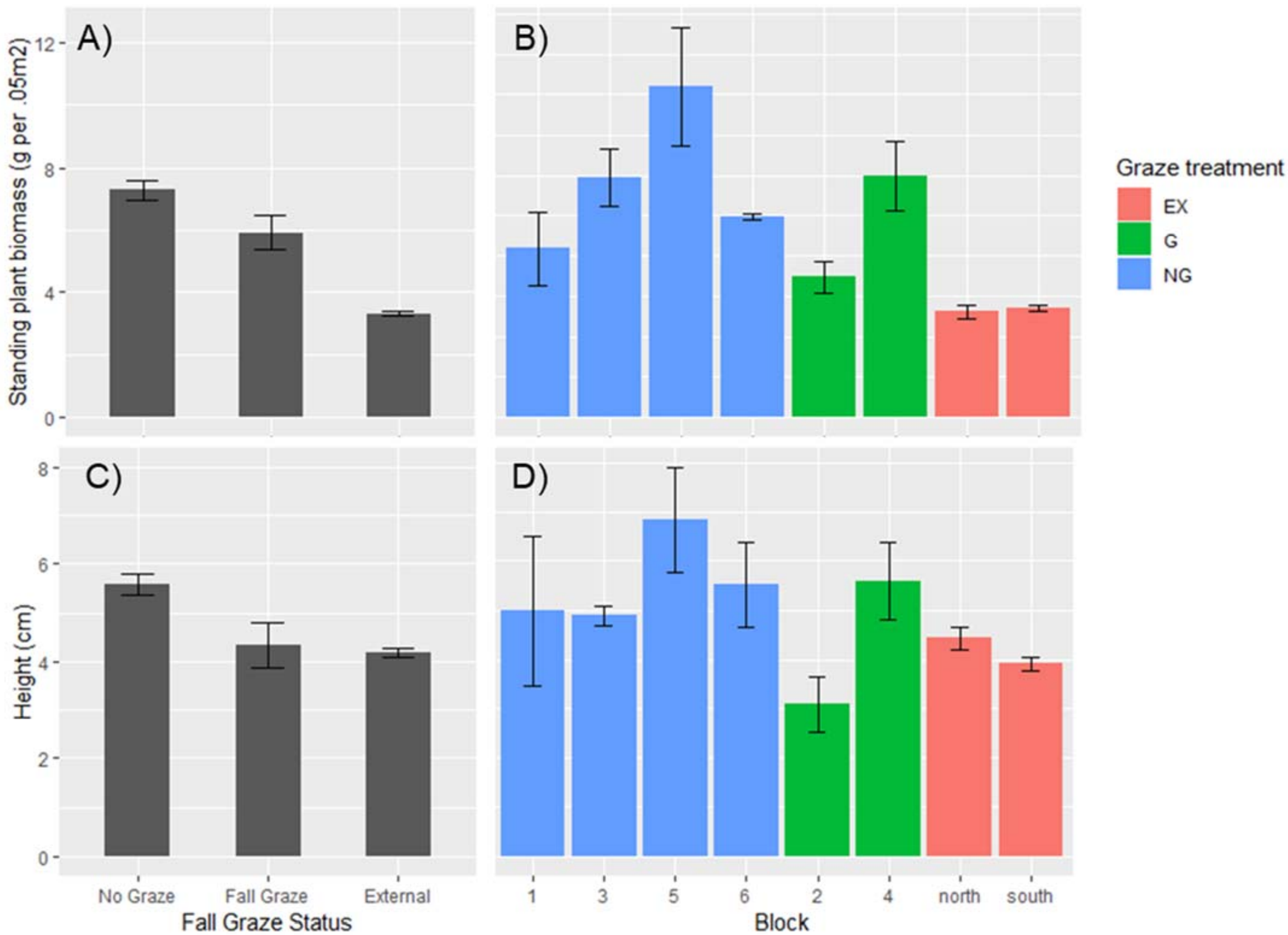


Fig. 10A – D. Initial effects of the fall graze treatment on plant biomass (A & B) and plant height (C & D) over a three week graze in October 2018 (external sampling locations [pink bars] grazed for 6 weeks). A) Standing plant biomass (current year's growth) within no-graze blocks (n=8 plots), fall graze blocks (n=4) and external sampling sites (n=12, approx.. 40m away from the study site). B) Data from (A) are further separate to see differences between blocks. C) Average plant height within no-graze blocks (n=8 plots), fall graze blocks (n=4) and external sampling sites (n=12). D) Data from (C) are further separate to see differences between blocks (including external sampling sites north or south of the site).

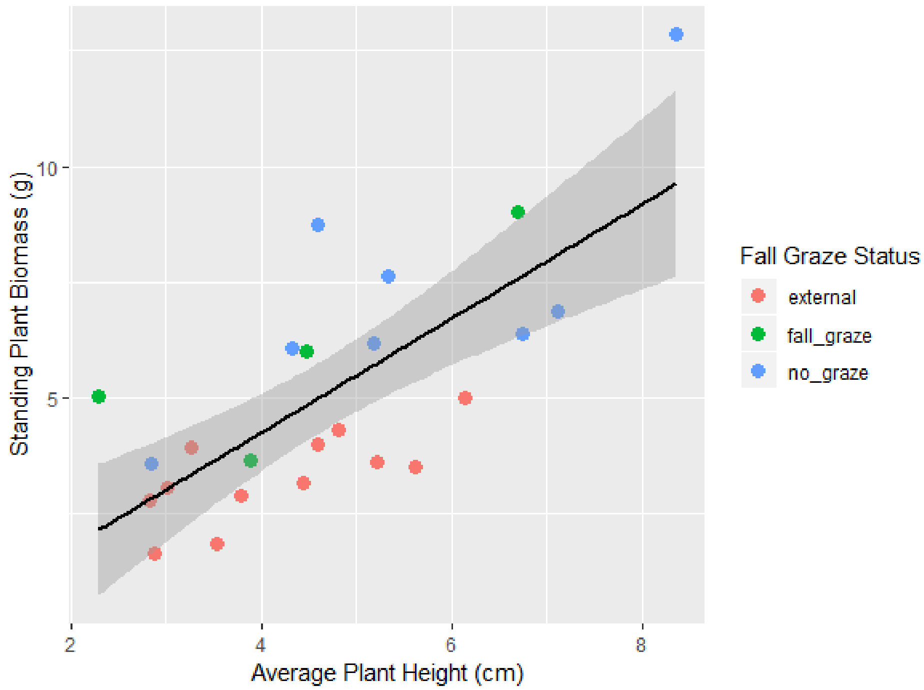


Fig. 11. Relationship between community-level plant height (each point is average of 10 samples per .05m2) and standing plant biomass (current year's growth clipped to the surface in the same .05m2 area). The relationship is strong ($p<0.001$, $R2=0.53$), and we will seek to improve it with greater sampling effort moving forward.

References

Jager, D., and A. Andreas. 2018. NREL National Wind Technology Center (NWTC): M2 Tower; Boulder, Colorado (Data). NREL Report No. DA-5500-56489.

Madole, R. F. 1991. Quaternary geology of the northern Great Plains, Colorado Piedmont section. Pages 456-462 *Quaternary nonglacial geology; conterminous United States, Geological Society of America. Geology of North America*.