Variation of Carbon Sequestration in *Halocnemum strobilaceum* and Soil under Livestock Grazing (Case Study: Salt Lands of Golestan Province, Iran)

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Received: 8 May 2017 / Accepted: 23 June 2017 / Published Online: 23 September 2017

**Background:** Grazing intensity has a major effect on soil and biomass organic carbon storage (C). Various plants and ecosystems with different characteristics have their unique responses to the grazing. Recognizing the effects of grazing intensity on C in various environments dominated by *Halocnemum strobilaceum*, such as Incheboron salt land of Golestan province, helps planning the grazing strategies.

**Materials and Methods:** Three grazing sites, viz. heavy grazing (in vicinity of resting point), moderate grazing (at a distance of 650 meters from resting point) and light grazing (at a distance of 2050 meters from resting point) were selected. Sampling of soil and *H. strobilaceum* species was carried out with systematic-random method in Nov. 2012. In this regard, fifteen plants along the five 100 meters transects (with distance of 30m) were randomly selected and underground and aboveground organs were extracted. The amount of biomass C was determined by combustion method. Also, soil sampling with five repetition was carried out in two depths (0-20 cm and 20-40 cm) and the amount of stored C was determined by Walkly and Black method.

**Results:** The sum of underground and aboveground biomass C in light grazing site was more than the heavy and moderate grazing sites, which were about 1.17, 1.07 and 0.567 ton/hectare respectively. The amount of soil C for the mentioned sites were 162.56, 137.39 and 80.76 ton/hectare, respectively. Besides, the depth 0-20 cm in all sites had a higher C. The soil C comprised more than 99 percent of ecosystem total stored C (biomass and soil C) in each site. In terms of total ecosystem C, the heavy and moderate grazing site had about 84.37 and 32.20 ton/hectare less C compared to light grazing site.

**Conclusions:** From a management perspective, it is concluded that light grazing intensity in saltland region can lead to maintenance of C in high level and grazing systems should avoid high stocking rates because it may adversely affect soil C. Heavy grazing has more negative effects on C compared to moderate grazing. Both soil and biomass respond similarly to different grazing intensities. The soil, especially first depth as the main resource of C, should be protected from deterioration to prevent C decline. In viewpoint of C sequestration, it is suggested to plan grazing intensity in light level and protect the soil of rangelands.

**Keywords:** Biomass, Carbon sequestration, Grazing intensive, *Halocnemum strobilaceum*, Soil
1. Background
Since 1980, the atmospheric concentration of CO₂ and other greenhouse gases, such as CO, CH₄ and N₂O, have been taken into attention mainly due to accelerated consumption of fossil fuels (1). These gases have devastating effects on the human life through increasing global warming and its consequences, such as damages to the ecosystems, flooding, drought and extreme climatic events (2). Warnings about increasing CO₂ in the atmosphere have stimulated investigations that may lead to ways of reducing its emissions through C sequestration processes (3), of which photosynthesis is considered as one of the options to reduce greenhouse gases in soil or plant bodies (4).

Rangeland ecosystems in arid and semi-arid regions have high potential to sequester C because they constitute almost half of terrestrial ecosystems that involve 10% of organic C storage (5). The vast expanse of salt rangeland makes it highly potential in the C sequestration and its management practices can, therefore, directly affect it. These ecosystems include important source of C cycle and are affected by any weather alteration, management methods and environmental conditions (6). Therefore, understanding the effects of management on changing C storage in soils and biomass can lead to better decisions in management of rangeland ecosystems. On the other hand, the sensitivity of rangelands ecosystem has increased the attention on proper land management to preserve the C sequestration potential. Livestock grazing is counted as one of the most common types of land utilization in the some parts of the world which has both direct and indirect effects in the soil and vegetation. Grazing can quantitatively and qualitatively alter C storage in rangeland ecosystems by allocation of C to the above-ground and underground biomass (7) as well as influencing species diversity and plant communities composition (8). Today, there are many uncertainties about whether increase in livestock grazing decreases the C storage, as there are conflicting reports in this regard. In global studies, both positive and negative effects have been related to the variation of climate, environment, soil properties, vegetation types, management strategies, etc. (9).

Understanding the effects of rangelands grazing on the C storage in Iran is very important, because almost 53 percent of the country is covered by rangelands and grazing is one of the most important utilization systems of land use (10). Moreover, due to the variety of ecological and historical conditions, there are many conflicts in this context (11). For instance, Menezes et al. found no significant differences between the grazed and un-grazed area on pH, C and nitrogen (12). Attaeian (13) extracted the data of Iran’s rangelands from 2006 to 2013 and found that mean CO₂ fixation was about 0.25 Mg C ha⁻¹y⁻¹. Considering the total rangeland area of Iran (≈ 84.8 million hectare), approximately 11770.011 Gg C y⁻¹ was stored in above-ground biomass annually, providing at least 5885 Gg organic C sequestration potential. Motamedi et al. (14) showed that there was a significant difference between the root and above-ground biomass of *Artemisia fragrans* under three grazing intensities. They pointed out that highest and lowest aerial biomass in two low and high grazing intensities were 5.18 and 76.42 g, respectively. Moreover, the maximum average of underground biomass per plant (95.32 g) and its minimum amount (46.56 g) belonged to low and high grazing intensities, respectively. Other researches in *Stipa grandis* and *Stipa bungeana* steppe in northern China (15) and also in a national park (7) found no changes in the soil organic materials. However it seemed that this process caused to accelerate the process of C cycle in grazed ecosystems, but in many cases the effects of grazing on C storage ecosystem were irregular and highly variable and difficult to predict the effects. Tavakoli (16) investigated the potential of C sequestration of *Hammada salicornica* vegetation type in desert...
areas of South Khorasan and found that C sequestered between 133 to 3293 kg/ha in different sites in which had the highest and the lowest amount of organic C in plant vegetation parts; soil organic C was about 6313 kg/ha on average in that site. The effects of grazing management on the ecosystem processes that control C cycling and distribution have not been sufficiently evaluated in native rangelands ecosystems.

2. Objective
C sequestration under the grazing intensity in the Incheboron salt lands, Golestan Province, has been investigated. Therefore, the manner of grazing effects on the C storage in salt lands can be determined and the C sequestration in ecosystems can be increased by using their findings towards the planning and management of rangeland. Increase in the knowledge about the effect of livestock grazing on the rate of C sequestration in ecosystems can provide the practical predictive models in grazing management. As low-grazing site is expected to be less affected by grazing disturbances, it is hypothesized that the C storage in both soil and biomass in this site is higher than the other higher grazing intensities.

3. Materials and Methods
3.1. Study area
Covering an area about 22200 hectares in a semi-hot and arid zone (37°07’N and 54°29’E), the Incheboron winter rangeland of Golestan province was selected as the representative rangeland with highly salty and alkaline soil (Figure 1). The long-term mean annual rainfall of this region is 181.5 mm (1986 to 2016) and the mean temperature is 17.06 °C; the soil texture is silt-loam to silt-clay-loam. The region is flat and slope is lower than five percent. The elevation above sea level is about -10 m. The Dalagh sheep race is the dominant livestock in this region. Shrubs constituted the major parts of vegetation with Halocnemum strobilaceum being the dominant species having class II palatability with 80 percent frequencies in plant community. Halostachys caspica, Artemisa siberi, Aeluropus lagopoides and A. littoralis are the other species of this region (17), (18).

Figure 1 study area in Golestan province, Iran
Sampling was carried out on three sites of low, moderate and high grazing intensity. Since the regions near the rest point and troughs were under high grazing intensity and trampling, they were considered as the high grazing intensity (10). The other two other sites (low and moderate) were selected within the distance of 650 and 2050 meters from the center of high grazing intensity site, based on an earlier study (19).

3.2. Sampling and laboratory analysis:

Due to the dominance of *H. strobilaceum* in the plant community, it was sampled and other species were neglected because of their temporality and seasonal coverage. Therefore, on Nov. 2012, when biomass grew completely, in each site, about fifteen *H. strobilaceum* were randomly selected at five transects with 100 meters length (three plant for each transect). The distance among transects was about 30 meters. The soil was caved and the above ground and underground organs of plants were extracted and then separated. All parts were weighed in the field and 500 gr of each fresh organ was transferred to lab for determining the content moisture coefficients and biomass C. Also, the plants density was counted on the 4m×10m quadrates in each site and the results were generalized in hectare (plants per hectare). Regarding the maximum rooting depth in the area (20), three soil samples were taken at each depth of 0-20 cm and 20-40 cm on each transects. After combining the samples, finally one composite soil sample was prepared for each transect. The intact soil after removing gravel was taken by cylinder to determine the soil bulk density and to calculate the storage C of soil. The 500gr biomass samples were dried in oven at 70°C for 24hr and the moisture coefficient was determined. Then, the overall dry weight was determined by using this coefficient. 10gr of dried samples was selected to estimate the C content by combustion in electrical kiln at 500°C for 5hr. The reduced weight in this method belongs to the organic matters, 56% of which is organic C. Then the C conversion coefficient for each organ was calculated by dividing the organic C weight into dried sample weight (10gr). After that, the storage organic C in each plants organ was determined by multiplying this one by overall dried organs weight (21).

The dried soil samples were sieved by 0.5mm sieve in laboratory and had its organic C content determined by Walkley-Black method, based on wet oxidation. The weight of stored SOC or sequestrated organic C storage within a certain depth was estimated using Equation 1 (22).

\[
SC = 100 \times OC \times Bd \times D
\]

where, SC is the C content at specified depth (ton per hectare in a given depth), OC is the soil organic C percent, Bd is the soil bulk density with removed gravel (gram per cubic centimeter), and D is the soil depth (m).

3.3. Statistical analysis

The data normality test for the sampling number was conducted using Anderson and Darling test and the abnormal data were normalized by data transformation before data analysis. The logarithm transformation was applied for the data. The two ways ANOVA was used to find the effect of grazing intensity and soil depth on the parameters of C conversion coefficient, organs C and organic C of soil. One-way ANOVA was used to analyse mixture of treatments (sites and depths) and the post hoc test (Duncan’s test, with a critical significance level of 0.05) was used to compare the mean of parameters. Levene’s test was also used for determining equality of variances. T-test was also used to compare the means between two organs of below and aboveground...
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The SPSS version 19 was used for all statistical analysis.

4. Results

The ANOVA results of C conversion coefficient of root and above ground organ in three sites (low, moderate and high grazing) and also t-test of the parameters between organs indicated a significant difference (p<0.05) between C conversion coefficient in above ground organ and root in three sites (Table 1). Root organ had a higher C conversion coefficient than the aboveground organ. This parameter for each organ was, however, not significantly different for the three sites. The C content of both root and the aboveground organs were significantly higher in light grazing than the other two sites. The C content of the aboveground organ was also significantly higher than that of root in the light grazing site, but the two other sites did not show significant differences in C content between the two organs (p<0.05). Finally the results of biomass C per hectare showed that both moderate and low grazing sites were in one group and had greater amount of biomass C than the high grazing site.

The two-way ANOVA revealed that grazing intensity (site) and soil depth had significantly (p<0.05) affected the amount of soil organic C (Table 2), but the interaction effect of livestock grazing and soil depth was not significant. Furthermore, the changes of C trend in different sites and depths remained constant in all three sites.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Organ</th>
<th>Light</th>
<th>Moderate</th>
<th>Heavy</th>
<th>F-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>C conversion Coefficient</td>
<td>Root</td>
<td>0.55±0.08</td>
<td>0.51±0.03</td>
<td>0.50±0.02</td>
<td>4.05*</td>
</tr>
<tr>
<td></td>
<td>Aerial</td>
<td>0.49±0.02</td>
<td>0.46±0.06</td>
<td>0.45±0.02</td>
<td>4.22*</td>
</tr>
<tr>
<td>t-value</td>
<td></td>
<td>5.79**</td>
<td>2.81*</td>
<td>12.31**</td>
<td></td>
</tr>
<tr>
<td>Organs C content (gram)</td>
<td>Root</td>
<td>42.6±24.6</td>
<td>24.3±15.18</td>
<td>12.9±10.77</td>
<td>10.32**</td>
</tr>
<tr>
<td></td>
<td>Aerial organ</td>
<td>39.2±22.75</td>
<td>22.8±12.58</td>
<td>21.9±6.56</td>
<td>6.24*</td>
</tr>
<tr>
<td>t-value</td>
<td></td>
<td>0.53±0.41</td>
<td>-3.76**</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>plant C content (gram)</td>
<td>-</td>
<td>81.31±46.88</td>
<td>34.97±16.23</td>
<td>9.80**</td>
<td></td>
</tr>
<tr>
<td>Density (shrub per hectare)</td>
<td>-</td>
<td>14510</td>
<td>23100</td>
<td>16230</td>
<td></td>
</tr>
<tr>
<td>Total biomass C content per unit area (ton per hectare)</td>
<td>-</td>
<td>1.17±1.08</td>
<td>0.567±0.45</td>
<td>6.13**</td>
<td></td>
</tr>
</tbody>
</table>

a, b, c: Significant parameter among sites A, B: Significant parameter between organs in specific site
**: difference at p<0.01 probability level *
: difference at p<0.05 probability level, ns: non-significant between groups

<table>
<thead>
<tr>
<th>Source</th>
<th>sum of square</th>
<th>d.f</th>
<th>mean square</th>
<th>F_value</th>
<th>Sig.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Site</td>
<td>855.439</td>
<td>2</td>
<td>4277.71</td>
<td>30.50</td>
<td>0.00</td>
</tr>
<tr>
<td>Depth</td>
<td>1164.895</td>
<td>1</td>
<td>1164.89</td>
<td>8.31</td>
<td>0.008</td>
</tr>
<tr>
<td>depth x region</td>
<td>204.406</td>
<td>2</td>
<td>102.20</td>
<td>0.72</td>
<td>0.49</td>
</tr>
<tr>
<td>Error</td>
<td>3365.074</td>
<td>24</td>
<td>140.21</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>130938.989</td>
<td>30</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**: Significant at 1% probability level n.s: non-significant

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The results of post hoc for soil C storage and soil bulk density in sites and depths showed that the low grazing site in 0-20 cm and 20-40 cm depths and moderate grazing site for 0-20 cm depth had the maximum amount of C with values 89.15, 73.41 and 74.47 ton/hectare, respectively. Also, the average bulk density in high grazing intensity in both depths was more than the other sites (Table 3).

The differences of the total stored C (soil and biomass) showed the low grazing site had 32.3 ton/hectare of C more than moderate grazing site that, in turn, had 57.07 ton/hectare of C more than high grazing site (Figure 2). It should be noted that the biomass C is too low in value to be visible in the figure.

The relative contribution of each ecosystem components in C storage is shown in Figure 3. Share of the first depth (0-20m) is more than the second depth (20-40m) for the three grazing sites. The major C storage for first depth was found in the moderate grazing site (55.8%), while the major C storing in the second depth belonged to the high grazing site (46.52%). The highest level of biomass C was recorded in the moderate site (0.81%), but obviously it was less than soil C in all the three sites (Figure 3).

<table>
<thead>
<tr>
<th>Region</th>
<th>Soil depth</th>
<th>C storage</th>
<th>Standard Deviation</th>
<th>soil bulk density</th>
<th>Standard Deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Light grazing</td>
<td>0-20</td>
<td>89.15a</td>
<td>14.5</td>
<td>1.41</td>
<td>0.08</td>
</tr>
<tr>
<td></td>
<td>20-40</td>
<td>73.41a</td>
<td>14.28</td>
<td>1.62</td>
<td>0.069</td>
</tr>
<tr>
<td>Moderate grazing</td>
<td>0-20</td>
<td>74.47a</td>
<td>15.39</td>
<td>1.57</td>
<td>0.03</td>
</tr>
<tr>
<td></td>
<td>20-40</td>
<td>57.92b</td>
<td>10.93</td>
<td>1.70</td>
<td>0.05</td>
</tr>
<tr>
<td>Heavy grazing</td>
<td>0-20</td>
<td>42.93bc</td>
<td>5.87</td>
<td>1.86</td>
<td>0.05</td>
</tr>
<tr>
<td></td>
<td>20-40</td>
<td>37.83c</td>
<td>5.98</td>
<td>1.86</td>
<td>0.07</td>
</tr>
</tbody>
</table>

a, b. Significant or insignificant parameter between sites in each depth

![Figure 2](image_url)
5. Discussion

In this study, livestock had different effects on the various C storage parameters. A significant difference between C conversion coefficient in the above-ground organ and root within all three sites was observed. The C conversion coefficient of root in low, moderate and high grazing sites was higher than the above ground organ. Fang et al. (23) found the maximum and minimum C conversion coefficient was in the stems and leaves, respectively.

The above-ground organ of plant is the most important part of an ecosystem that has direct effect on the grazing. The highest and lowest reduction of C storage in the above-ground organ was related to the low and high grazing intensity sites, respectively. The grazing intensity on moderate grazing treatment was balanced so the C storage of the above-ground organ for this site was significantly lower. This is due to the direct effects of grazing on vegetation cover and its indirect role on reduction of storage C in overall ecosystem by soil erosion and desertification (24), which could have also negative effects on the above-ground organ and probably on the underground organ as well (25, 26).

Some studies have shown a direct relation between the biomass reduction of the above-ground and underground organ of plants with the grazing intensity and harvesting volume (14). In this study, too, the grazing intensity was associated with the reduction of C storage. The non-significance change of the underground organ biomass in moderate grazing treatment can be attributed to the lower grazing. Therefore, the lower harvest intensity from photosynthesizing organs in moderate grazing caused no significant effect on growth and extension of the underground organ biomass. However, there was a significant reduction in the underground organ biomass in the high grazing treatment due to harvesting of the above ground organ and problems in photosynthesis process and the reduced physiological activity of the plants (27). Niknahad Gharmaker et al. (28) also found a significant difference between grazing intensities for the stored C in biomass and soil.

Joneidi Jafari et al. (29) concluded that root growth and extension in high grazing site was limited when compared with the moderate grazing site. While the C storage in the above-ground organ was less than root in the three sites, the high grazing reduced the root biomass and increased the root ration in the total plant biomass.
biomass, which caused the C storage in the above-ground biomass less than the total C storage of the ecosystem. The results of organs and the total C per unit area showed the highest amount of C for root organ and low grazing site, followed by the moderate grazing site. Also, heavy grazing intensity led to higher levels of soil-plant C through changes in species composition. Nonetheless, heavy grazing markedly decreased vegetation cover and the above-ground biomass, that is undesirable for livestock production and sustainable rangeland development.

Grazing at light to moderate grazing intensity resulted in stable and diverse plant communities dominated by forage range with high above-ground biomass (30). Also, higher organic matter inputs from plant litter and root exudates in un-grazed soil, compared to lower organic input in the grazed soil, may have enhanced the rate of biomass C production, a labile organic C faction, in the soil (31). Some researches, such as Eccard et al. (32), stated the vague response of root to grazing, but it had been shown that the roots had a key role on C sequestration in ecosystems for being the major route of the entered C and nitrogen into the soil (9). Naseri et al. (33) found significant differences between treatments for the total C stocks (soil, biomass and litter) and showed that the soil had a main role in C sequestration, followed by biomass, roots and litters among the components of the ecosystem. Also, Khosravi Moshizi et al. (34) studied C sequestration hotspots and found that soil characters played effective roles in C stocking in semiarid rangeland ecosystems.

High vegetation cover of the ungrazed site increases the underground biomass and the C storage as well. The results of organic C in soil showed that there was a significant difference in C storage between all sites and depths. Plants use the litter falling to transfer their organ C to the soil (35). The livestock grazing cann't change the C content in soil that depends on grazing intensity and soil depths. Mahdavi and Esmaiali (36) showed that the rates of C sequestration in different species, vegetation organs and soil layers differed, being higher in woodying species and the upper soil layer. Henderson et al. (3) investigated the reaction of soil C on long term enclosure in Canada and found a higher amount of C in vegetation cover and litter in non-grazing treatment. The C content in 0-20cm depth was more than in 20-40cm depth in the three sites of the present study. The highest C storage was recorded in the moderate grazing site at the first depth (55.8%) and in the high grazing site at the second depth (46.52%). Also, the moderate grazing site had the highest C biomass (0.81%) at the three sites, which was obviously less than the soil C in the three sites. This shows the importance of soil conservation, especially in surface layers to reduce the C sequestration loss in ecosystem, since the recovery of soil C loss is slow.

Finally, the soil erosion could causes C loss as most part of C is stored in the soil. Therefore, the soil C storage for low grazing site is maximum and for high grazing site is minimum. This is due to the reduction of litter by harvesting the plant organ in high grazing site (31). Also, the high bulk density in region with high grazing intensity may cause erosion.

The high bulk density and reduction of organic matter in soil could produce from reduction of ground and underground organic matter in soil and livestock movement in field (9). Well-managed grazing improves nutrient cycling in rangeland ecosystems, stimulating the above-ground production as well as root respiration and exudation rates (31). Differences in methodology of sampling and analyses, plant response to grazing, and photosynthesis of grazed plants may be responsible for different results of grazing on organic matters (31).
6. Conclusion
The livestock grazing could influence the vegetation composition and the ratio of plant material allocation. The stored C in rangeland ecosystem could be controlled by the grazing intensity and frequency through grazing systems (such as rotation, deferred rotation, and rest rotation grazing system). If the vegetation cover and rangeland production capacity is not affected by high livestock grazing and the livestock number is based on the maintenance capacity, the organic matter of soil does not change. However, destruction of vegetation cover, high soil erosion, and loss of C storage as a consequence could be occurred in high grazing site. The soil especially first depth as main resources of C should be protected from deterioration to prevent C declining. In view point of C sequestration, it is suggested to plan grazing intensity in light level and protect the soil of rangelands.

Conflict of Interest
The authors declare that they have no competing interests.

Acknowledgment
We would like to thank anonymous reviewers for their valuable comments and suggestions.

Authors’ Contributions
Each of the authors has contributed in writing of this paper equally.

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تغییرات ترسیب کربن در گونه Halocnemum strobilaceum و خاک بر اثر چرای دام (مطالعه موردی: استان گلستان، ایران)

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تمامی مطالعات قبلی نشان دادند که چرای دام تاثیر عمده‌ای بر روی کربن آلی خاک و زیست‌ده دارد. گیاهان مختلف و اکوسیستم‌های با شرایط متفاوت، پاسخ‌های منحصر به فردی به چرای دام دارند. شناخت تاثیر شدت چرای دام بر روی کربن در محیط‌های گوناگون نظیر اراضی شور اینچه درون، استان گلستان که پوشیده از گونه Halocnemum strobilaceum (باتلاقی شور) است، به برنامه‌ریزی استراتژی‌های جهت حفظ کربن می‌گردد.

مواد و روش‌ها: به این منظور، با توجه به فاصله از محل استراحت و نیمروزی دام‌ها در مرتع، سه محل شامل چرای شدید دام (در مجاورت منطقه استراحت)، چرای متوسط (در 156 متری محل استراحت) و چرای متوسط (در 2656 متری محل استراحت دام) انتخاب شدند. نمونهبرداری به صورت سیستماتیک تصادفی از خاک و گیاه گونه H. strobilaceum، در آبان 1391 صورت پذیرفت. در این خصوص استخراج نمونه‌برداری از هر یک از سایت‌ها به ترتیب در دو عمق 0-20 و 20-60 سانتی‌متری صورت گرفت و کربن خاک به روش والکی-بلك تعیین گردید.

نتایج: مجموع کربن بیوماس اندام زیرزمینی و هوایی برای گونه مذکور در سایت چرای سنگین بیشتر از سایت‌های چرای متوسط و سنگین بود. با انتخاب نقاط محل استراحت و نیمروزی در حدود 0-11, 0-61 و 0-511/6 تن در هکتار، سایت‌های چرای سنگین و متوسط به ترتیب در حدود 0-11/1, 0-61/1 و 0-511/6 تن کربن کمتری نسبت به سایت چرای سنگین داشتند.

بحث و نتیجه‌گیری: از دیدگاه مدیریتی، یکی از منافع اصلی کشت چرای دام در منطقه شور به ذخیره کربن در خاک باید به‌شمارآید. شرایط محیطی سایت‌های چرای دام، بایستی به‌طور کامل در نظر گرفته شوند و تخصیص منابع مالی به برنامه‌های حفاظت خاک در این منطقه ضروری می‌باشد.

کلمات کلیدی: Halocnemum strobilaceum، ترسیب کربن، خاک، زیست‌ده