Land management controls on soil carbon fluxes in Asia's largest tropical grassland

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MB, CM and ATV conceived and designed the research; MB and CM collected the data; MB and CM analyzed the data; MB, CM, AJH and ATV wrote and edited the manuscript.

Abstract

Land management changes threaten the ecological functionality of grassland ecosystems worldwide, including the ability of these ecosystems to store carbon in their soils. The Banni grasslands, Asia's largest tropical grassland, is no different in this regard. Despite being a highly biodiverse ecosystem with an extensive land use history, information on soil carbon fluxes in this ecosystem remains conspicuously absent. In this study, we map soil organic carbon (SOC) stocks within the upper 30cm depth across land use-land cover (LULC) classes by combining high-resolution satellite imagery with field-based soil carbon data from a network of long-term monitoring sites. We find that Banni currently stores 27.69 MtC in its soils, with an average SOC density of 119.61 ± 3.50 tC/ha (Mean ± Std. Error). These estimates compare favourably with arid and semi-arid grasslands as well as seasonally-flooded savannas across the tropics. The highest SOC densities are found in restored grasslands (142.72 ± 5.72 tC/ha), reaffirming the potential of carbon recovery from ecological restoration. Tracing 10 years of LULC change in the region indicates the further expansion of the invasive tree *Prosopis juliflora* across the landscape, but its impacts on changes in SOC concentrations estimates remain varied. Our results indicate the large soil carbon storage potential and the SOC fluxes associated with woody encroachment in Banni, and point towards the need for the active management of Banni so that carbon benefits can accrue at meaningful timescales for reversing land degradation and contributing to land-based climate action.

1. Introduction

The soil organic carbon (SOC) pool is one of the most important reservoirs in the global carbon cycle (Lal, 2004; Lal et al., 2015). This reservoir contains more carbon than the atmospheric pool and the biotic pool combined. The role of the SOC pool is considered as a potential sink of greenhouse gases. About one-third of this soil carbon pool is currently found in grassland ecosystems, including savannas with scattered trees and open-canopy grassy woodlands. These ecosystems cover approximately 20% of the tropics (Buisson et al., 2019), playing a vital role in soil carbon sequestration in addition to providing several provisioning, regulating and cultural ecosystem services (Bai and Cotrufo, 2022).

These tropical grasslands are undergoing fundamental transformations in land use, leading to changes in their structure and function (Buisson et al., 2022, 2020). These land use changes include both land cover changes, for example, changes in canopy cover through woody encroachment, as well as land management changes, for examples, land use intensity changes within the same land cover type (Erb et al., 2017). These changes are being largely driven by anthropogenic activities, including grazing pressure, increasing soil salinity and the introduction of invasive tree species (Dakhil et al., 2021; Dayal et al., 2018; Joshi and Kiran, 2021; Mbaabu et al., 2020). Despite their crucial role in providing various ecosystem services and supporting local livelihoods, major knowledge gaps exist in our qualitative and quantitative understanding of the carbon storage and the rates of carbon fluxes across different land use types in these ecosystems (Bossio et al., 2020; Stockmann et al., 2013; Tian et al., 2023; Zhou et al., 2023).

Situated in the western-most part of mainland India over an area of more than 2,300 km^2, the Banni grasslands (hereafter, Banni) is Asia's largest tropical grassland. Despite harsh environmental features and the high salinity of its soils, Banni is endowed with rich floral and faunal biodiversity and has often been considered Asia's finest grassland (Dayal et al., 2018; Joshi et al., 2009; Manjunatha et al., 2022). It consists of two ecosystems in juxtaposition, i.e., grasslands and wetlands, which are extensively populated by pastoralist communities (Joshi et al., 2009).

Significant land use changes have occurred in Banni grasslands after India's independence in 1947, including its declaration as a Protected Forest in 1955, shifts in community grazing rights, increasing soil salinity and the introduction of *Prosopis juliflora (*hereafter, *Prosopis)* in the 2^{nd} half of the 20^{th} century (Manjunatha et al., 2022). Over time, saline brushlands and grasslands with mixed stands of *Prosopis* has become the most widespread ecosystem in Banni. likely playing an important role in SOC storage as well. Although *Prosopis* establishment may contribute to increases in SOC (Bhojvaid and Timmer, 1998), this must be balanced against its negative effects on grasslands, which are among the vegetation types with the highest SOC storage capacities (Bossio et al., 2020; Jobbágy and Jackson, 2000).

Till now, no study has attempted to establish estimates of SOC stocks in this grassland ecosystem, address the distribution of SOC and assess SOC fluxes accruing due to land cover and land management changes over time. Examining these issues through long-term monitoring plots has been known to generate robust estimates, but such monitoring plots are predominantly deployed to monitor forest carbon fluxes, and their use in grassland ecosystems has been severely limited in South and Southeast Asia (Malhi et al., 2021; Phillips et al., 1998). These monitoring plots have the potential to yield SOC accumulation estimates that are at once both geographically extensive and of high spatial and temporal resolution (Poorter et al., 2021, 2016). Moreover, few studies in South and Southeast Asia have quantified SOC in grasslands directly by monitoring sampling plots over time (Dhaulakhandi et al., 2000; Ghosh and Mahanta, 2014; Thokchom and Yadava, 2016). Existing studies have either estimated SOC as a snapshot in time or used space-for-time substitution approaches to visualize change (Bhan et al., Restoration Ecology, in press). Even global or regional meta-analyses have excluded data from Banni, making it an outlier in the study of SOC fluxes in tropical grasslands (Abdalla et al., 2018; Archer et al., 2001; Bardgett et al., 2021; Guerra et al., 2020; Phillips et al., 1998).

To bridge this knowledge gap, we present an estimate for the average SOC densities within the upper 30cm soil depth across different land use types in Banni. To produce SOC stock estimates at the landscape scale, we apply these per-hectare estimates to a high-resolution land use-land cover

(LULC) map for the year 2023. Further, recognizing the absence of systematic long-term monitoring in Banni, we trace 10 years of land use change by opportunistically resampling in SOC monitoring plots established for previous studies to account for the impacts of these LULC changes on SOC concentrations (SOC%) in Banni.

In this way, we address 2 research questions (RQ):

RQ 1: What are the current SOC stocks across different LULC classes in Banni?

RQ2: How have different land management regimes regulated grassland SOC fluxes in Banni over the last decade?

Based on these analyses, we describe the SOC dynamics of different LULC classes in Banni, paving the way for a better understanding of future land management and restoration strategies.

2. Materials and Methods

2.1 Study site

The Banni grasslands (23°19’N to 23°52’N and 68°56’E to 70°32’) is the western-most end of the Gujarat state, as well as India, and is located 60 km from the Bhuj-district headquarters. The climate of Banni is arid; its mean annual rainfall of about 300m is concentrated in the Summer Monsoon months between June and September, with a coefficient of variation of 65% and recurring droughts (Basu et al., 2019; Joshi et al., 2009).

Similar to other grasslands, vegetation in Banni is sparse and highly dependent on seasonal variations in monsoon. Banni is dominated by low-growing forbs and graminoids, many of which are halophiles (salt tolerant), as well as scattered tree cover and scrub. Prominent grass species include *Desmostachya bipinnata (Dhrab), Sporobolus marginatus (Dhrabad) and Dichanthium annulatum (Jinjvo).* Common tree species found in Banni include *Acacia nilotica subsp. indica (Bavar) and Prosopis juliflora (Gando Bavar)* (Joshi et al., 2009; Joshi and Kiran, 2021). Banni’s soils are inherently saline and consist of recent alluvium mixed at places with Aeolian sandy deposits, and the entire area has deep clayey and coarse textured soils in discontinuous patches.

2.2 Research Design

We utilized a combination of ground-based and spatial methods to investigate RQ1 and RQ2.

For RQ 1, we developed a land use-land cover classification for Banni using high-resolution Landsat imagery for the year 2023. We also estimated per-hectare values of SOC densities by estimating SOC% and soil bulk density (BD) at 30cm depths in sampling plots across the identified LULC classes. The average SOC density per hectare (sampling and carbon accounting methodology described in subsequent sections) was then multiplied by the area covered by each LULC class without considering any accumulation or depletion factors to obtain a landscape-level SOC account at 30cm depth.

For RQ 2, we compared SOC% values across 3 different timeperiods (2014, 2017 and 2023) to visualize changes in SOC concentrations across different LULC classes. We used high-resolution Landsat imagery to visualize LULC change over this 10-year timeperiod. Data on SOC% in 2014 and 2017 was obtained from earlier field investigations in sampling plots focused on developing alternative approaches to the management of *Prosopis juliflora* in Banni (Nerlekar et al., 2022) *(see Text S1)*. We were able to access some of these sampling plots during the field campaign in November 2023, allowing us to develop a temporal account of SOC% change.

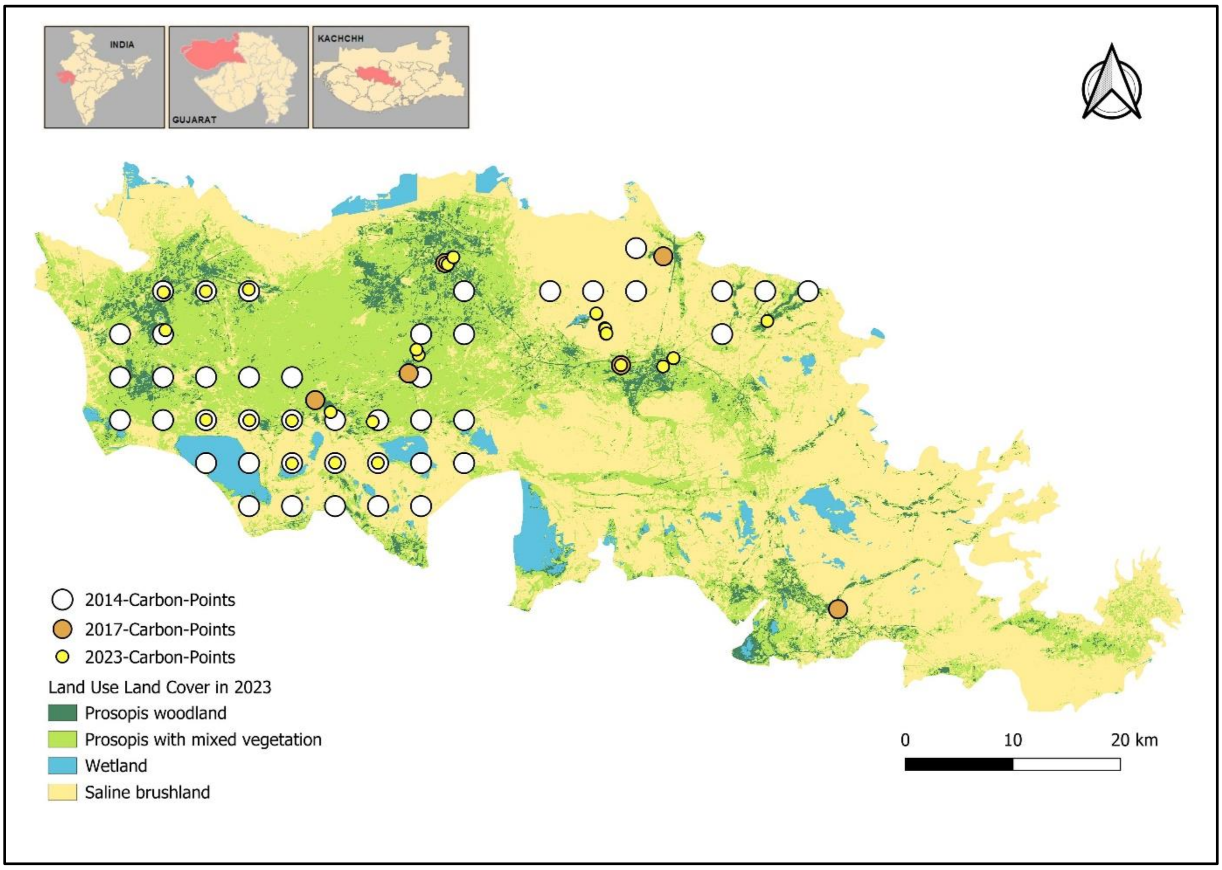

Figure 1: Map of the Banni grasslands, along with the locations of the field sampling points for 2014, 2017 and 2023.

2.3 Classification of satellite images

We performed an LULC classification for the current sampling year (2023) and for every third year since 2014 (ie. the years 2014, 2017, 2020 and 2023) to coincide with the availability of ground-based SOC% estimates using Landsat multispectral bands with a 30m spatial resolution. All images between December and March, i.e. the winter season following the monsoon (based on concurrent soil sampling), were stacked and a median composite was generated. This median layer served as the basis for training points. We employed a Support Vector Machine (SVM) classifier to generate the LULC layer in Google Earth Engine. The classification output was validated using ground truth points collected during the 2023 sampling. For previous years' images (excluding 2023), validation points were adjusted based on visual assessment of true color composites. These images were then used for buffer analysis around each sampling point. The zonal histogram tool in QGIS was utilized to determine the proportion of each land cover class within each buffer for every sampling point.

In this way, four distinct habitat types were identified in the landscape. These were: (1) Dense *Prosopis* woodlands (Pw), (2) *Prosopis* with mixed vegetation (Pm), (3) saline brushlands (Sb), and (4) wetlands (Wl).

2.4 Buffer analysis

For each point sampled in 2014 and 2017, which was subsequently resampled in 2023, we performed a buffer analysis to examine any correlation between changes in land cover and soil carbon. A total of 14 such points were included in the analysis. At each point, we created buffers of three different sizes - 60m, 80m, and 100m. Within each buffer, we calculated the area covered by each land cover type for each point in time. This process resulted in the generation of a matrix displaying the dominant land cover type (>50%) within each buffer for the years 2014, 2017, 2020, and 2023. Landsat 8 imagery with a spatial resolution of 30m was utilized for this analysis.

Subsequently, we categorized the LULC transformations at each point into two main categories: invasion-oriented and restoration-oriented. LULC change towards denser *Prosopis* stands was considered invasion-oriented, and change towards mixed grasslands was considered restoration-oriented, to correspond to the nature of restoration currently practiced in Banni (*Prosopis* removal). This helped us visualize the overall direction of LULC transition in Banni. We then correlated this transition trend with SOC% change at these sampling points to investigate whether the direction of land cover transformation at all influences SOC%. It is worth noting that our analysis was constrained by the small sample size, limiting the ability to draw statistically significant conclusions from the findings.

2.5 Restored grasslands in Banni

Banni has had a recent history of grassland restoration through *Prosopis* removal. We classified restored grasslands (locally known as *'Vaadas'*) based on the landscape's recent land cover change and land management history and by consulting leaders of local pastoralist and agro-pastoralist communities. Restored grasslands were considered those that had been restored by these communities through the manual removal of *Prosopis juliflora* over a decade ago. These are usually only a few hectares in size and fenced from all sides. They are traditionally managed for high-quality fodder, with restricted grazing for a few select cattle, and periodic harvest of standing biomass. The *Vaadas* were enclosed at least 2 decades ago (approximate range = 20–45 years), and were tilled and seeded with fodder grasses with the primary aim of forage improvement (Nerlekar et al., 2022).

We were able to physically access 6 such *Vaadas* for soil sampling based on the willingness of local village heads to collaborate with us *(Table 1)*. For the purposes of this study, these *Vaadas* were considered akin to natural old-growth grasslands of the region since it was not possible to find natural old-growth grasslands sites without the existence of Prosopis.

Because the *Vaadas* are not widespread across Banni and due to the potential for their misclassification with the *Sb* and *Pm* LULC types, we did not consider these *Vaadas* as a separate LULC type for the landscape-level SOC account for Banni. We only undertook field sampling in these *Vaadas* to estimate their typical SOC densities to illustrate differences among carbon storage capacities across LULC types and the carbon benefits that can be potentially accrued from restoration. The field sampling methodology is given in subsequent sections below.

2.6 Field Measurements of SOC%

We related observed LULC changes with observed changes in SOC% from field data collected in separate field campaigns in the years 2014, 2017 and 2023. These field measurements involved the estimation of SOC% at a depth of 30cms for 2023, but only till 10cms for 2014 and 2017, limiting the interpretation of our results of SOC% change.

2.6.1 Sampling in 2023

A total of 41 sampling plots were established in a total of 28 sites across these 5 LULC classes *(Table 1)*. The choice of these sites was determined by the location of sampling plots established in earlier field campaigns (see Sampling in 2017 and Sampling in 2014 below) and the need to make up for limited sampling sites in certain LULC classes if required *(Figure 2)*.

We established 15 plots across 6 *Vaadas* to collect soil cores. From each plot, we obtained two soil cores at a depth of 30 cm, which were then combined to create a composite sample.

For *Prosopis* woodlands, we selected four sites under dense *Prosopis* canopies. Two of these sites had been previously sampled in 2017. For the *Prosopis* with mixed vegetation class, we selected four sites overlapping with transects previously sampled in 2014. Similarly, we sampled a total of 8 sites under the saline bushland habitats, again overlapping with the 2014 sampling points. Finally, we collected soil samples from six wetland sites for SOC analysis.

2.6.2 Sampling in 2017

In a study describing Prosopis management strategies for Banni, 3 random soil samples were collected from multiple 20m x 20m plots across 8 villages *(Figure 2)* (Nerlekar et al., 2022). Due to logistical constraints and the willingness of local village heads to collaborate with us, it was only possible to visit 2 such villages in 2023 for repeat soil sampling. More information on sampling strategy and sampling protocol is provided in *Text S1*.

2.6.3 Sampling in 2014

To study the impacts of vegetation cover on soil carbon fluxes, soil sampling was conducted across 43 sampling plots to analyze SOC% across Banni in 2014. These 43 plots were selected out of 200 such points marked at 4 km intervals across the landscape. In this way, a number of points were geo-referenced across the landscape for ground truthing and for collecting soil samples to assess SOC%. The ecosystem type of each such sample point was recorded. At every plot, we established a 100m-long transect and collected soil samples from a depth of 10 cm at both ends *(Figure 2)*. These individual samples were then combined to form composite samples for subsequent SOC analysis. More information on sampling strategy and sampling protocol is provided in *Text S1*.

Table 1: Distribution of field sampling points across the 3 years – 2014, 2017 and 2023 – where field investigations were conducted in Banni.

Field-based LULC classes	2023		2017		2014	
	Site	Plot	Site	Plot	Site	Plot
Vaadas	6	15	0	0	0	0
Dense *Prosopis* woodlands	4	8	9	18	1	1
***Prosopis* with mixed vegetation**	4	4	0	0	15	15
Saline brushlands	8	8	0	0	26	26

Wetlands	6	6	0	0	1	1

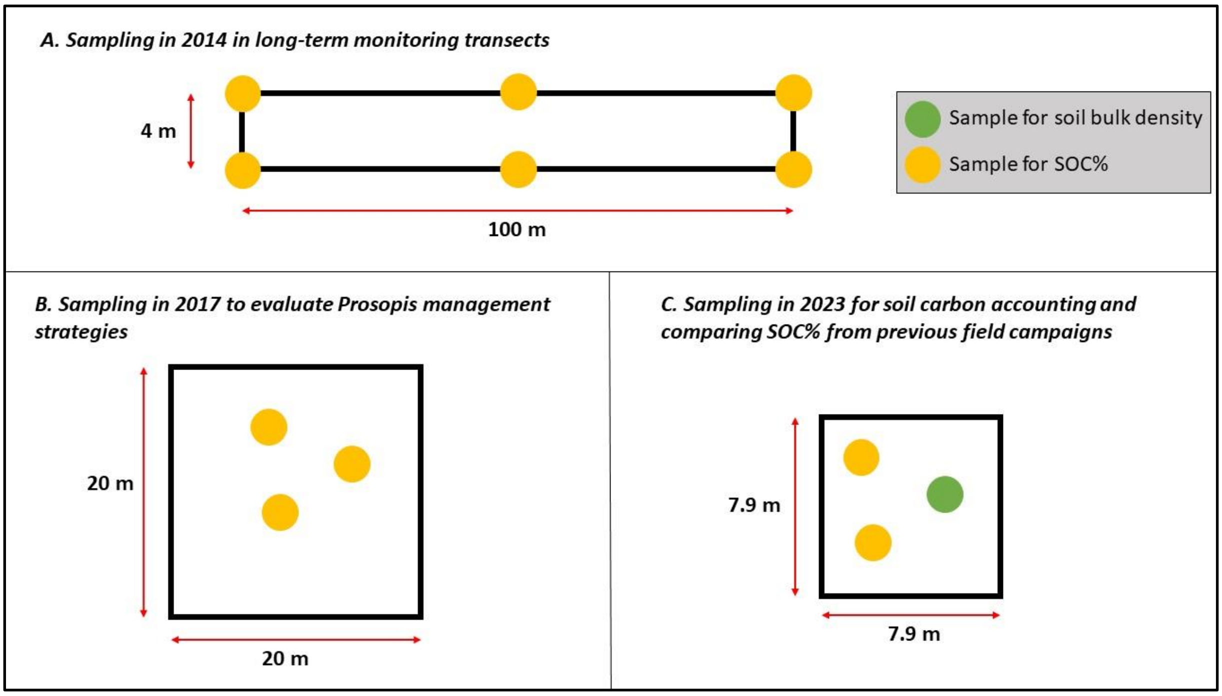

Figure 2: Stylized description of the plot design for soil sampling in 2014, 2017 and 2023. In each plot, samples for SOC% were collected at random and mixed well, and their composite for used for estimating SOC%. In 2023, a separate sample from each plot was taken for bulk density estimation as well.

2.7 Estimation of SOC densities

We estimated both the SOC% and BD of the soil samples to calculate per-hectare SOC values for the year 2023. IPCC guidelines state that it is good practice to measure the SOC pool to a depth of at least 30 cm (Eggleston et al., 2006). This is the depth where the changes in the soil carbon pool are likely to be fast enough to be detected with monitoring at realistic time intervals. It is important to note that in these landscapes, there is limited data on the depth at which SOC responds to changes in ecosystem types, management practices and disturbance regimes.

We collected soil samples by using a soil core of fixed volume (height, h = 10cm; diameter, d = 5cm). We cored the soil thrice to reach the required depth of 30 cm. We excavated around the core without disturbing or loosening the soil that it contained and carefully removed it with the soil intact.

We removed any excess soil from the outside of the soil core and cut any plants or roots off at the soil surface with scissors.

To keep the sampling comparable, soil samples were taken by laying a 100m transact at sites that were sampled in 2014 and collecting 2 soil samples from its ends. For sites sampled in 2017, we collected soil samples from 3 soil cores placed randomly within the 20x20m plot. Finally, we collected soil samples in *Vaadas* by making 7.9x7.9m sub-plots, based on previous soil sampling methodologies developed for semi-arid grasslands (Bhan et al., Restoration Ecology, in press). In all these cases, soil samples collected from each site were mixed well and their composite was collected in an airtight vial and taken back to the ATREE campus in Bengaluru for further analysis of SOC%.

To assess BD in each plot, one additional core was collected from each site. Each sample was well-mixed in the bag and clumps were broken down. The moist weight was noted using a standard weighing scale. The average weight of each soil sample was found to be ~700 gms. We took a sub-sample weighing ~200 gms for further processing. This sub-sample was oven-dried at $70^{o}C$ for 72 hours and its dry weight was recorded.

We sieved the sample using a 2mm sieve to separate fine earth particles from the coarse mineral fraction. Evidence suggests that the coarse mineral fraction has a negligible capacity to store carbon, therefore it was removed before analysis and SOC% was measured for the fine earth fraction (FAO, 2019).

SOC % analysis: We determined the SOC% of each soil composite using the combustion gas chromatography method in a CHNS analyzer. Approx. ~2 gm of the section was dried for 1 hour at $105^{o}C$. A small proportion of soil (0.110 - 0.111 mg) was weighed and packed into a small tin foil to be inserted into the CNHS analyzer to get the SOC % value *(Figure 3)*.

Bulk density: BD is the mass per unit volume of the soil. Here, we estimated $BDfine_2$, denoted as the mass of fine earth per total volume of the soil sample. To estimate the mass of fine earth particles in each soil core, we oven-dried the soil section again at $105^{o}C$ for 24 hours to ensure complete loss of moisture. Comparison of the final weight of the section with the wet weight of the sample allowed us to estimate the proportion of moisture content in the soil section. As we had taken a random sample from the original soil core, we assumed that the original soil core collected on site would have the same moisture content as the sub-sample. In this way, we could calculate the dry weight of the fine earth particles of the original soil core. We combined that with the volume of the soil core (known already) to estimate $BDfine_2$ (FAO, 2019) *(Figure 3)*.

Finally, SOC stock for each sample was determined by the following equation:

$$\textbf{SOC stock (tC/ha)} = OC_i \times BDfine_2 \times T_i \times 0.1$$

where,

SOC stock (tC/ha) is the soil organic carbon stock of the sampled depth increment;

OC_i *(mgC/g of fine earth)* is the organic carbon content of the fine earth fraction (< 2 mm) in the sampled depth increment;

$BDfine_2$ *(g fine earth per cm^3 of soil)* is the mass of fine earth per total volume of the soil sample (equivalent to the mass (g) of fine earth/total volume of soil sample (cm^3) in the given depth increment;

T is the thickness (depth, in cm) of the depth increment;

0.1 is a factor for converting mgC/cm^2 to tC/ha.

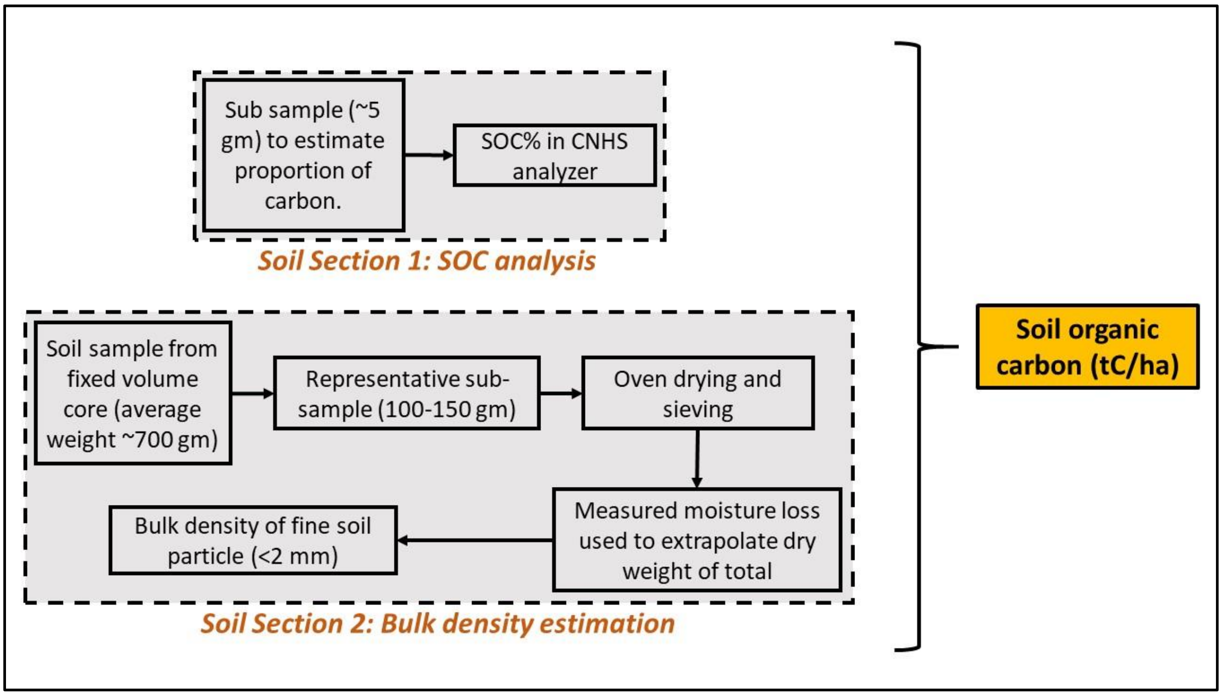

Figure 3: The steps outlining the procedure for SOC analysis for Banni's current soil carbon account.

2.8 Statistical analysis of soil carbon fluxes

To compare SOC% across different LULC classes, we conducted a mixed-effect ANOVA for the samples collected in 2023. Given the dispersion of our sampling sites across Banni, we included individual plot ID as a random effect variable and restoration type as a fixed effect variable. This analysis utilized the *'lme'* function from the R package *'nlme'*. Additionally, we performed Tukey's post-hoc analysis to evaluate differences in SOC between pairs of treatments using the *'multcomp'* package. We used *ggplot2* for visual representation of results.

For the SOC% estimates from 2014, we conducted a separate simple linear regression model with SOC% as the response variable. The area under different land cover types - (1) *Prosopis* woodland, (2) saline brushlands, (3) *Prosopis* with mixed vegetation and (4) wetlands within a 100m buffer was taken as a continuous predictor variable. No sampling point from this survey fell into the restored grassland class. Due to the correlation between areas of different land cover types, we constructed individual models for each habitat type. The best model was selected based on the R-squared value to assess the effect size of the variable.

To evaluate changes in SOC% over 10 years from 2014 to 2023, we performed a simple Student T-Test. We looked at the change in dominant LULC class within a 100m buffer of each point. Only transects sampled in both 2014 and 2023 (n = 12) were included in the analysis.

3. Results

3.1 Current land cover and SOC dynamics in Banni

In 2023, the area under saline brushlands was highest in Banni, covering an area of approx. 127,521 ha. This was followed by Prosopis with mixed vegetation (78,726 ha) and Prosopis woodlands (14,658 ha).

All LULC classes demonstrated an average SOC density of more than 100 tC/ha. The highest SOC density was found in *Vaadas* (142.72 tC/ha). This was closely followed by SOC densities in wetlands (138.59 tC/ha) and brushlands (125.75 tC/ha). *Prosopis* with mixed vegetation was found to have the lowest SOC density (105.56 tC/ha) *(Table 2)*.

Table 2: SOC densities in 5 land use-land cover classes in Banni in 2023.

Habitat	n	SOC density (Mean ± Std. Error, tC/ha)
Saline brushland	8	125.75 (± 11.61)
Vaadas	15	142.72 (± 5.72)
Prosopis with mixed vegetation	4	105.56 (± 11.01)
Dense Prosopis woodlands	8	118.91 (± 14.83)
Wetland	6	138.59 (± 13.14)

Combining these areal density estimates with the total area under the 4 LULC classes identified from spatial imageries, we determined the total SOC stock of Banni to be 27.69 MtC up to a depth of 30cms. The most SOC stock was found in saline brushlands, followed by mixed *Prosopis* stands *(Table 3)*. Given its small size in relation to the total area under grasslands globally, the total SOC stock in Banni is approx. 0.012% of the total global SOC stock in grazing lands upto 30cms (214 PgC) (Georgiou et al., 2022).

Table 3: Total SOC stocks in each land use-land cover class in Banni.

LULC class	Area under each LULC class in 2023 (ha)	SOC density (Mean ± std. error, tC/ha)	Total SOC stock (MtC)
Saline brushlands	127,521	125.75 (± 11.61)	16.03
Prosopis with mixed vegetation	78,726	105.56 (± 11.01)	8.31
Dense Prosopis woodlands	14,658	118.91 (± 14.83)	1.74
Wetlands	11,563	138.59 (± 13.14)	1.6
TOTAL	**232,468**	**119.61 (± 3.50)**	**27.69**

Highest mean SOC% was found in *Vaadas*, followed by *Pw* and *Wl*. The lowest mean SOC% was found in *Pm*. However, high variation was seen in the SOC% among these LULC types, and the differences were not found to be statistically significant *(Figure 4)*.

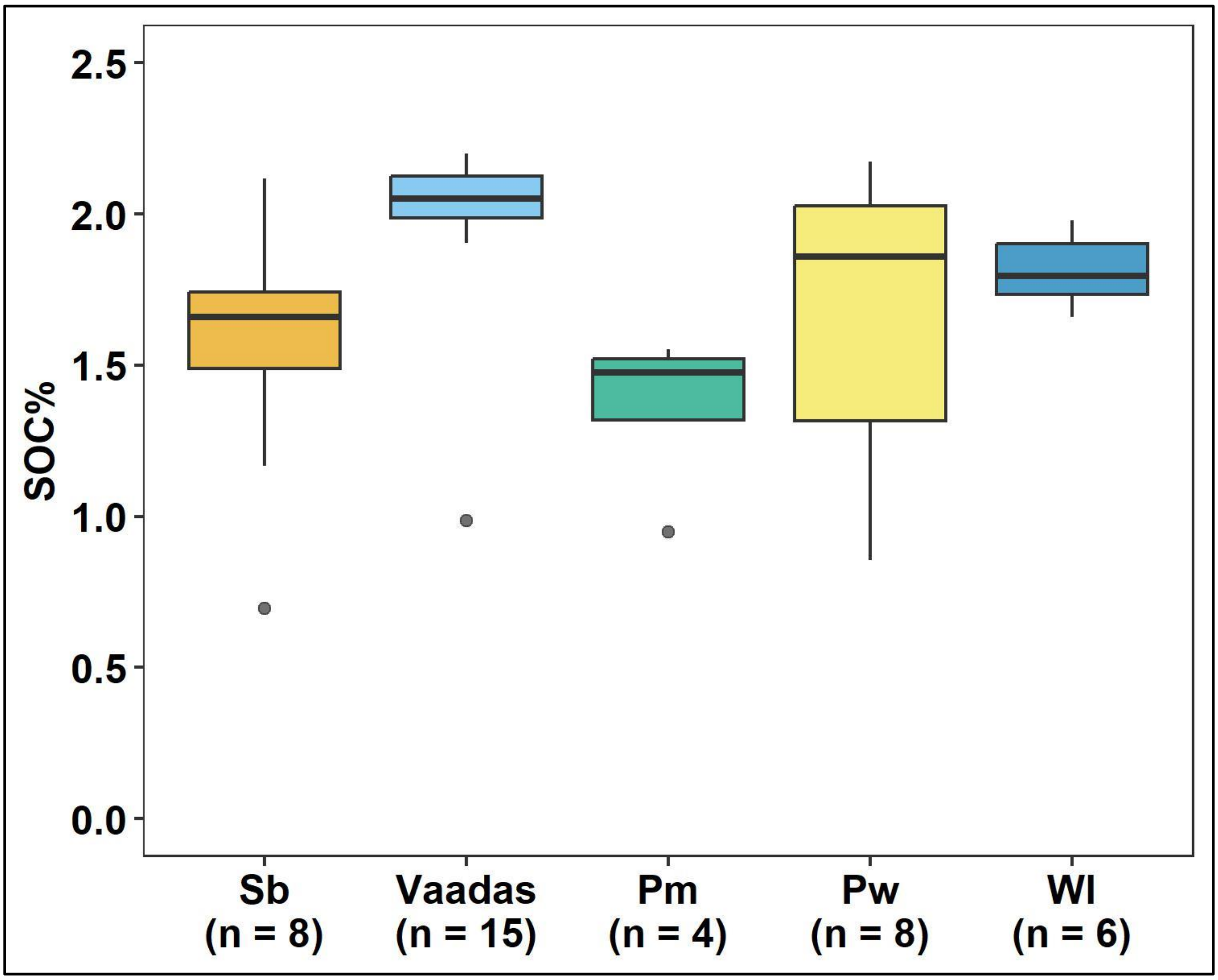

Figure 4: Distribution of SOC% across the LULC classes in 2023 obtained from field sampling. Here, Sb: saline brushlands, Vaadas: restored grasslands, Pm: Prosopis with mixed vegetation, Pw: Dense Prosopis woodlands, Wl: wetlands.

Considering SOC% as a response variable, the linear mixed-effects model demonstrated that 87.6% of the residual variation (Intercept, $\Psi = 0.33$, Residual $\sigma = 0.12$) was explained by the random intercept term (Plot ID), highlighting the impact of variation in individual sampling sites on estimated SOC% *(Table 4)*. Tukey's post-hoc pairwise comparison reveals that SOC% is significantly higher in *Vaadas* compared to both the *Sb* and *Pm* LULC classes. However, it is not significantly different compared to *Pw* and *Wl*. The SOC% in *Vaadas* was 0.43x higher compared to *Sb* soils ($\beta = 0.42$, $z = 2.77$, $p < 0.05$) and 0.63x higher compared to *Pm* soils ($\beta = -0.63$, $z = -3.17$, $p < 0.01$) *(Table 5)*.

Table 4: Linear mixed effect model describing the variation in SOC% across different LULC classes with a random intercept term (Plot ID). Associated β estimates, standard error (SE), degrees of freedom (DF), z-statistics, and p-values are shown.

Random effects:					
Formula: ~1 | ID					
	(Intercept)	Residual			
Std Dev:	0.3290787	0.1234045			
Fixed effects:					
	Estimate	SE	DF	t-value	p value
(Intercept)	1.564	0.124	36	12.586	0.000
Habitat-Grassland	0.426	0.154	36	2.769	0.009
Habitat-GrassPros	-0.200	0.215	36	-0.930	0.359
Habitat-Prosopis	0.110	0.176	36	0.628	0.534
Habitat-Wetland	0.249	0.190	36	1.314	0.197

Table 5: Results of Tukey- pairwise comparisons for variation in soil organic carbon. The estimates are differences between the means of two groups along with associated standard error (S.E.), Z-statistic, and p-value. Values in bold show statistically significant differences.

	Estimate	**SE**	**z-value**	**p-value**
Vaadas – Sb	**0.426**	**0.1539**	**2.769**	**0.0431**
Pm – Sb	-0.2001	0.2152	-0.93	0.8826
Pw - Sb	0.1103	0.1757	0.628	0.9698
Wl – Sb	0.2494	0.1898	1.314	0.6767
Pm - Vaadas	**-0.6262**	**0.1978**	**-3.166**	**0.0131**
Pw - Vaadas	-0.3157	0.1539	-2.052	0.236
Wl - Vaadas	-0.1766	0.1698	-1.04	0.8329
Pw – Pm	0.3105	0.2152	1.442	0.5937
Wl - Pm	0.4495	0.2269	1.982	0.2694
Wl - Pw	0.1391	0.1898	0.733	0.9475

This is in comparison to SOC% data from 2014, where *Sb* showed a significant negative ($\beta = -0.002$, $z = -3.27$, $p < 0.01$) effect on SOC%, while *Pm* showed a significant positive ($\beta = 0.002$, $z = 3.21$, $p < 0.01$) effect. A unit area increase in *Sb* led to a ~10% decrease in SOC%, whereas a unit area increase in *Pm* led to a ~17% increase in SOC% *(Figure S2)*.

3.2 Impacts of LULC changes on soil carbon in Banni

The area under Pw increased over the observed timeperiod (2014-2023), from 12,604 ha in 2014 to 14,658 ha in 2023. The area under Pm largely remained similar (76,568 ha in 2014, 78,726 ha in 2023), barring a sudden spike in 2020. The area under *Wl* almost doubled over the 10-year timeperiod (from 6,412 ha to 11,563 ha), although it is known locally that area under Wetlands fluctuates rapidly year-on-year, and the Monsoon rains in 2023 were recorded to be well above the regional average, while the timing of our field sampling (November 2023) meant that the rainwater may not have completely drained out. Lastly, the area under Sb decreased, from 136,884 ha in 2014 to 127,521 ha in 2023, indicating the combined effects of Wl seasonality and the further expansion of *Prosopis* in Banni *(Table S5)*.

If the SOC densities calculated for 2023 are used to develop a SOC account for 2014, we found a modest change of 8,519 tC in the total SOC account in Banni for 2014. This included an increase in the SOC under *Pw*, *Pm* and *Wl*, offset by substantial SOC losses in *Sb (Table S1)*.

Land use transition analysis with a 100m buffer around the sampling points revealed that degradation-oriented land use changes, for example, the shift from *Pm* to *Pw*, was widely observed across the 10-year period for sampling points which were sampled in 2014/17 and in 2023. In a majority of cases, LULC class of the sampling points remained the case across the years *(Table 6)*. Restoration-oriented changes, for example, a shift from Pw to Pm, were not observed. Similar results were observed for transition analysis with 60m and 80m buffers as well *(Tables S2-S4)*. In effect, there was no significant change in SOC% in the 10-year timeperiod between 2014 and 2023 ($t = -0.6455$, $p = 0.532$, $df = 11$) *(Figure S1)*.

Table 6: Changes in SOC% across sampling points which were sampled across multiple years (2014/17 and 2023) with a 100m buffer. Estimates with # refer to SOC% from 2017, the rest are from 2014. Note that SOC% for 2014/17 are from 10cm depth, where SOC% for 2023 is from 30cm depth. LTM = Long-term Monitoring plots; PROS: Dense Prosopis Woodlands; BHIND: Bhirandiyara, a village in Banni.

Sample	SOC% in 2014/2017	SOC% in 2023	SOC% Change	LULC class in 2014/17	LULC class in 2023	Transition
LTM_10	1.68	1.554	-0.126	Pm	Pros	Degradation-oriented
LTM_11	1.65	1.646	-0.004	Pm	Pm	No change
LTM_15	1.13	1.9403	0.8103	Pm	Pm	No change
LTM_18	1.34	1.1663	-0.1737	Sb	Pm	Degradation-oriented
LTM_23	1.13	1.5099	0.3799	Sb	Pm	Degradation-oriented
LTM_26	1.32	0.69497	-0.62503	Sb	Pm	Degradation-oriented
LTM_29	1.68	1.5969	-0.0831	Sb	Sb	No change
LTM_30	0.9	0.94748	0.04748	Sb	Sb	No change
LTM_36	1.29	2.1162	0.8262	Sb	Sb	No change
LTM_37	1.93	1.6769	-0.2531	Pm	Pm	No change
LTM_43	1.62	1.674	0.054	Sb	Sb	No change
LTM_44	1.5	1.4439	-0.0561	Pm	Pm	No change
BHIND-PROS-1	1.59[#]	1.3383	-0.2551	Pm	Pw	Degradation-oriented
BHIND-PROS-2	1.40[#]	1.2432	-0.1584	Pm	Pw	Degradation-oriented
GORE-PROS-1	1.82[#]	2.0225	0.1986	Pm	Pw	Degradation-oriented
GORE-PROS-2	1.85[#]	1.8061	-0.0475	Pm	Pw	Degradation-oriented

4. Discussion

4.1 Banni's current soil carbon account

The study quantifies, for the first time, the spatial heterogeneity of SOC stocks of a tropical dry grassland undergoing woody encroachment in the Indian subcontinent. Overall, our results show that the Banni grasslands, spanning an area of more than 2,300 sq.kms, is an important carbon store in India. It stores a total of 27 MtC upto a depth of 30cms across 4 major LULC types. With an average SOC density of almost 120 tC/ha, it is one of the most carbon-rich arid or semi-arid tropical grassland systems in the world today. Observed SOC densities are driven by pedoclimatic conditions in Banni which can support high SOC% and soil BD. We find that SOC% and soil BD are significantly above equivalent values for semi-arid savanna grasslands in Peninsular India (Bhan et al., Restoration Ecology, in press), which experience similar amounts of seasonal rainfall due to the Summer Monsoon. However, in Banni, the soil is predominantly clayey in nature and large low-lying tracts are seasonally inundated after the advent of the Summer Monsoon. Such high SOC densities reaffirm evidence from seasonally-flooded grasslands in South America that the combined effects of geomorphology, soil texture and the nature of plant communities facilitates high SOC concentrations (Martín-López et al., 2023).

In fact, the marked differences between SOC densities in Banni and in other arid/semi-arid open natural ecosystems in the pantropical region mean that a relevant comparison from a soil carbon accounting perspective may instead be other similar seasonal wetland-grassland complexes. With this consideration as well, observed SOC stocks for Banni compare favourably with several temperate and tropical flooded grasslands (Gomes et al., 2019; Martín-López et al., 2023), signaling Banni's importance in carbon cycling among diverse grassland ecosystems.

Table 7: Soil carbon stocks in some tropical arid/semi-arid flooded savannas. Here, # refers to total areas and SOC densities that have been calculated by the authors themselves using ecosystem area estimates of the two Brazilian grasslands taken from a separate study (Roesch et al., 2009).

Ecosystem	Area (ha)	Soil carbon stocks upto 30 cm (MtC)	SOC density (tC/ha)	Mean Annual Precipitation (mm)	Mean Annual Temperature (°C)	Source
Banni Grasslands	232,468	27.69	119.11 (± 10.25, Std. Dev.)	300	10-48 (Range)	This study
Flooded grasslands and savannas across Africa	69,500,000	2,896	41.7	-	-	(Henry et al., 2009)
Colombian Casanare flooded savannas	664,752	55.07	83.1 (± 24.3, Std. Error)	2684	21-33	(Martín-López et al., 2023)
Brazilian Pantanal	15,035,500#	770	51.21#	1000-1600	22-24	(Gomes et al., 2019)
Brazilian Pampa	17,649,600	1,490	84.42#	1300-2500	14-20	(Gomes et al., 2019)

We find that restored grasslands (*'Vaadas'*) in Banni managed by local pastoralist communities have the highest SOC densities, indicating the positive outcomes from restoration from a carbon accounting perspective. It backs up existing evidence of *Prosospis* removal as an ecologically-effective strategy to conserve native herbaceous plant diversity in Banni (Nerlekar et al., 2022). Conversely, our results indicate that planting more trees in tropical grasslands does not lead to more SOC concentration in the soils, reaffirming previous evidence from Sub-Saharan Africa (Zhou et al., 2023). For Banni, this indicates that the introduction of *Prosopis* in Banni done decades ago from a perspective of greening the landscape and arresting soil erosion, has not led to benefits in terms of higher SOC storage.

Although the management of *Prosopis* may be expensive, our results suggest that Prosopis removal may be a viable strategy to increase SOC sequestration potential in Banni. Should effective *Prosopis* removal occur, it is possible for grasslands to establish themselves and accumulate carbon within <30 years, especially if they are not overgrazed or under dense *Prosopis* cover already. It is possible that part of the carbon benefits from restoration maybe realized within <10 years already (Eschen et al., 2021). To further accumulate such carbon benefits over the medium-term, it may be beneficial to focus on *Prosopis* removal from areas which have been recently invaded, because they may have a more intact seed bank than those invaded years ago and may be easier to revert to grasslands (Eschen et al., 2021). The need for fodder to support Banni's growing livestock population may be able to provide the financial incentives required to undertake such restoration at scale (Manjunatha et al., 2022).

4.2 Challenges in attributing SOC% change to LULC change

Woody plant invasion of grassland ecosystems is a geographically extensive phenomenon that is known to alter regional biogeochemical cycles (Asner et al., 2004; Schlesinger et al., 1990). Given its widespread occurrence, there is considerable interest in quantifying its impacts on local carbon pools (Liu et al., 2011; Naikwade, 2021).

Studies have found increased SOC storage after woody invasion in some ecosystems (McKinley and Blair, 2008), but no net change, or decreased SOC storage, in others (Jackson et al., 2002). Typically, studies of this kind require the extrapolation of fine scale, local inventories of SOC storage to broad scales. Spatial heterogeneity of soil properties is amplified as woody plant abundance increases in grasslands, thus making it difficult to accurately estimate the impact of this vegetation change on SOC (Zhou et al., 2017). Change detection is further complicated by spatial variability, land use legacies, erosion, differing sampling depths across successive field campaigns and small signal-to-noise ratios. This is why such studies remain typically quite limited in their sample density and distribution, which partially accounts for the wide divergence of perspectives (increase/decrease/no change) as to how this vegetation change has affected SOC% (Liu et al., 2011).

We encounter similar challenges in Banni. In spite of significant LULC changes in Banni over the 10 years of our analysis, we do not find statistically robust impacts on SOC%. While there were differences in soil sampling depths in 2014, 2017 and 2023 (30cms in 2023, and 10cms in 2014 and 2017), these differences may not significantly impact our findings since evidence suggests that soil carbon cycling initially occurs at shallower depths, because that is where the majority of the root production occurs, before percolating deeper (Steinbeiss et al., 2008). These changes eventually recede with age as soil strata approach equilibrium (McSherry and Ritchie, 2013).

We find that degradation-oriented land use changes, like the further invasion of *Prosopis* into mixed-grassland patches due to the lack of active management predominates, but its impacts on SOC% remain without any directional trends. This effect is sustained even when buffers of varying sizes (60, 80 and 100m) are placed around sampling sites to identify the impacts of LULC change at different spatial resolutions. A possible explanation for these patterns could be that woody patches are relatively young (only about 50 years) from a SOC stabilization perspective, whereas some dynamic simulation models indicate that it may take decades, if not centuries, to reach SOC stabilization (Zhou et al., 2017). Changes in SOC% and BD will continue to drive SOC change in Banni, since we find that area changes may not be the dominant driver of SOC change in Banni: SOC changes due to LULC changes largely offset each other if SOC densities are kept constant.

Based on these observations, we predict that, without any disturbances or active interventions, woody encroachment into this landscape will continue to alter the spatial heterogeneity of SOC for the next several decades. However, given the carbon storage differences observed between *Vaadas* and other LULC classes, we believe the appropriate management strategy to maximise SOC% is to actively restore sparse *Prosopis*-bearing patches by *Prosopis* removal.

We suggest that future intervention strategies in Banni in particular, and arid/semi-arid open natural ecosystems in India in general, should be based on a more collaborative and integrated approach that takes into account the management strategies employed by local communities. Our results can feed into the National Grassland Policy currently under development at the national level in India and can be taken up by policy makers and land use managers to guide future management of degraded lands as well as *Prosopis* invasions, as a means to maximize the abundant soil carbon sequestration potential that exists in India's open natural ecosystems.

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References

Abdalla, M., Hastings, A., Chadwick, D., Jones, D., ..., 2018. Critical review of the impacts of grazing intensity on soil organic carbon storage and other soil quality indicators in extensively managed grasslands. Agric. Ecosyst. ….

Archer, S., Boutton, T., Hibbard, K., 2001. Trees in grasslands: biogeochemical consequences of woody plant expansion. Glob. Biogeochem. Cycles ….

Asner, G.P., Elmore, A.J., Olander, L.P., Martin, R.E., Harris, A.T., 2004. Grazing Systems, Ecosystem Responses and Global Change. Annu. Rev. Environ. Resour. 29, 261–299. https://doi.org/10.1146/annurev.energy.29.062403.102142

Bai, Y., Cotrufo, M.F., 2022. Grassland soil carbon sequestration: Current understanding, challenges, and solutions. Science 377, 603–608. https://doi.org/10.1126/science.abo2380

Bardgett, R., Bullock, J., Lavorel, S., ..., 2021. Combatting global grassland degradation. Nat. Rev. Earth ….

Basu, S., Sanyal, P., Pillai, A.A.S., Ambili, A., 2019. Response of grassland ecosystem to monsoonal precipitation variability during the Mid-Late Holocene: Inferences based on molecular isotopic records from Banni grassland, western India. PLOS ONE 14, e0212743. https://doi.org/10.1371/journal.pone.0212743

Bhojvaid, P.P., Timmer, V.R., 1998. Soil dynamics in an age sequence of Prosopis juliflora planted for sodic soil restoration in India. For. Ecol. Manag. 106, 181–193. https://doi.org/10.1016/S0378-1127(97)00310-1

Bossio, D.A., Cook-Patton, S.C., Ellis, P.W., Fargione, J., Sanderman, J., Smith, P., Wood, S., Zomer, R.J., Von Unger, M., Emmer, I.M., Griscom, B.W., 2020. The role of soil carbon in natural climate solutions. Nat. Sustain. 3, 391–398. https://doi.org/10.1038/s41893-020-0491-z

Buisson, E., Archibald, S., Fidelis, A., Suding, K.N., 2022. Ancient grasslands guide ambitious goals in grassland restoration. Science 377, 594–598. https://doi.org/10.1126/science.abo4605

Buisson, E., Fidelis, A., Overbeck, G.E., Schmidt, I.B., Durigan, G., Young, T.P., Alvarado, S.T., Arruda, A.J., Boisson, S., Bond, W., Coutinho, A., Kirkman, K., Oliveira, R.S., Schmitt, M.H., Siebert, F., Siebert, S.J., Thompson, D.I., Silveira, F.A.O., 2020. A research agenda for the restoration of tropical and subtropical grasslands and savannas. Restor. Ecol. rec.13292. https://doi.org/10.1111/rec.13292

Buisson, E., Le Stradic, S., Silveira, F.A.O., Durigan, G., Overbeck, G.E., Fidelis, A., Fernandes, G.W., Bond, W.J., Hermann, J.-M., Mahy, G., Alvarado, S.T., Zaloumis, N.P., Veldman, J.W., 2019. Resilience and restoration of tropical and subtropical grasslands, savannas, and grassy woodlands: Tropical grassland resilience and restoration. Biol. Rev. 94, 590–609. https://doi.org/10.1111/brv.12470

Dakhil, M.A., El-Keblawy, A., El-Sheikh, M.A., Halmy, M.W.A., Ksiksi, T., Hassan, W.A., 2021. Global Invasion Risk Assessment of Prosopis juliflora at Biome Level: Does Soil Matter? Biology 10, 203. https://doi.org/10.3390/biology10030203

Dayal, D., Dev, R., Sureshkumar, M., Manjunatha, B.L., 2018. Managing Banni grassland: Issues and Opportunities. Indian Farming 68, 112–114.

Dhaulakhandi, M., Rajwar, G.S., Kumar, P., 2000. Primary productivity and system transfer functions in alpine grassland of Western Garhwal Himalaya. Trop. Ecol. 41, 99–101.

Eggleston, H.S., Buendia, L., Miwa, K., Ngara, T., Tanabe, K., 2006. IPCC guidelines for national greenhouse gas inventories. Inst. Glob. Environ. Strateg. Hayama Jpn. 48–56.

Erb, K.-H., Luyssaert, S., Meyfroidt, P., Pongratz, J., Don, A., Kloster, S., Kuemmerle, T., Fetzel, T., Fuchs, R., Herold, M., Haberl, H., Jones, C.D., Marín-Spiotta, E., McCallum, I., Robertson, E., Seufert, V., Fritz, S., Valade, A., Wiltshire, A., Dolman, A.J., 2017. Land management: data availability and process understanding for global change studies. Glob. Change Biol. 23, 512–533. https://doi.org/10.1111/gcb.13443

Eschen, R., Bekele, K., Mbaabu, P.R., Kilawe, C.J., Eckert, S., 2021. Prosopis juliflora management and grassland restoration in Baringo County, Kenya: Opportunities for soil carbon sequestration and local livelihoods. J. Appl. Ecol. 58, 1302–1313. https://doi.org/10.1111/1365-2664.13854

FAO, 2019. Measuring and modelling soil carbon stocks and stock changes in livestock production systems: Guidelines for assessment (Version 1). Livestock Environmental Assessment and Performance (LEAP) Partnership, Rome.

Georgiou, K., Jackson, R.B., Vindušková, O., Abramoff, R.Z., Ahlström, A., Feng, W., Harden, J.W., Pellegrini, A.F.A., Polley, H.W., Soong, J.L., Riley, W.J., Torn, M.S., 2022. Global stocks and capacity of mineral-associated soil organic carbon. Nat. Commun. 13, 3797. https://doi.org/10.1038/s41467-022-31540-9

Ghosh, P.K., Mahanta, S.K., 2014. Carbon sequestration in grassland systems. Range Manag. Agrofor. 35, 173–181.

Gomes, L.C., Faria, R.M., De Souza, E., Veloso, G.V., Schaefer, C.E.G.R., Filho, E.I.F., 2019. Modelling and mapping soil organic carbon stocks in Brazil. Geoderma 340, 337–350. https://doi.org/10.1016/j.geoderma.2019.01.007

Guerra, C.A., Heintz-Buschart, A., Sikorski, J., Chatzinotas, A., Guerrero-Ramírez, N., Cesarz, S., Beaumelle, L., Rillig, M.C., Maestre, F.T., Delgado-Baquerizo, M., Buscot, F., Overmann, J., Patoine, G., Phillips, H.R.P., Winter, M., Wubet, T., Küsel, K., Bardgett, R.D., Cameron, E.K., Cowan, D., Grebenc, T., Marín, C., Orgiazzi, A., Singh, B.K., Wall, D.H., Eisenhauer, N., 2020. Blind spots in global soil biodiversity and ecosystem function research. Nat. Commun. 11, 3870. https://doi.org/10.1038/s41467-020-17688-2

Henry, M., Valentini, R., Bernoux, M., 2009. Soil carbon stocks in ecoregions of Africa. Biogeosciences Discuss. 6, 797–823. https://doi.org/10.5194/bgd-6-797-2009

Jackson, R.B., Banner, J.L., Jobbágy, E.G., Pockman, W.T., Wall, D.H., 2002. Ecosystem carbon loss with woody plant invasion of grasslands. Nature 418, 623–626. https://doi.org/10.1038/nature00910

Jobbágy, E.G., Jackson, R.B., 2000. The Vertical Distribution of Soil Organic Carbon and Its Relation to Climate and Vegetation. Ecol. Appl. 10, 423–436. https://doi.org/10.1890/1051-0761(2000)010[0423:TVDOSO]2.0.CO;2

Joshi, P.N., Kumar, V., Koladiya, M., Patel, Y.S., Karthik, T., 2009. Local perceptions of grassland change and priorities for conservation of natural resources of Banni, Gujarat, India. Front. Biol. China 4, 549–556. https://doi.org/10.1007/s11515-009-0041-6

Joshi, U., Kiran, G.S., 2021. Evaluating the Impact of Different Vegetation Types on NEE: A Case Study of Banni Grasslands, India. J. Environ. Prot. 12, 490–507. https://doi.org/10.4236/jep.2021.127031

Lal, R., 2004. Soil Carbon Sequestration Impacts on Global Climate Change and Food Security. Science 304, 1623–1627. https://doi.org/10.1126/science.1097396

Lal, R., Negassa, W., Lorenz, K., 2015. Carbon sequestration in soil. Curr. Opin. Environ. Sustain. 15, 79–86. https://doi.org/10.1016/j.cosust.2015.09.002

Liu, F., Wu, X.B., Bai, E., Boutton, T.W., Archer, S.R., 2011. Quantifying soil organic carbon in complex landscapes: an example of grassland undergoing encroachment of woody plants. Glob. Change Biol. 17, 1119–1129. https://doi.org/10.1111/j.1365-2486.2010.02251.x

Malhi, Y., Girardin, C., Metcalfe, D.B., Doughty, C.E., Aragão, L.E.O.C., Rifai, S.W., Oliveras, I., Shenkin, A., Aguirre-Gutiérrez, J., Dahlsjö, C.A.L., Riutta, T., Berenguer, E., Moore, S., Huasco, W.H., Salinas, N., da Costa, A.C.L., Bentley, L.P., Adu-Bredu, S., Marthews, T.R., Meir, P., Phillips, O.L., 2021. The Global Ecosystems Monitoring network: Monitoring ecosystem productivity and carbon cycling across the tropics. Biol. Conserv. 253, 108889. https://doi.org/10.1016/j.biocon.2020.108889

Manjunatha, B.L., Naorem, A., Hajong, D., Tewari, P., 2022. Dynamics of Household and Regional Economy in Banni Grasslands, India: A Cross-Sectional Study. Sustainability 14, 11236. https://doi.org/10.3390/su141811236

Martín-López, J.M., Verchot, L.V., Martius, C., Da Silva, M., 2023. Modeling the Spatial Distribution of Soil Organic Carbon and Carbon Stocks in the Casanare Flooded Savannas of the Colombian Llanos. Wetlands 43, 65. https://doi.org/10.1007/s13157-023-01705-3

Mbaabu, P., Olago, D., Gichaba, M., Eckert, S., Eschen, R., ..., 2020. Restoration of degraded grasslands, but not invasion by Prosopis juliflora, avoids trade-offs between climate change mitigation and other ecosystem services. Sci. Rep. https://doi.org/10.1038/s41598-020-77126-7

McKinley, D., Blair, J., 2008. Woody Plant Encroachment by Juniperus virginiana in a Mesic Native Grassland Promotes Rapid Carbon and Nitrogen Accrual. Ecosystems. https://doi.org/10.1007/s10021-008-9133-4

McSherry, M.E., Ritchie, M.E., 2013. Effects of grazing on grassland soil carbon: a global review. Glob. Change Biol. 19, 1347–1357. https://doi.org/10.1111/gcb.12144

Naikwade, P., 2021. Changes in Soil Carbon Sequestration during Woody Plant Encroachment in Arid Ecosystems. Plantae Sci. 4, 266–276. https://doi.org/10.32439/ps.v4i4-5.266-276

Nerlekar, A.N., Mehta, N., Pokar, R., Bhagwat, M., Misher, C., Joshi, P., Hiremath, A.J., 2022. Removal or utilization? Testing alternative approaches to the management of an invasive woody legume in an arid Indian grassland. Restor. Ecol. 30, e13477. https://doi.org/10.1111/rec.13477

Phillips, O.L., Malhi, Y., Higuchi, N., Laurance, W.F., Núñez, P.V., Vásquez, R.M., Laurance, S.G., Ferreira, L.V., Stern, M., Brown, S., Grace, J., 1998. Changes in the Carbon Balance of Tropical Forests: Evidence from Long-Term Plots. Science 282, 439–442. https://doi.org/10.1126/science.282.5388.439

Poorter, L., Bongers, F., Aide, T.M., Almeyda Zambrano, A.M., Balvanera, P., Becknell, J.M., Boukili, V., Brancalion, P.H.S., Broadbent, E.N., Chazdon, R.L., Craven, D., de Almeida-Cortez, J.S., Cabral, G.A.L., de Jong, B.H.J., Denslow, J.S., Dent, D.H., DeWalt, S.J., Dupuy, J.M., Durán, S.M., Espírito-Santo, M.M., Fandino, M.C., César, R.G., Hall, J.S., Hernandez-Stefanoni, J.L., Jakovac, C.C., Junqueira, A.B., Kennard, D., Letcher, S.G., Licona, J.-C., Lohbeck, M., Marín-Spiotta, E., Martínez-Ramos, M., Massoca, P., Meave, J.A., Mesquita, R., Mora, F., Muñoz, R., Muscarella, R., Nunes, Y.R.F., Ochoa-Gaona, S., de Oliveira, A.A., Orihuela-Belmonte, E., Peña-Claros, M., Pérez-García, E.A., Piotto, D., Powers, J.S., Rodríguez-Velázquez, J., Romero-Pérez, I.E., Ruíz, J., Saldarriaga, J.G., Sanchez-Azofeifa, A., Schwartz, N.B., Steininger, M.K., Swenson, N.G., Toledo, M., Uriarte, M., van Breugel, M., van der Wal, H., Veloso, M.D.M., Vester, H.F.M., Vicentini, A., Vieira, I.C.G., Bentos, T.V., Williamson, G.B., Rozendaal, D.M.A., 2016. Biomass resilience of Neotropical secondary forests. Nature 530, 211–214. https://doi.org/10.1038/nature16512

Poorter, L., Dylan, C., C., J.C., T., van der S.M., Lucy, A., Bongers Frans, Chazdon Robin L., Farrior Caroline E., Kambach Stephan, Meave Jorge A., Muñoz Rodrigo, Norden Natalia, Rüger Nadja, van Breugel Michiel, Almeyda Zambrano Angélica María, Amani Bienvenu, Andrade José Luis, Brancalion Pedro H. S., Broadbent Eben N., de Foresta Hubert, Dent Daisy H., Derroire Géraldine, DeWalt Saara J., Dupuy Juan M., Durán Sandra M., Fantini Alfredo C., Finegan Bryan, Hernández-Jaramillo Alma, Hernández-Stefanoni José Luis, Hietz Peter, Junqueira André B., N'dja Justin Kassi, Letcher Susan G., Lohbeck Madelon, López-Camacho René, Martínez-Ramos Miguel, Melo Felipe P. L., Mora Francisco, Müller Sandra C., N'Guessan Anny E., Oberleitner Florian, Ortiz-Malavassi Edgar, Pérez-García Eduardo A., Pinho Bruno X., Piotto Daniel, Powers Jennifer S., Rodríguez-Buriticá Susana, Rozendaal Danaë M. A., Ruíz Jorge, Tabarelli Marcelo, Teixeira Heitor Mancini, Valadares de Sá Barretto Sampaio Everardo, van der Wal Hans, Villa Pedro M., Fernandes Geraldo W., Santos Braulio A., Aguilar-Cano José, de Almeida-Cortez Jarcilene S., Alvarez-Davila Esteban, Arreola-Villa Felipe, Balvanera Patricia, Becknell Justin M., Cabral George A. L., Castellanos-Castro Carolina, de Jong Ben H. J., Nieto Jhon Edison, Espírito-Santo Mário M., Fandino Maria C., García Hernando, García-Villalobos Daniel, Hall Jefferson S., Idárraga Alvaro, Jiménez-Montoya Jaider, Kennard Deborah, Marín-Spiotta Erika, Mesquita Rita, Nunes Yule R. F., Ochoa-Gaona Susana, Peña-Claros Marielos, Pérez-Cárdenas Nathalia, Rodríguez-Velázquez Jorge, Villanueva Lucía Sanaphre, Schwartz Naomi B., Steininger Marc K., Veloso Maria D. M., Vester Henricus F. M., Vieira Ima C. G., Williamson G. Bruce, Zanini Kátia, Hérault Bruno, 2021. Multidimensional tropical forest recovery. Science 374, 1370–1376. https://doi.org/10.1126/science.abh3629

Roesch, L.F., Vieira, F., Pereira, V., Schünemann, A.L., Teixeira, I., Senna, A.J., Stefenon, V.M., 2009. The Brazilian Pampa: A Fragile Biome. Diversity 1, 182–198. https://doi.org/10.3390/d1020182

Schlesinger, W.H., Reynolds, J.F., Cunningham, G.L., Huenneke, L.F., Jarrell, W.M., Virginia, R.A., Whitford, W.G., 1990. Biological Feedbacks in Global Desertification. Science 247, 1043–1048. https://doi.org/10.1126/science.247.4946.1043

Steinbeiss, S., BEßLER, H., Engels, C., Temperton, V.M., Buchmann, N., Roscher, C., Kreutziger, Y., Baade, J., Habekost, M., Gleixner, G., 2008. Plant diversity positively affects short-term soil carbon storage in experimental grasslands: BIODIVERSITY INCREASES CARBON STORAGE. Glob. Change Biol. 14, 2937–2949. https://doi.org/10.1111/j.1365-2486.2008.01697.x

Stockmann, U., Adams, M.A., Crawford, J.W., Field, D.J., Henakaarchchi, N., Jenkins, M., Minasny, B., McBratney, A.B., Courcelles, V. de R. de, Singh, K., Wheeler, I., Abbott, L., Angers, D.A., Baldock, J., Bird, M., Brookes, P.C., Chenu, C., Jastrow, J.D., Lal, R., Lehmann, J., O'Donnell, A.G., Parton, W.J., Whitehead, D., Zimmermann, M., 2013. The knowns, known unknowns and unknowns of sequestration of soil organic carbon. Agric. Ecosyst. Environ. 164, 80–99. https://doi.org/10.1016/j.agee.2012.10.001

Thokchom, A., Yadava, P., 2016. Carbon dynamics in an Imperata grassland in Northeast India. Trop. Grassl.-Forrajes ….

Tian, D., Xiang, Y., Seabloom, E., Wang, J., Jia, X., Li, T., Li, Z., Yang, J., Guo, H., Niu, S., 2023. Soil carbon sequestration benefits of active versus natural restoration vary with initial carbon content and soil layer. Commun. Earth Environ. 4, 83. https://doi.org/10.1038/s43247-023-00737-1

Zhou, Y., Bomfim, B., Bond, W.J., Boutton, T.W., Case, M.F., Coetsee, C., Davies, A.B., February, E.C., Gray, E.F., Silva, L.C.R., Wright, J.L., Staver, A.C., 2023. Soil carbon in tropical savannas mostly derived from grasses. Nat. Geosci. 16, 710–716. https://doi.org/10.1038/s41561-023-01232-0

Zhou, Y., Boutton, T.W., Wu, X.B., 2017. Soil carbon response to woody plant encroachment: importance of spatial heterogeneity and deep soil storage. J. Ecol. 105, 1738–1749. https://doi.org/10.1111/1365-2745.12770

Land management controls on soil carbon fluxes in Asia's largest tropical grassland

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This Supplementary Information file contains:

Text S1

Tables S1-S5

Figures S1-S2

Text S1: Sampling protocol for estimating SOC concentrations in 2014 and 2017

Sampling Protocol for 2014

In 2014, the entire Banni landscape was divided into 4 x 4kms grids for vegetation sampling. Permanent transects were then laid inside these grids. These transects were located at the centre of each grid cell in a systematic sampling design. The protocol for laying these transects is described below.

A transect of 100m length was laid out in due North-South direction, such that the centre of the 4 x 4 kms grid falls in the center of the transect. Each transect was 100 m long and 4 m wide (i.e., with a 2 m wide band on either side of the central line).

In cases where an obstacle (a road, a building, a permanent water body) was located within this transect, the transect was displaced in a due North direction (or South direction if displacement in North was not possible) for not more than 100 m, without changing the N-S orientation. Notes on the distance displaced and the new coordinates of the transect was then recorded. Transects that fell within a permanent water body were omitted. In cases where vertical displacement was not possible, horizontal displacement of the transect was done. In such cases, the transect was first shifted due east. If east did not work, then it was shifted west. Finally, the end points and mid-point of each transect was marked with 120 cm long concrete stakes buried in the ground such that the top 90 cm was visible above ground.

The soil sampling for each transect was then performed. A total of six samples was initially taken for each transect. Samples were collected at the extreme corners of the 4x100 m belt transect, and at the midpoints along the length of the transect (ie. at 50m along the transect length). Black circles indicate locations of soil samples relative to belt transect. Soil was collected up to a depth of 10 cm at every sampling transect. A well-mixed composite of the six soil samples per transect was made, from which an approx. 500gm sample was taken for further analysis of SOC concentrations (SOC%).

Sampling Protocol for 2017

The study was designed to compare the effectiveness of two different methods for managing *Prosopis juliflora*, which are a concern in grassland restoration and recovery efforts (Nerlekar et al., 2022). The methods for managing Prosopis was (1) mechanical removal, for example, using an earthmover, where trees are completely removed above ground, but roots partially remain in the ground, and (2) lopping, where trees are cut down close to the ground (about 5–10 cm high) using machetes. This technique is commonly used by locals to harvest the trees for charcoal production. After lopping, the trees are allowed to regrow for about three years before being cut again. In addition to these two methods of management, there was a control group where no *Prosopis* management was applied.

The soil sampling for each of these three cases was then performed. The experiment was carried out in eight villages across the Banni region, which provided a diverse range of conditions due to the east–west rainfall gradient. The selection of villages was based on the willingness of village elders to participate in the study. The study used a randomized block design. This means that the different treatments (mechanical removal, lopping, and control) were assigned randomly to plots within each block. The size of each of these plots was 20x20m. We used three random soil samples collected at a depth of 10 cm from these plots.

Table S1: SOC stocks in different land use-land cover classes for 2014 and 2023, assuming the same SOC density for both years.

LULC Class	2014			2023			SOC change
	Area in 2014 (ha)	SOC density in 2014 (ha)	Total SOC (tC)	Area in 2014 (ha)	SOC density in 2014 (ha)	Total SOC (tC)	
Dense Prosopis woodlands	12,604	118.91	14,98,741.64	14,658	118.91	17,42,982.78	2,44,241.14
Prosopis with mixed vegetation	76,568	105.56	80,82,518.08	78,726	105.56	83,10,316.56	2,27,798.48
Saline brushlands	1,36,884	125.75	1,72,13,163.00	1,27,521	125.75	1,60,35,765.75	-11,77,397.25
Wetlands	6,412	138.59	8,88,639.08	11,563	138.59	16,02,516.17	7,13,877.09
	2,32,468		2,76,83,061.80	2,32,468		2,76,91,581.26	8,519.46

Table S2: Changes in SOC% across sampling points which were sampled across multiple years (2014/17 and 2023) with a 60m buffer. Estimates with # refer to SOC% from 2017. Note that SOC% for 2014/17 are from 10cm depth, where SOC% for 2023 is from 30cm depth. LTM = Long-term Monitoring; PROS: Dense Prosopis Woodlands; BHIND: Bhirandiyara, a village in Banni.

Transect	**SOC% in 2014/2017**	**SOC% in 2023**	**SOC% change**	**LULC class in 2014**	**LULC class in 2017**	**LULC class in 2020**	**LULC class in 2023**	**Transition**
LTM_10	1.68	1.554	-0.126	Pm	Pm	Pros	Pm	Degradation-oriented
LTM_11	1.65	1.646	-0.004	Pm	Pm	Pm	Pm	No change
LTM_15	1.13	1.9403	0.8103	Pm	Sb	Pm	Pm	Degradation-oriented
LTM_18	1.34	1.1663	-0.1737	Sb	Sb	Pm	Pm	Degradation-oriented
LTM_23	1.13	1.5099	0.3799	Sb	Sb	Pm	Pm	Degradation-oriented
LTM_26	1.32	0.69497	-0.62503	Sb	Sb	Pm	Pm	Degradation-oriented
LTM_29	1.68	1.5969	-0.0831	Sb	Sb	Sb	Pm	Restoration-oriented
LTM_30	0.9	0.94748	0.04748	Sb	Sb	Sb	Sb	No change
LTM_36	1.29	2.1162	0.8262	Sb	Mix	Sb	Pm	Degradation-oriented
LTM_37	1.93	1.6769	-0.2531	Pm	Pm	Pm	Pm	No change
LTM_43	1.62	1.674	0.054	Sb	Sb	Sb	Sb	No change
LTM_44	1.5	1.4439	-0.0561	Pm	Pm	Pm	Pm	No change
BHIND-PROS-1	1.59#	1.3383	-0.2551		Pw	Pw	Pw	No change
BHIND-PROS-2	1.40#	1.2432	-0.1584		Pw	Pw	Pw	No change
GORE-PROS-1	1.82#	2.0225	0.1986		Pw	Pw	Pw	No change
GORE-PROS-2	1.85#	1.8061	-0.0475		Pw	Pw	Pw	No change

Table S3: Changes in SOC% across sampling points which were sampled across multiple years (2014/17 and 2023) with a 80m buffer. Estimates with # refer to SOC% from 2017. Note that SOC% for 2014/17 are from 10cm depth, where SOC% for 2023 is from 30cm depth. LTM = Long-term Monitoring; PROS: Dense Prosopis Woodlands; BHIND: Bhirandiyara, a village in Banni.

Transect	**SOC% in 2014/2017**	**SOC% in 2023**	**SOC% change**	**LULC class in 2014**	**LULC class in 2017**	**LULC class in 2020**	**LULC class in 2023**	**Transition**
LTM_10	1.68	1.554	-0.126	Sb	Pm	Pm	Pros	Degradation-oriented
LTM_11	1.65	1.646	-0.004	Sb	Pm	Pm	Pm	Degradation-oriented
LTM_15	1.13	1.9403	0.8103	Sb	Pm	Pm	Pm	Degradation-oriented
LTM_18	1.34	1.1663	-0.1737	Mix	Sb	Pm	Pm	Degradation-oriented
LTM_23	1.13	1.5099	0.3799	Mix	Sb	Pm	Pm	Degradation-oriented
LTM_26	1.32	0.69497	-0.62503	Sb	Sb	Pm	Pm	Degradation-oriented
LTM_29	1.68	1.5969	-0.0831	Mix	Sb	Pm	Sb	Restoration-oriented
LTM_30	0.9	0.94748	0.04748	Sb	Sb	Sb	Sb	No change
LTM_36	1.29	2.1162	0.8262	Pm	Pm	Pm	Sb	Degradation-oriented
LTM_37	1.93	1.6769	-0.2531	Sb	Pm	Pm	Pm	Degradation-oriented
LTM_43	1.62	1.674	0.054	Sb	Sb	Sb	Sb	No change
LTM_44	1.5	1.4439	-0.0561	Sb	Pm	Pm	Pm	Degradation-oriented
BHIND-PROS-1	1.33#	1.5934	0.2551		Pw	Pw	Pw	No change
BHIND-PROS-2	1.24#	1.4016	0.1584		Pw	Pw	Pw	No change

GORE-PROS-1	2.02#	1.8239	-0.1986		Pw	Pw	Pw	No change
GORE-PROS-2	1.80#	1.8536	0.0475		Pw	Pw	Pw	No change

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Table S4: Changes in SOC% across sampling points which were sampled across multiple years (2014/17 and 2023) with a 100m buffer. Estimates with # refer to SOC% from 2017. Note that SOC% for 2014/17 are from 10cm depth, where SOC% for 2023 is from 30cm depth. LTM = Long-term Monitoring; PROS: Dense Prosopis Woodlands; BHIND: Bhirandiyara, a village in Banni.

Transect	SOC% in 2014/2017	SOC% in 2023	SOC% change	LULC class in 2014	LULC class in 2017	LULC class in 2020	LULC class in 2023	Transition
LTM_10	1.68	1.554	-0.126	Pm	Pm	Pm	Pw	Degrdation-oriented
LTM_11	1.65	1.646	-0.004	Pm	Pm	Pm	Pm	No change
LTM_15	1.13	1.9403	0.8103	Pm	Pm	Pm	Pm	No change
LTM_18	1.34	1.1663	-0.1737	Sb	Sb	Pm	Pm	Degrdation-oriented
LTM_23	1.13	1.5099	0.3799	Sb	Sb	Pm	Pm	Degrdation-oriented
LTM_26	1.32	0.69497	-0.62503	Sb	Sb	Pm	Pm	Degrdation-oriented
LTM_29	1.68	1.5969	-0.0831	Sb	Sb	Pm	Sb	Restoration-oriented
LTM_30	0.9	0.94748	0.04748	Sb	Sb	Sb	Sb	No change
LTM_36	1.29	2.1162	0.8262	Sb	Pm	Pm	Sb	Degrdation-oriented
LTM_37	1.93	1.6769	-0.2531	Pm	Pm	Pm	Pm	No change
LTM_43	1.62	1.674	0.054	Sb	Sb	Sb	Sb	No change
LTM_44	1.5	1.4439	-0.0561	Pm	Pm	Pm	Pm	No change
BHIND-PROS-1	1.59[#]	1.3383	-0.2551		Pm	Pw	Pw	Degrdation-oriented
BHIND-PROS-2	1.40[#]	1.2432	-0.1584		Pm	Pw	Pw	Degrdation-oriented
GORE-PROS-1	1.82[#]	2.0225	0.1986		Pm	Pm	Pw	Degrdation-oriented

GORE-PROS-2	1.85[#]	1.8061	-0.0475		Pm	Pm	Pw	Degrdation-oriented

Table S5: Area under each land use-land cover class in Banni over 2014, 2017, 2020 and 2023.

LULC Class	Area in 2023 (ha)	Area in 2020 (ha)	Area in 2017 (ha)	Area in 2014 (ha)
Dense *Prosopis* woodland	14,658	138.63	10,724	12,604
Prosopis with mixed vegetation	78,726	1077.38	78,908	76,568
Wetland	11,563	82.65	1,601	6,412
Saline brushland	127,521	1026.03	141,235	136,884

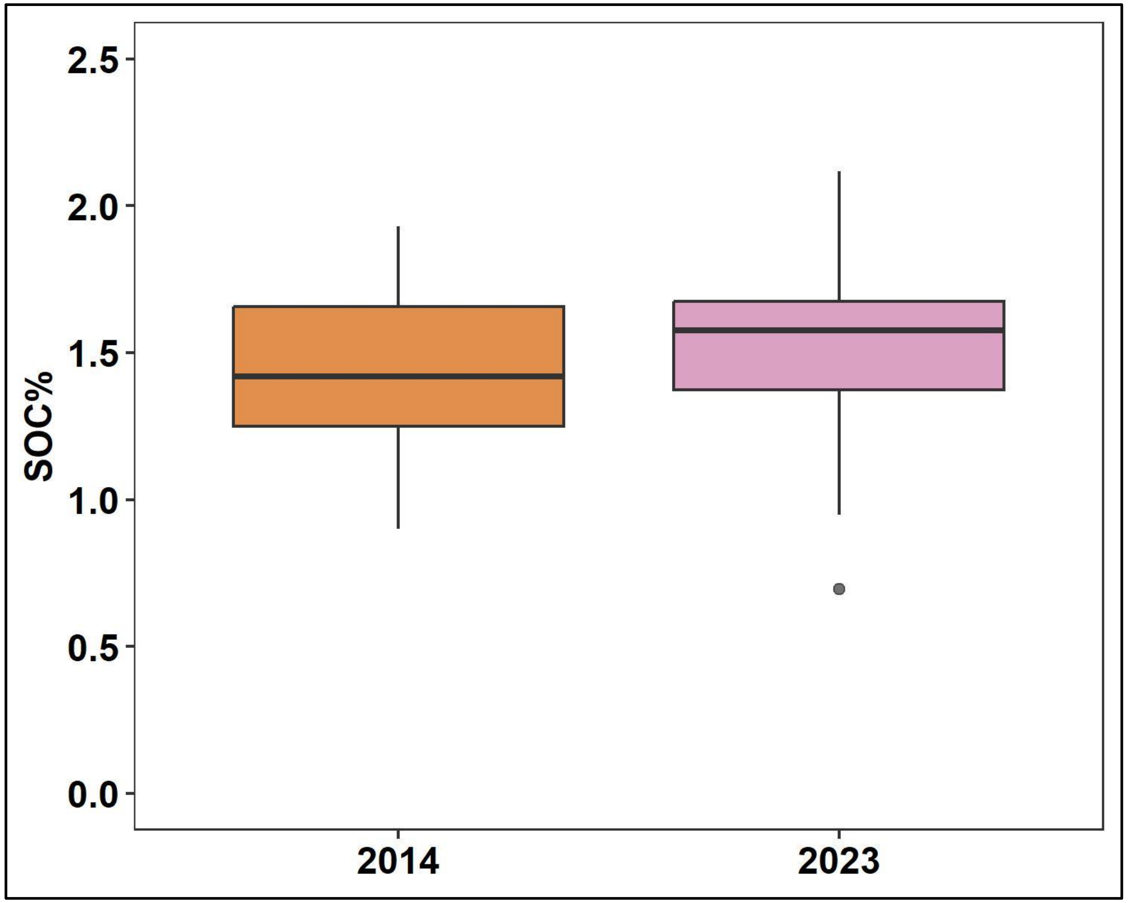

Figure S1: Distribution of SOC% for 2014 and 2023 in Banni. Note that SOC% for 2014 have been estimated at 10cm soil depth, whereas for 2023 at 30cm soil depth (n = 12).

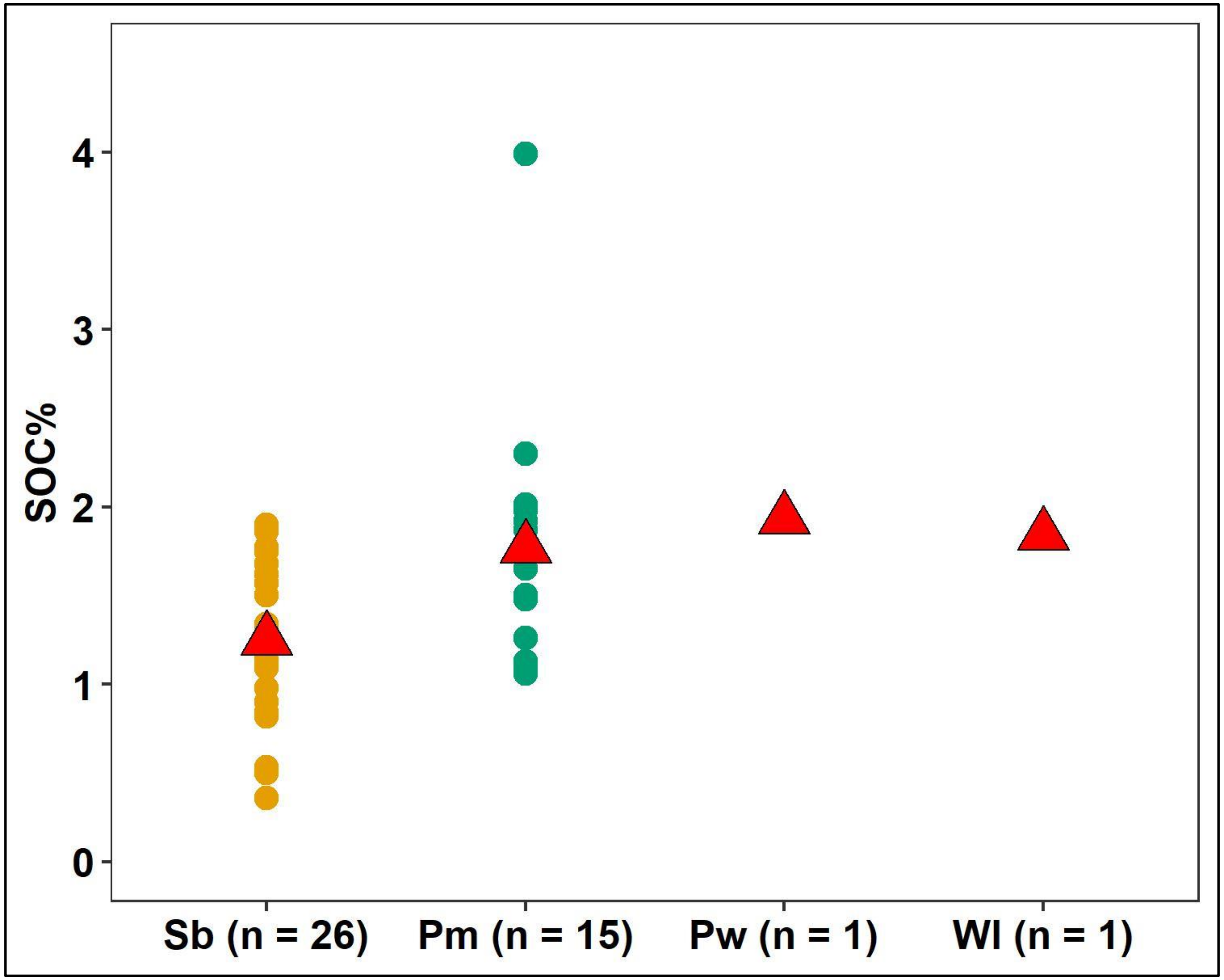

Figure S2: SOC% in sampling points in 2014. The red triangle indicates mean of all observations in that LULC class. In Pw and Wl – there was only 1 sampling point each. Here, Sb: saline brushlands, Pm: Prosopis with mixed vegetation, Pw: Dense Prosopis woodlands, Wl: wetlands.