Soil Seed Bank Characteristics in Relation to Distance from Watering-Points in Arid Ecosystems (Case Study: Kahnuj, Kerman Province)

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ABSTRACT Epizoochory and endozoochory are well-known mechanisms by which seeds are dispersed by animals. Since, livestock gather around the watering-points to rest and drink, we expected to find that seeds would be frequently moved to the areas surrounding watering-points, resulting a higher soil seed density (and diversity) closest to the water. We investigated this issue by assessing soil seed bank density and composition along a distance gradient from the water and compared them with an ungrazed-control area in 2010. Therefore, three watering-points were selected in a dry rangeland of Kahnuj, Kerman Province, Iran. Soil seed bank characteristics were measured at eight distances with different intervals from the watering-points (totally 80 sampling points) and in an ungrazed area (30 sampling points) after seed dispersion in autumn. The results showed that in spite of the assumption, soil seed density and similarity between the soil seed bank and above ground vegetation increased with distance from the watering-points. A clear trend was not detected for the diversity of soil seed bank with distance from water source. Our findings suggested that the restoration of degraded sites could not rely on soil seed bank. Areas surrounding watering-points, where soil seed losses are potentially higher than those at greater distances from water source, should be given more attention for conservation by the rangeland managers.

1   INTRODUCTION

Gradients of grazing over increasing distances from watering-points have been used extensively to investigate the impact of livestock on rangeland vegetation. It can be concluded from the literature that the heavy use of vegetation around watering-points had always occurred and the intensity of grazing decreased with distance from the water (e.g. Van Rooyen *et al.*, 1994; Howes and Mc Alpine, 2008).

Patterns in soil seed banks has been highly varied spatially and temporally, and strongly influenced by factors are that influence seed production (Erfanzadeh *et al.*, 2010b), such as grazing. In fact, the magnitude of spatial heterogeneity in soil seed banks is strongly influenced by livestock pressure due to selective grazing (e.g. Solomon *et al.*, 2006). For instance, high grazing pressure favours annual species, which show various adaptations due to tolerate or avoid intensive grazing and trampling, such as the production of huge

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numbers of small seeds which could be buried easily and join the soil seed bank (Erfanzadeh *et al.*, 2010a; Wu *et al.*, 2015). As a result, it could be assumed that soil seed size might be higher when closer to water points because of higher grazing intensity. However, this assumption could also suggest that soil seed density might increase at greater distances from water locations because it has more opportunity for seed production under conditions of lower grazing intensity.

Moreover, epizoochory (seed dispersal on the body surface of mammals) and endozoochory (seed dispersal via ingestion by mammals) were well-known mechanisms by which seeds were dispersed by grazing animals (Poschlod *et al.*, 2005). Since grazers gather around the watering-points to rest and drink, we expected to find that seeds were frequently transported to the area in the immediate vicinity of the watering-point and penetrate the soil. Therefore, it could be hypothesised that a proportion of the seeds produced by plants further away might be dispersed, deposited closer to watering-points and finally became a part of the soil seed bank. Seed banks closest in distance to watering-points would then be a combination of seeds of all species growing further away, resulting in increased soil seed density and richness around the watering-point (Nsinamwa, 2007).

Hence, future plans and decisions for restoration of degraded sites may require more information on potential capacities of soil seed banks and studies dealing with vegetation in the arid rangeland of Iran have been restricted to the above ground vegetation communities, and have ignored seed banks stored in the soil (which were part of the plant diversity), the recording of which requires more effort. The objectives of this study were i) to examine seedling density and composition along distance gradient from the water locations and ii) to assess the similarity between the seed bank and aboveground vegetation. iii) to assess the potential of soil seed bank for restoration of degraded sites. We also used an exclosure about 5000m away from the watering-points as a control area.

2 MATERIALS AND METHODS
2.1 Description of the study sites
The study was carried out in the Kahnuj rangelands situated in the south of Kerman Province, Iran. The climate was arid, with an average annual rainfall of 216 mm. The monthly average temperature was maximum in January with 51°C and is minimum in July with 0°C. The study area was plain and less hilly with low variation in topographical characteristics. The vegetation type was shrubland with dominancy of *Hammada salicornia* (Moq.) Iljin and *Gaillonia aucheri* (Guill.) Jaub. and Spach.

Drinking water for livestock was provided mostly by digging semi-deep wells (with ca. 15 m in depth; water was taken out by pail) (personal communications). In the Kahnuj rangelands, three water-points were selected: Kuhsorkh (57°, 42´, 45.02" E. and 28°, 04´, 41.05" N.), Talkhab (57°, 46´, 28.81" E. and 28°, 02´, 43.23" N.) and Rishehkonar (28°, 04´, 21.04" E. and 57°, 41´, 45.18" N.) (Figure 1) (the latitudes and longitudes were estimated by GPS apparatus in the field).

In addition, a small part of the area (ca. 5km far away the watering-points) was excluded from grazing by fencing 11 years ago (in 2001) by the Iranian Research Center of Natural Resources. Since all exclosure characteristics were similar to grazing area, climatologically and topographically, this exclosure created an opportunity to study some vegetation traits and to compare them with the grazing area. Sheep and goats were the most important livestock that roam throughout the year (personal observation).
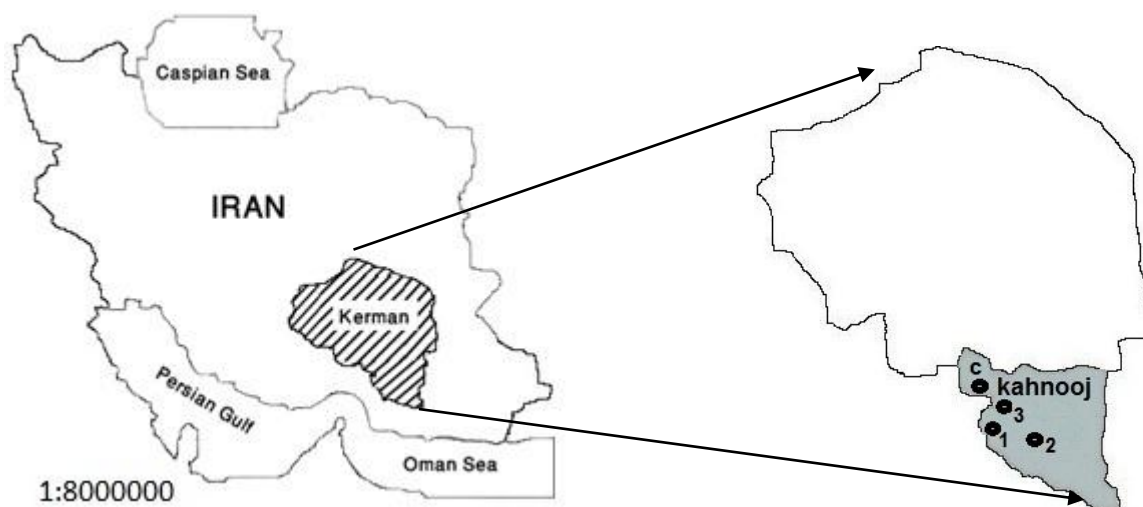
Figure 1 The location of three watering-points in southern Kerman Province: 1) Rishehkonar, 2) Talkhab, 3) Kuhsorkh and c) Control area

2.2 Soil sampling

According to statistical requirement (10 replications in each distances) and previous studies (e.g. Nsinamwa, 2007), we selected 8 distances from each water-point i.e. 0 m (adjacent to the water-point), 20, 50, 100, 120, 150, 200 and 300 m. At each distance, 10 ($4m^2$ in size) quadrats were randomly established. In addition, 30 quadrats ($4m^2$ in size) were also established inside the exclosure. Soil samples in all quadrats were then collected in February 2010 before the seeds germinated (Nsinamwa, 2007). In each quadrat, 20 soil cores were collected at random down to a depth of 10cm and these samples were divided into two subsamples (0-5 and 5-10 cm) which were then pooled per soil layer for each quadrat. The subsamples were transported to the green house and were spread out in a thin layer (maximum 0.4 mm thick) in 26×40 cm trays filled with sterilized potting soil. The trays were placed randomly on shelves with a natural light regime and were kept moist by regular hand watering. In addition, 20 control trays, filled with the same sterilized potting soil were placed randomly on the shelves to test for possible glasshouse contamination. Seedlings were identified as soon as possible and were removed regularly during the 7-month greenhouse trial. Any plant that could not be identified at the seedling stage was allowed to grow until identification was possible (e.g. Erfanzadeh *et al.*, 2010b).

The above ground vegetation composition was determined during the 2010 growing season by visually estimating (using nested plots) the cover of all vascular plant in all quadrats (Heady and Child, 1999).

2.3 Data analysis

Seed density was calculated per m^2 for each 5 cm soil depth layer and for each species separately. Seed density was transformed to log10 (X+1) to meet normal distribution, assessed by Kolmogorov-Smirnov test. Correlation between the above-ground vegetation and the soil seed bank was assessed with Sørensen`s qualitative similarity index (Kent and Coker, 1994) for each depth separately. To determine the species diversity of the soil seed bank, the Shannon-Wiener index for each quadrat and for each depth was determined as following:

Shannon-Wiener diversity index: H' = -ΣPi ln *Pi*,

In which *Pi* was proportion of the *i*th species (Kent and Coker, 1995) for each depth.

Univariate general linear modeling (GLM) and LSD test were used to compare the mean of seed density, diversity and similarity (between above-ground vegetation and the soil seed bank) between depths and between 9 distances from water source. Seed density and other seed characteristics were introduced as dependent variables, while depth categories (0-5 and 5-10 cm) and distance from water categories (0 m, 20, 50, 100, 120, 150, 200, 300 and 5000 m) were introduced as fixed factors. An interaction between depth and distance from water source

was also considered in the model. All statistical analyses were done using SPSS. 17.

3 RESULTS

Species composition in the soil seed bank and above-ground vegetation was determined. In total, 15 species germinated from the soil samples. The dominant plant species in the entire area (three water-points and exclosure) were *Stipa capensis, Asphodelus tenuifolius* and *Astragalus triboloides* (Table 1).

In total, 20 species were recorded in the above-ground vegetation in the entire study area. The dominant plant species were *Stipa capensis* and *Asphodelus tenuifolius* (Table 2).

Table 1 Average seed density per square meter of each species in the soil seed bank around different watering-points and exclosure area

Area (Watering-point and Ungrazed)	Kuhesorkh		Rishekonar		Talkhab		Exclosure	
Depth (cm)	0-5	5-10	0-5	5-10	0-5	5-10	0-5	5-10
Density /m^2	299.16	28.99	260.60	11.21	181.40	1.81	786.75	14.26
Asphodelus tenuifolius L.	36.69	2.04	45.10	0.25	27.86	0.00	64.20	33.12
Astragalus triboloides Dlile.	29.21	2.49	36.18	1.27	29.21	0.68	78.47	40.76
Astragalus sp.	2.04	0.23	5.61	0.00	0.23	0.00	0.00	0.00
Chenopodium album L.	2.26	0.00	2.04	0.00	0.23	0.00	0.00	0.00
Fagonia bruguieri DC.	19.48	5.89	8.66	0.25	0.45	0.00	0.00	0.00
Forsskaolea tenacissima L.	0.00	0.00	2.16	0.00	0.00	0.00	0.00	0.00
Hordeumvulgare L.	0.45	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Infloga spicata Boiss.	26.72	3.17	22.17	5.61	29.44	0.00	214.01	108.54
Infloga sp.	17.66	4.53	4.84	0.00	3.40	0.00	101.91	50.96
Malvaneglecta L.	12.46	2.04	7.64	0.51	1.59	0.00	0.00	0.00
Medicagosativa L.	0.00	0.00	0.00	0.00	1.13	0.00	0.00	0.00
Paronychia sp.	0.00	0.00	2.19	0.00	0.00	0.00	0.00	0.00
Plantago stocksii Boiss. and Decne.	15.17	0.23	41.27	0.00	12.00	0.00	61.15	31.59
Solanum lycopersicum L.	1.13	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Stipa capensis Thunb.	11.50	8.38	82.55	3.31	70.66	1.13	127.39	0.00
Taraxacum officinale F.H.Wigg.	15.63	0.00	2.29	0.00	5.21	0.00	16.31	8.15
Unidentified Graminoides	4.76	0.00	0.00	0.00	0.00	0.00	0.00	0.00

Table 2 Average cover (%) of each species in the above ground vegetation in the grazed and exclosure areas. A=annual and P=perennial

Species	Life form	Around watering-points	Exclosure
Asphodelus tenuifolius L.	A	5.31	10.50
Astragalus tribuloides Dlile.	A	3.09	10.00
Calligonum bungei Boiss.	P	0.52	5.60
Convolvulus leptocladus Boiss.	P	0.61	0.00
Fagonia bruguieri DC.	A	1.07	5.32
Forsskaolea tenacissima L.	A	0.07	0.00
Gaillonia aucheri Jaub. and Spach	P	0.31	2.12
Gymnocarpus decander Forssk.	P	0.06	0.00
Hammada salicornica (Moq.)Iljin	P	2.39	5.10
Infloga spicata Boiss.	A	3.69	10.03
Lycium edgeworthii Dun.	P	0.07	0.00
Plantago stocksii Boiss. and Decne.	A	2.93	7.10
Prosopis spicigera L.	P	0.04	0.00
*Pteropyrum aucheri*Jaub. and Spach	P	0.19	2.01
Rhazya stricta Decne.	P	0.28	1.00
Stipa capensis Thunb.	A	8.80	15.31
Taraxacum officinale F.H.Wigg.	A	0.19	0.00
Taverniera cuneifolia (Roth) Arn.	P	0.41	0.00
Ziziphus spina-christi (L.) Willd.	P	0.31	0.00
Zygophyllum eurypterum Boiss. and Buhse	P	0.04	0.00

3.1 Soil seed bank density

The GLM results showed that distance from water source (F=7.97 and P<0.01) and soil depth (F=142.80 and P<0.01) affected the density of the soil seed bank significantly, while the effect of distance from water × depth on soil seed bank density was not significant (F=6.95 and P=0.08). Mean seed densities per square meter in the upper layer of Kuhsorkh, Rishekonar and Talkhab were 299.16, 260.60 and 181.40, respectively. Mean seed densities per square meter in the lower layer of Kuhsorkh, Rishekonar and Talkhab were 28.99, 11.21 and 1.81, respectively (Table 1).

Mean seed densities in the 0-10 cm depth, at 0, 20, 50, 100, 120, 150, 200, 300 and 5000 m from water source were significantly different, 105.37, 135.54, 186.84, 158.30, 152.18, 337.66, 368.33, 463.35 and 786.75 seeds per m^2

(F=15.39 and P<0.001), respectively. The lowest seed density occurred at distances of 0 and 20 m and the highest was related to furthest distance from water source, i.e., 200, 300 and 5000 m (F=15.39 and P<0.001) (Figure 2).

3.2 Species diversity in the soil seed bank

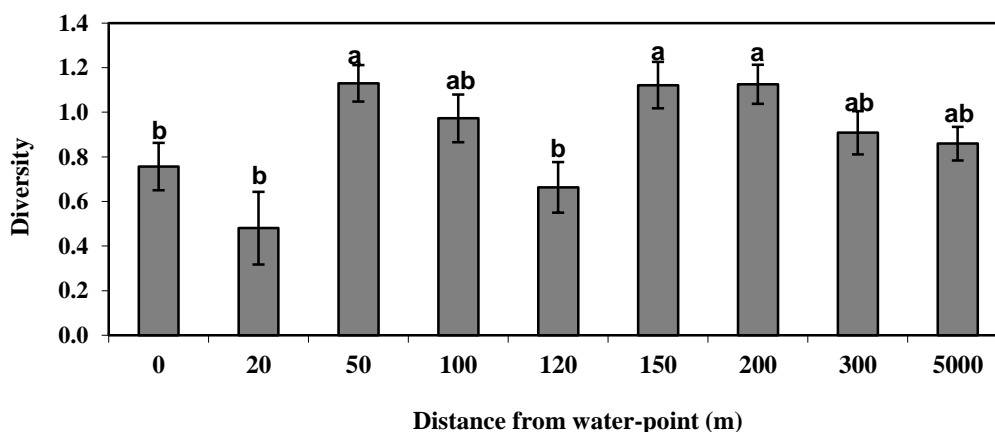The GLM results showed that distance from water source (F=1.30 and P<0.05) and soil depth (F=97.30 and P<0.01) significantly affected species diversity of the soil seed bank. The interaction effect between depth and distance from water source on soil seed bank diversity was not significant (F=1.83 and P=0.09).

Mean species diversity of the soil seed bank in the 0-10cm depth was lowest at 0, 20 and 120 m from water source while it was highest at 150 and 200m from water source (Figure 3).
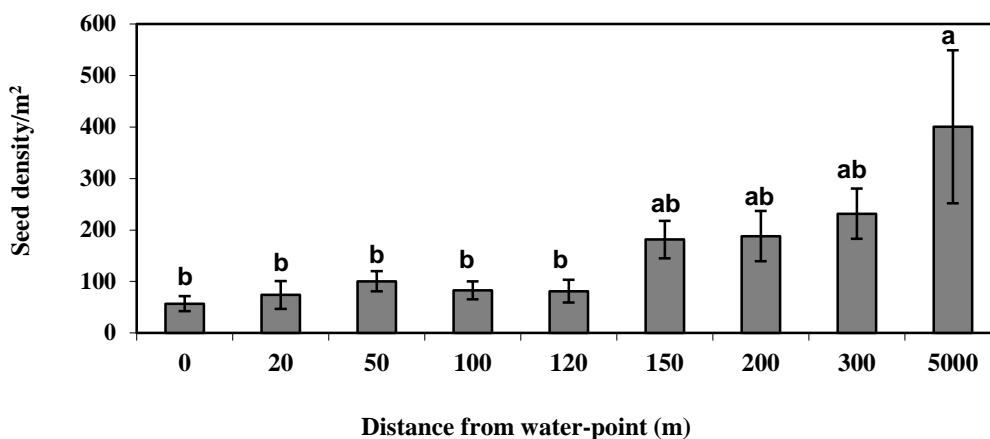
Figure 2 Variation of soil seed density (per m^2) along distance from water source

Small successive letters indicate significant differences of soil seed bank density between distances from water source within 0-10 cm depth
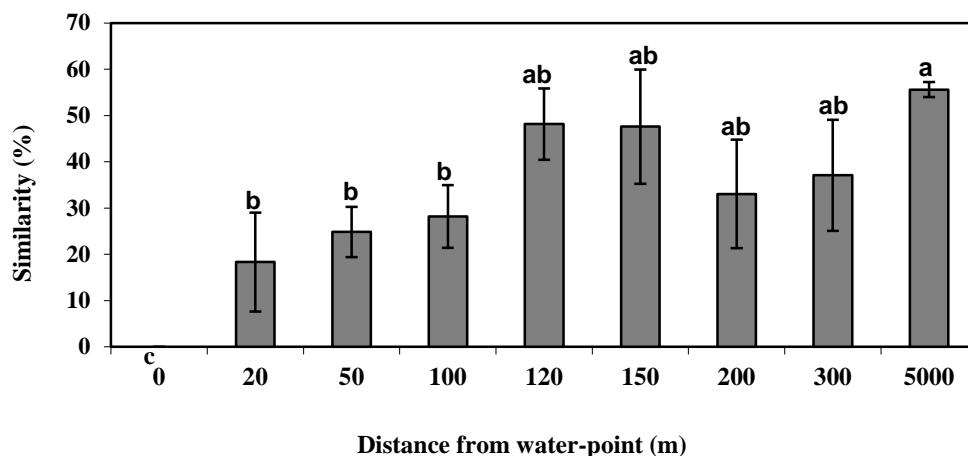

Figure 3 Variation of species diversity of soil seed bank along distance from water source

Small successive letters indicate significant differences of soil seed bank diversity between distances from water source within 0-10 cm depth

3.3 Similarity between the soil seed bank and above-ground vegetation

The results showed that distance from water source (F=5.85 and P<0.01) and soil depth (F=28.06 and P<0.01) significantly affected the similarity between the soil seed bank and above-ground vegetation. The interaction effect between depth and distance from water source on the similarity between soil seed bank and above-ground vegetation was not significant (F=2.06 and P=0.08). Mean similarity between the soil seed bank and above-ground vegetation was highest in the ungrazed area while it was lowest (zero) in quadrats closest to the watering-points (Figure 4).

Figure 4 Variation of Sørensen`s similarity index along distance from water source.

Small successive letters indicate significant differences of the similarity index between distances from water source within 0-10 cm depth

4   DISCUSSION

In an arid zone, the animals forage outwards from a watering-point, to which they were obliged to return frequently to drink. Therefore, animals graze and had an uneven impact on habitats relative to distance from water source, with a higher intensity in areas which were closer to watering-points (Heady and child, 1999). Our results showed that intensive grazing close to watering-point strongly decreased the seed bank density. Comparison of the soil seed bank density at different distances from water source confirmed that the grazing impact was one of the most important causes of variation in seed bank population in arid environments. Heavy utilization and defoliation (Hempy-Mayer and Pyke, 2008) and preventing of plant full-growth by grazing livestock (Omar *et al.*, 2013) might result in a reduction of the seed production capacity of plants. In other words, grazing by livestock affected the reproductive output of plant species negatively by removing reproductive tissues and reducing seed production (Paruelo *et al.*, 2008). In addition, trampling by livestock may also affect

the structure of the upper soil layer negatively by destroying the soil aggregates (Du Toit, *et al.*, 2009; Bertiller and Ares, 2011). Consequently, the contribution of seeds to the seed bank was reduced by intensive grazing. Other investigations in dry and semi-dry rangelands have been reported similar results regarding the influence of increased grazing on the reduction of the soil seed bank size (Sternberg *et al.*, 2003; Osem *et al.*, 2006; Solomon *et al.*, 2006; Kassahun *et al.*, 2009; Dreber *et al.*, 2011). In contrast, Dreber and Esler (2011) reported that long-term heavy grazing increased seed densities and species richness of the soil seed bank by favoring small-seeded and tiny-seeded annual species. Some researchers found that grazing had no impact on total seed density (Kinloch and Friedel, 2005). However, the results showed that, at distances up to 120m from water source, soil seed density was drastically lower than at greater distances. Thus, it can be concluded here that not only above ground vegetation, but also the soil seed bank were negatively affected by grazing and that they were intensively degraded around watering-points.

Moreover, previous studies showed that the impact of grazing on seed bank size would be more intensive if it coincided with drought. For instance, Kinloch and Friedel (2005) concluded that the impact of grazing on the seed bank depended on the severity of grazing during the preceding years and the accompanying drought at that time. Li *et al.* (2011) showed that annual rainfall had a significant and positive correlation with seed density and species number in the soil seed bank. During the previous years, there were signs of frequent and severe drought in Iran. For instance, from 1999 to 2002, the entire country experienced a multi-year drought (Salemi *et al.*, 2009). Therefore, this might have increased the intensity of the negative grazing effect on the soil seed bank in these years.

The results of the present study suggest that the displacement of watering-points every few years was necessary to allow degraded sites to renew/conserve their soil seed bank density (and richness). Moreover, in arid and semiarid rangelands, recovery processes in degraded sites (here closest to watering-points) was slow and stochastic (Call and Roundy, 1991; Visser *et al.*, 2004) and the time span for improvement might last several years (Allington and Valone, 2010). As a result, active seed species introduction may be the only way to achieve the recovery of degraded sites through seeding. Perhaps seed rich topsoil replacement and sowing of target species (Visser *et al.*, 2004; Solomon *et al.*, 2006; Dreber *et al.*, 2011) implemented within a long-term strategic restoration framework, will be necessary in order to improve rangeland condition and prevent local species extinction. However, the success of species recruitment and establishment was strongly dependent on climatic conditions, particularly in Kerman Province, which suffers from drought once every few years (Beheshti Rad and Beheshti Rad, 2013).

We did not detect a clear trend for species diversity of the soil seed bank along a distance gradient from water source. However, up to 20m from the watering-points, species diversity of the soil seed bank was lowest compared to the ungrazed area, where species diversity of the soil seed bank was intermediate. In previous studies, species diversity (or richness) in soil seed banks were often reported to decrease with grazing intensity, to increase with grazing intensity or to show no clear response (Snyman, 2004; Dreber and Esler, 2011).Similarities between the seed bank and above-ground vegetation ranged from a low of zero at the distance closest to the water-points (i.e. no species in common) to 0.56% in ungrazed sites. In areas closest to water-points which had been subjected to heavy grazing, vegetation production capacities and their ultimate contribution of seeds to the seed bank were probably reduced (Kassahum *et al.*, 2009) and consequently this similarity decreased strongly.

Overall, similarity between the seed bank and above-ground vegetation was low, particularly in the grazing area. The absence of above-ground woody species from the seed bank (e.g. *Zygophyllum eurypterum*, *Ziziphus spina-christi*, *Taverniera cuneifolia*, *Pteropyrum aucheri*, *Prosopis spicigera*, *Hammada salicornica*, *Gymnocarpus decander*) caused the low similarity between the soil seed bank and above-ground vegetation, suggesting that woody species were not able to produce a persistent seed bank in arid rangelands. In contrast, annual species were found in the seed bank while they were absent in the above ground vegetation (e.g. *Chenopodium album*, *Hordeum vulgare*, *Malva neglecta*). Indeed, one another reason for the dissimilarity could be related to the late vegetation sampling time in the area. If we sampled vegetation sooner, at the beginning of the growing season, then annual species might be recorded, leading an increase in the

similarity between vegetation and soil seed bank. However, the generally low similarity between above-ground vegetation and the soil seed bank in our study confirmed that the above-ground vegetation did not necessarily reflect the soil seed bank. Therefore, it was argued that restoration of degraded sites in the study area could not depend on a high level of recruitment from the seed bank of species, many of which have disappeared from the established vegetation even after grazing cessation (Kassahun *et al.*, 2009; de las Heras *et al.,* 2015). In contrast, few researchers reported relatively high similarities between soil seed banks and above-ground vegetation in part or all of their study area (Hopfensperger, 2007; Tessema *et al.*, 2012).

5   CONCLUSION

According to the results of this study, assumption of higher soil seed density and diversity in the areas closest to watering-points, because of epizoochory and endozoochory, could not be true in our study area, since soil seed bank density and diversity were lowest around the watering points compared to the further areas. Managers should pay more attention to areas surrounding watering-points, which have more potential for soil seed losses than those further away.

6   ACKNOWLEDGMENT

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خصوصیات بانک بذر خاک در ارتباط با فاصله از آبشخوار در اکوسیستم‌های خشک
(مطالعه موردی: کهنوج، استان کرمان)

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چکیده پخش بذر بهوسیله دستگاه هاضمه و پشم دام از روش‌های مشهور انتشار بـذر در مراتـع اسـت. از آنجـائی کـه دام‌های چرا کننده مکرراً در اطراف آبشخوارها برای شرب آب و استراحت جمع مـی‌شـوند، بـذرهای مرتعـی مـی‌تواننـد بهوسیله آن‌ها از سایر قسمت‌های مرتع به اطراف آبشخوار منتقل شوند و باعث افزایش تراکم و تنوع بانک بذر خـاک در این نقاط شوند. برای مطالعه یک چنین فرضیه‌ای، تراکم و ترکیب بانک بذر خاک را در طول گرادیان فاصـله از آبشخوار اندازه‌گیری و با یک مرتع قرق در سال ۱۳۸۹ مقایسه شد. بنابراین، سه آبشخوار در مراتع خشک کهنوج، اسـتان کرمـان انتخاب شدند و خصوصیات بانک بذر خاک در هشـت فاصـله از آبشـخوارها (مجموعـاً ۸۰ نقطـه نمونـه‌بـرداری شـده) و هم‌چنین منطقه قرق (۳۰ نقطه نمونه‌برداری شده) بعد از بذرپاشی گیاهان در پائیز تخمین زده شدند. نتایج نشان داد که بر خلاف تصور، تراکم بانک بذر و تشابه آن با پوشش روزمینی با فاصله از آبشخوار افزایش یافت. تغییرات تنوع بانک بـذر خـاک از الگوی مشخصی تبعیت نکرد. نتایج این تحقیق پیشنهاد می‌کند که جابجا کردن آبشخوارها هر چند سال یک بار جهت حفظ بانک بذر خاک و احیا پوشش در نقاط اطراف آبشخوارها ضروری است. نقاط نزدیک‌تر بـه آبشـخوارها یعنـی جاهایی که پتانسیل بیش‌تر برای تخریب بانک بذر خاک دارند بایستی بیش‌تر مورد توجه مرتعـداران قرار بگیرد.

کلمات کلیدی: آبشخور، بوته‌زار، *Gaillonia aucheri*، *Hammada salicornia*