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Germination success of gut-passed seeds of plant species in semi steppe rangelands: survival and ecological correlate with seed traits and standing vegetation

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Abstract

During last two decades studies on endozoochorous seed dispersal indicated that a large numbers of plant seeds are potentially dispersed and suceefully germinated via animal dung. However, very little is known about the relative importance of endozoochory in germination success of plant species in semi-steppe rangelands.. In this paper we examined dung germinating seed content, seed deposition patterns of different domestic animals (Cattle, Sheep and goat), ecological correlate with seed traits (Seed weight, length, width and shape) and the possible correlate of dung seed content characteristics with vegetation in a simulated feeding experiment and a field study in semi-steppe rangelands of Karsanak, Iran. 39 native plant species were fed to domestic animals and their germination successes were recorded in a simulated glasshouse experiment. In the field study, Animal dung (Sheep and goat) was collected during summer 2009 and 2001 and was placed under greenhouse conditions to record seed germination. The resulting species germination was compared to the standing vegetation of 60 4-m2 plots installed randomly along 6 200m transects.

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Introduction

Plant regeneration opportunities and dispersal has become an important issue in plant ecology and restoration management (Cosyns *et al.*, 2005). Large numbers of plant seeds are potentially dispersed and germinated via endozochory (Welch 1985; Malo & Suarez 1995a ; Pakeman *et al.*, 2002; Cosyns *et al.*, 2005) improving seed disperal and enhancing the diversity of plant species and consequently the stability of plant population and communities, (Couvreur *et al*, 2005). Endozoochory covers the consumption of fleshy fruits by frugivores and the consumption of seeds of grasses and herbs by herbivorous mammals (Pakeman *et al.,* 1998, Heinken *et al.,* 2001, Cosyns 2004, Cosyns *et al.,* 2005). Since many plant species cannot relied only on soil seed bank, germination success through the endozoochorus seed dispersal has a significant role on their survival of plant species and studies on possible seed dispersal and germination mechanisms are of key interest in the understanding of the colonization abilities of plants at the landscape scale (Verhagen *et al.,* 2001; Pywell *et al.,* 2002).

During last two decades, an increasing number of attempts have been undertaken to get reliable knowledge of which plants are dispersed zoochorously and which factors and plant traits direct zoochorous dispersal (Cosyns *et al.,* 2005). There are now many studies which have quantified such effects on a wide range of plants and habitats, such studies, are still very limited in semi-steppe rangelands (Jaroszewicz *et al.,* 2009). Steppe and semi-steppe rangeland systems are structurally diverse, exhibiting differences in woody plant canopy cover, stature, shrub functional form (evergreen vs. deciduous; broad-leaved vs. needle-leaved vs. succulent-leaved; shallow vs. deeply rooted), grass functional form[annual vs. perennial, C3 vs. C4 photosynthetic pathway] and spatial arrangement [random, regular, or clumped trees, bunch vs. rhizomatous grass] (Archer *et al.,* 2001).

The effectiveness of endozoochory and germination success of plant species after passage through the animal gut is a function of quantitative and qualitative traits. The quantity of dispersal depends on the amount of seeds ingested, animal type and livestock digestive system (Herrera and Jordano, 1981). This can happen either deliberately due to high palatability or accidentally when a herbivore consumes seeds along with palatable leaves or neighboring palatable plants (Janzen, 1984; Pakeman *et al.,* 2002). While the quality of dispersal depends on the percentage of undamaged seeds that are defecated. Germination may be enhanced by the softening of the coats during the digestive process, but destruction of seeds or inhibition of germination can also occur (Ramos *et al.,* 2006). Some seeds adapted for endozoochory require scarification, the abrasion or chemical degradation of the seed coat that may be required by some species in order for the seeds to germinate (Davise, 2007). Deposition of seeds with faecal material may provide nutrients that promote seedling establishment, but seed germination and seedling establishment could also be inhibited due to the toxicity and hydrophobic nature of dung (Ramos *et al.,* 2006). Edible vegetative plant parts and seed resistance to digestion (Pakeman *et al*., 2002) are hypothesized to be adaptations to endozoochory by large herbivores (Couvreur *et al.*, 2005). Seed traits such as: hard shell, size, length, shape and seed width are among the most important traits that available studies revealed their prelateship with endozochory. For example, it has been hypothesized that seeds of small size and thick skins are better able to pass through the digestive system (Cosyns *et al.,* 2005). Studies of the seed content in herbivore dung have often found that small seed size is correlated with presence in dung (Cosyns and Hoffmann 2005, Couvreur *et al.,* 2005). However, a higher survival rate during gut passage of small seeds is not the only possible explanation for this pattern. Smaller seed size having smaller seeds implies having more numerous seeds per plant individual due to an evolutionary trade-off between seed size and seed number produced per plant per year (Brunn and Poschold, 2006).

Additionally, Seed germination success after gut passage is not simply a function of the parent plant. Animal species have different effects on seed germination because of the differences in the oretical mean retention times of digestive products (Wallander *et al.,* 1995; Mueller *et al.,* 1998). Different intake of forage plant seeds, defecation frequencies, digestive systems and mean dung volumes, for example it has been indicated that most large herbivores have the strongest effect on the distribution and germination success of vegetable seeds .Existing studies dealing with the colonization of cattle faeces revealed high seedling emergence and high establishment rates of endozoochory dispersed diasporas compared to sheep and goat (Olson and Wallender, 2001).

Including different sized herbivore species, with different digestive physiology and morphology and by combing the results of a simulated feeding experiment and in a field study we aimed to [1] assess germination success of 39 native plant species after gut passage; [2] To explore the possible ecological correlates of the dung germinable seed content with seed traits [3] To compare dung seed content characteristics between different animal type, [4] to explore the possible correlate of dung seed content characteristics with vegetation in semi-steppe rangelands.

Materials and methods

Feeding experiment

39 native plant species were selected with a range of morphological characteristics within different plant life forms and considering their role in structure and composition of semi- steppe plant communities (Table 2). Seeds were either collected from the semi-steppe rangeland in Karsank during the summer of 2011 or bought from a commercial supplier. 50 air-dried seed of each plant species were chosen to measure seed weight, length, width and shape (Grime *et al.,* 1988). Three individuals of each animal species were kept under similar conditions in the stable separated from each other and were fed and freely access to water from 6 days before experiment. In the seventh day, a known amount of seeds was mixed with the previous food and offered to the animals. Then animals were carefully observed to ensure that the seeds were completely consumed. After seed feeding, all dung from each individual animal was collected regularly until 6 days and kept at 2–4 °C for 2 weeks. Several subsample were extracted from each animal dung and spread out on 0.4 m^2 trays kept for 6 month at greenhouse condition (temperature 20°C, ventilation, humidity 50% , 16 h light and 8 h dark, Watering, twice a day). The same number of seed for each plant species was dried and kept in 2-4°C for 2 weeks and then planted on the bare soil substrate to compare germination success of seed passed gut with control. Then, seedlings were identified and counted and simulatansouly were removed from trays. Moreover the date of seedling emergences was also recorded for each plant species. This continued for six months since the seedling emergence was abruptly decreased after three months. Six trays without seed addition were used to avoid possible germination from the potting soil substrate and contamination in the greenhouse.

Field study

The field study was conducted in the Karsanak region (32° 30' N, 50° 26' E, area 800ha), Chahar Mahaal-Bakhtiari province in Iran. This region is characterized by a transitional situation between arid and temperate climate, with hot summers and cool winters and 600 mm annual rainfall, mostly in winter and early spring. Differences in geological substrate and topography further contribute to diversity of vegetation types in the region. The natural vegetation in the study area presents itself as grassland-shrub land mosaics. Grazing is started from to by goat and sheep. Since the entire area is not grazed by cattle, it was removed from our field experiment. During summer 2009 and 2011, goats and sheep feces were collected by following the herds which graze different parts of the region. The samples were also collected in location where animals gathered to rest. To avoid possible seed contamination, deposited dung was collected leaving the lowermost part of the dung untouched. Because distinguish between sheep and goat faces were difficult, mixed sample of pallets were analyzed. The samples were aired dried and kept under same treatment as the greenhouse experiment. The standing vegetation was estimated in 60 plots (2×2m) that were randomly installed along 6, 200m transects in order to compare to the germination success of plant species.

Table 1. different germination index used for germination success of plant species.

Index	Germination value (GV)	Mean daily germination (MDG)	Peak value (PV)	Mean germination time (MGT)	Coefficient of the rate germination(CRG)	Mean germination rate (GR)
Formula	PV*MDG	Gn/final day	$((GC_{1max}+GC_{2max})/2)/((t_{1\,max}+t_{2\,max})/2)$	∑(t*n)/∑n	R*100	∑n/∑(t*n)
Unit	day^(-2)	day^(-1)	day^(-1)	day	_	day^(-1)

Data Analysis

Germination success was calculated using different index (Table1), each has different definition and application. Based on germination forms prepared for recorded daily germination, germination rate is means that the species that germinate to begin sooner than the other species will be higher germination rate (GR). If germination is associated with fewer days, increases coefficient of the rate germination (CRG) for each species. Mean germination time (MGT), there have Inverse relationship with coefficient of the rate germination. To calculate the peak value (the maximum germination value) or PV, first, the maximum number of seedling to days germination recorded in those days, was determined, then divided by the mean cumulative germination days which had the highest average of the day, the index was calculated (Table No. 1). Index germination value (GV) was calculated by multiplying the two parameters PV*MDG. The results of indicate T test (Paired samples t test) for animal treatment and control treatment showed that there are significant differences for all the parameters so that the germination rate, coefficient germination rate and the germination treatments for animal control is higher (Table 2).Greenhouse conditions may not be suitable for germination of some seeds, therefore the seeds did not germinate in any of the treatments (sheep, goat, cattle and control) were removed from the analysis among which seeds of plant species and analises were performed with the rest of the plant species (21 plant species were excluded from the analysis, and 18 plant species were compiled to analyzed).

Data were analyzed using software SPSS17 using Chi-square test was performed to distinguish between plant species and growth form. Using the Kruskal-Wallis test to compare between different cattle densities were performed for germination. To calculate the index of germination Paired samples t-test was used for analysis.

Result

Seeds pass through the digestive system of the plant species

Plant species are able to germinate in 3 treatments per animal and control treatment (Table 2), The results showed that the germination percentage equal to 82% of the treated cows *Festuca ovina*, 10/6% in the treated sheep and 14/6% for the treatment of goats. Between cows treated with control, there was no significant difference for any *F. ovina* ($p < 0/05$) and germination control equal to 34/ 6% (Fig. 1).

Table 2. Complete list of plant species studied in experiment.

Number	Scientific Name	Vegetative Form	Family	Number of seed in treatment	Number of seed germination in each treatment			
					Cow	Sheep	Goat	Control
1	*Festuca ovina L.*	perennial grasses	*Gramineae*	75	62	8	11	26
2	*Festuca arundinacea Schreb*	perennial grasses	*Gramineae*	112	0	0	0	12
3	*Agropyron elongatum (Host) .Beeauv.*	perennial grasses	*Gramineae*	66	16	2	3	24
4	*Agropyron tricophorom (Link) Richter.*	perennial grasses	*Gramineae*	70	0	0	0	8
5	*Agropyron cristatum (L.) Gaert.*	perennial grasses	*Gramineae*	55	0	0	0	5
6	*Dactylis glomerata L.*	perennial grasses	*Gramineae*	112	8	1	0	35
7	*Secale montanum Guss.*	perennial grasses	*Gramineae*	95	0	0	0	9
8	*Rumex ponticus E.H.L. Krause*	perennial forbs	*Polygonaceae*	428	86	2	2	0
9	*Rumex crispus L.*	perennial forbs	*Polygonaceae*	1400	65	14	11	395
10	*Rheum ribes L.*	perennial forbs	*Polygonaceae*	270	0	0	0	0
11	*Salvia officinalis L.*	perennial forbs	*Labiatae*	350	72	8	6	179
12	*Stachys spectabilis Choisy ex DC.*	perennial forbs	*Labiatae*	1450	0	0	0	0
13	*Ziziphora tenuir L.*	perennial forbs	*Labiatae*	1100	0	0	0	0
14	*Conium maculatum L.*	perennial forbs	*Umbelliferae*	1800	20	5	4	275
15	*Dorema Aucheri Boiss.*	perennial forbs	*Umbelliferae*	66	0	0	0	0
16	*Ferula gumosa Boiss.*	perennial forbs	*Umbelliferae*	134	0	0	0	0
17	*Ferula Assa-foetida L.*	perennial forbs	*Umbelliferae*	200	0	0	0	0
18	*Ferulago angulata (Schlecht.) Boiss.*	perennial forbs	*Umbelliferae*	120	0	0	0	0
19	*Cynara Scolymus*	perennial forbs	*Compositae*	200	10	2	2	23
20	*Silybum marianum (L.) Gaertn.*	perennial forbs	*Compositae*	170	28	4	3	140
21	*Artemisia aucheri Boiss.*	perennial forbs	*Compositae*	90	0	0	0	0
22	*Achillea sp*	perennial forbs	*Compositae*	126	0	0	0	0
23	*Artemisia sieberi Besser*	bush	*Compositae*	360	0	0	0	0
24	*Onopordon leptolepis DC.*	perennial forbs	*Compositae*	2000	0	0	0	0
25	*Eurotia ceratoides(L.) C. A. Mey.*	bush	*Chenopodiaceae*	112	0	0	0	0
26	*Atriplex leucoclada (Boiss.) Aellen*	perennial forbs	*Chenopodiaceae*	83	0	0	0	0
27	*Kochia.prostrata*	perennial forbs	*Chenopodiaceae*	150	0	0	0	0
28	*Plantago lanceolata L.*	perennial forbs	*Plantaginaceae*	65	26	4	1	21
29	*Vicia sativa L.*	perennial forbs	*Papilionaceae*	75	5	0	0	11
30	*Asragalus adsendence*	perennial forbs	*Papilionaceae*	120	0	0	0	0
31	*Trigonella elliptica Boiss.*	perennial forbs	*Papilionaceae*	200	0	0	0	23
32	*Nigella sativa L.*	perennial forbs	*Ranunculaceae*	200	0	0	0	6
33	*Fritillaria persica L.*	perennial forbs	*Ranunculaceae*	285	0	0	0	0
34	*Cerasus Mahaleb(L.) Miller*	shrub	*Rosaceae*	200	0	0	0	0
35	*Sanguisorba minor*	perennial forbs	*Rosaceae*	63	0	0	0	5
36	*Hypericum scabrum L.*	perennial forbs	*Hypericaceac*	1750	0	0	0	0
37	*Alyssum linifolium Steph. ex Willd.*	annuals forbs	*Cruciferae*	250	0	0	0	0
38	*Allium hirtifolium Boiss.*	perennial forbs	*Liliaceae*	200	0	0	0	0
39	*Ixiolirion tataricum(Pall.) Herb*	perennial forbs	*Amaryllidaceae*	1500	0	0	0	0

Table 3. List of plants entered in the analysis.

Plant species	Number of seed in each treatment	Number of seed germination in animal treatment			Number of seed germination in control treatment
		Cow	Sheep	Goat	
Festuca ovina L.	75	62	8	11	26
Agropyron elongatum (Host) P.Beeauv.	66	16	2	3	24
Dactylis glomerata L.	112	8	1	0	35
Rumex ponticus E.H.L. Krause	428	86	2	2	0
Rumex crispus L.	1400	65	14	11	395
Salvia officinalis L.	350	72	8	6	179
Conium maculatum L.	1800	20	5	4	275
Cynara Scolymus	200	10	2	2	23
Plantago lanceolata L.	65	26	4	1	21
Vicia sativa L.	75	5	0	0	11
Silybum marianum (L.) Gaertn.	170	28	4	3	140
Festuca arundinacea	112	0	0	0	12
Agropyron tricophorom	70	0	0	0	8
Agropyron cristatum	55	0	0	0	5
Secale montanum	95	0	0	0	9
Trigonella elliptica	200	0	0	0	23
Nigella sativa	200	0	0	0	6
Sanguisorba minor	63	0	0	0	5

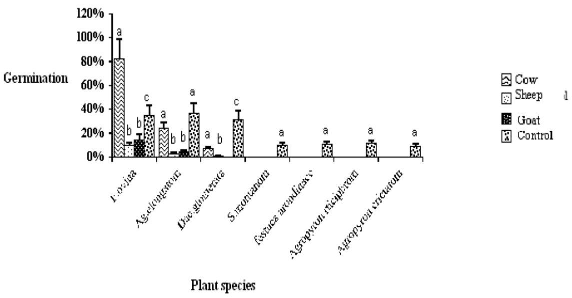

Fig. 1. Comparison of germination perennial grasses (cattle, sheep, goats, and control) at 5% significance level based on Chi-square test. Same letters indicate no significant difference.

For species *Agropyron elongatum*, there a significant difference between the treatment of cattle, sheep and goats, so that germination in treatment cow, are more of sheep and goats treatments, and the percentage of cattle treatment is equal to 24.2%. Comparison between sheep and goats treatments, there was no significant difference, and germination percentage, equal to 3% for sheep and goats with 4.5%. For *Ag. elongatum* species, no was significant difference between cow treatments and control treatment (p> 0/05), also is germination percentage for controls with 36.6%.The results showed that the germination of *Dactylis glometara*, between cattle treatment with sheep and goat treatments, there is a significant difference, so that germination in plants cattle treatment, is higher than the sheep and goats treatments. Germination for these species, respectively, in the treatment of cattle, sheep and goats with 7.1 of 0.89 percent and zero percent._For these species, the percentage of seed germination between treatments sheep with goat, there was no significant difference. For *D.glomerata* species, between treatments with control animals, there were significant differences. The results for *D.glometara* species indicate there are significant differences between animal treatment and control treatments, and germination percentage in control treatment, were higher than of animal treatments and equal to 31.2 percent. The results showed that the species in Table 2, could not germinate after passing through the digestive system.

Table 4. Chi-square values significant at the 5% level of significance between the treatments of animals. (ns: Not significant at the 5% level,*: Is significant at the 5% level).

Compare of animals treatment	perennial grasses	Chi-Square	perennial forbs	Chi-Square	annuals forbs	Chi-Square
Cow-sheep	0*	65.28	0*	194.68	0*	20.94
Cow-goat	0*	94.68	0*	217.69	0.017*	22.75
Sheep-goat	0.304 ns	0.497	0.126 ns	1.62	0.5	0.145
Cow-control	0.047 *	3.11	0*	373.21	0*	143.36
Sheep-control	0*	47.48	0*	807.06	0*	116.06
Goat-control	0*	71.37	0*	917.28	0*	180.74

The result of Chi-square test for *Rumex ponticus* species showed there was significant difference between cattle treatments with sheep and goats treatments, so that germination percentage for cow treated with 20.9%, and for both sheep and goat treatments with 0.46%. For *R. ponticus* species no green shoots in the control treatment (Fig. 2). The results showed for *Rumex crispus* species, there were significant difference between cow treatment with goat and sheep treatment, ($p < 0.05$).

Germination percentage for this species, in cattle treatment equal with 64/4 percent and for sheep and goats respectively 1 and 0.87 percent. The results of the comparison between animals treatment and control treatment for R. crispus species there was shown a significant difference.So that germination percentage in control treatment higher than animal treatments and equal with 1/28 percent. Test results for *Salvia officinalis* species showed, there were significant difference between cattle treatment with sheep and goat treatments, and germination percentage in cow treatment, higher than other animal treatments and with 20.57 percent. Also, there is no significant difference between the treatments sheep fed goats ($p>0.05$), and germination percentage for sheep treatment equal with 2.28% and for goat treatment equal whit 1.71%. The results of the comparison between animal treatments with control treatment, Showed, there was significant differences for *S. officinalis*. So that germination in control treatment is higher than animal treatments and equal with 1/51 percent.

Chi-square test results showed that for *Conium maculatum* species, between cattle treatment with goat and sheep treatments, there is a significant difference. So that germination in cow treatment is higher than two other animal treatment and equal with 1/11 percent, and for sheep and goats treatments, respectively equal with 0.27% and 0.22 percent. The results of the comparison between animal treatments with control treatment show there is significant difference for *Conium maculatum* specie. So that germination percentage in control treatment is higher than animal treatment and is equal with 15 percente. Results for *Cynara Scolymus* species showed, there was significant differences between cattle treatments with sheep and goat treatments. So that germination percent for cow treatment equal with 5 percent and germination percentage for goat and sheep in both the treatment 1 percent. Germination percent in control treatment higher than the animal treatments and equal with 11.5 percent. For species *Plantago lanceolata*, there was significant difference between cattle treatment with sheep and goat treatments. And germination in cow treatment, higher than other animal treatments and is equal to 40 percent. For this species, there was no significant difference between the treated sheep fed goats (Fig. 2).

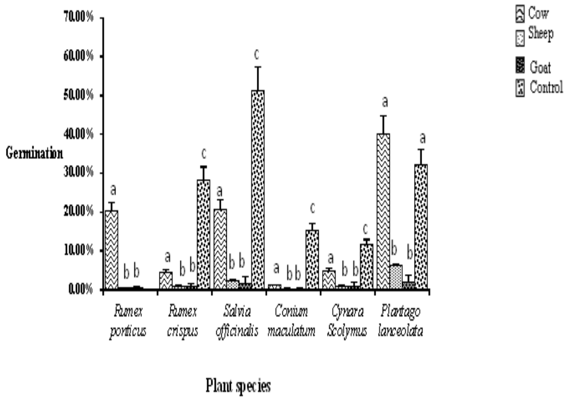

Fig. 2. Comparison of germination perennial forbs (cattle, sheep, goats, and control) at 5% significance level based on Chi-square test. Same letters indicate no significant difference.

The results showed for *Vicia sativa* species, there was significant differences between cow treatment and control treatment for germination. Germination in cow treatment equal with 6.61% and control treatment equal with 14.6 percent. The results showed for *Silybum marianum* species, there was a significant difference between the cattle treatment with sheep and goats treatments. Germination percent in cattle treatment higher than other animal treatments and equal with 4/16 percent. For these species, there was no significant difference between the sheep and goats treatments (Table 4 and Fig. 3).

The results of the germination for vegetative form

Perennial grasses

The results of Chi-square for the perennial grasses vegetative forms showed, between treatments of cattle, sheep, goats, and control treatments, there was a significant difference ($p < 0.05$). So that percentage of germination in control treatment, higher than the sheep and goats treatments and equal with 27%. This is while, germination in control treatment, began one month later animal treatments. The results showed there was no significant difference between sheep treatment with goat treatment, and sheep treatment germination is equal with 1/3% and for goat treatment equal with 4%. For Grass several, between cow treatment with control treatment there were significant difference, and germination percentage for cow treatment is equal with 24.7 percent (Fig. 4 and Table 4).

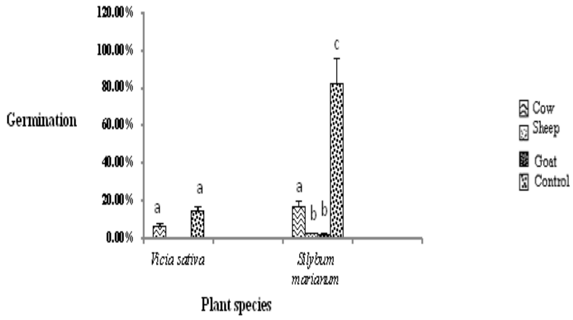

Fig. 3. Comparison of germination annuals forbs (cattle, sheep, goats, and control) at 5% significance level based on Chi-square test. Same letters indicate no significant difference.

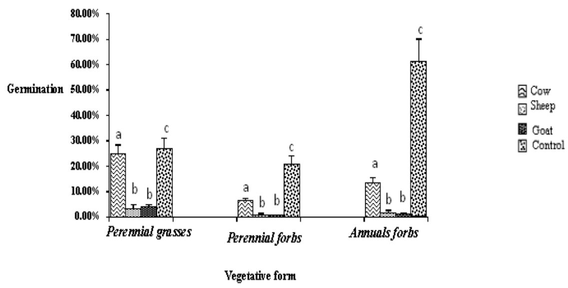

Fig. 4. Comparison of germination vegetative form (cattle, sheep, goats, and control) at 5% significance level based on Chi-square test. Same letters indicate no significant difference.

Perennial forbs

Results of Chi-square for perennial forbs showed there was significant difference between cattle treatment with sheep and goat treatments, so that germination percentage in cow treatment higher than two other treatments and is equal to 7 percent. Between sheep and goats treatments for perennial forbs, there was no significant difference, and germination percentage is for sheep treatment equal to 0.88 % and for goat treatment equal to 0.66%. The results showed that the germination percentage in control treatment for perennial forbs, there was significant difference with the animal treatments. In control treatment, germination percentage higher than animal treatments and equal with 21.04% germination in control treatment, but the start date germination in control treatment is later than the animal treatments.

Annuals forbs

Results of Chi-square for annuals forbs showed there was significant difference between cattle treatment with sheep and goat treatments, so that germination percentage in cow treatment higher than two other treatments and is equal to 13.4 percent, so that for sheep treatment germination percentage is equal with 1.65% and for goat treatment equal to 1.25%. The results showed that the germination percentage in control treatment for annuals forbs, there was significant difference with the animal treatments.

In control treatment, germination percentage higher than animal treatments and equal with 61% germination in control treatment.

3- Seeds properties and pass of the digestive system

According to studies, many features of the seed through the digestive system are affected. Scattering by endozoochory include: capture and eat plant seeds and being under the influence of digestive fluids during the passage through the digestive system. Endozoochory assessment, need to assess the viability of seeds in the digestive tract of livestock and other properties, Such as: hard shell, size, length, shape and seed width (the cosynse *et al.*, 2005). The results showed that in this experiment, 491 seeds, sprouted animal treatments. Plants sprouted are from different plant families such as: *Gramineae*, *Polygonaceae*, *Labiatae*, *Compositae*, *Plantaginaceae* and the *Papilionaceae*. Also, the number 68 seed, only able to germinate were in control treatment. Between the seeds of grasses family, Most of the species *Festuca ovina* seeds pass through the animal's digestive tract. *F.ovina* seed compared to *Ag*. *Cristatum* species and *D. glomerata* weight, length and width is less. Also in the family *Gramineae*, species *Festuca arundinaceae, Agropyron tricophorom*, *Agropyron cristatum* and *Secale montanom***,** germination was successful only in control treatment, and for these species in animal treatments, the buds did not grow, that probably due to the large size, is (Length, width and weight than other seeds of the same family). About two plant species *Rumex ponticus* and *Rumex crispus*, germination percentage between animal treatments, in cattle treatment is higher than, sheep and goats treatments. Also, the species Rumex ponticus compared with species *Rumex crispus,* weight, length and width is greater and the highest percentage of germination for this species, in cattle treatment and is equal with 20 percent. About two plant species *Rumex ponticus* and *Rumex crispus*, there was no clear relationship between seed characteristics and pass through the digestive system. Species S*alvia officinalis*, *Conium maculatom*, *Cynara scolymus*, *Silybum marianum* and *Plantago lanceolata*, germinated in all four treatments (3 animal treatments and one control treatment), that the percentage of germination for this species, in cattle treatment more than two other animals treatments, and germination percentage in control treatment is more than germination of each animal treatments. Species of *Trigonella elliptica, Nigella sativa* and *Sanguisorba minor* germination just for control treatment were able to germinate, that there was no clear relationship between seed characteristics and pass through the digestive system (Table 5).

Table 5. Characteristics of size, length and width of seed germination in animal and control treatments.

Plant family and species	Seed shape	Number of seeds in each treatment	Weight seed (gr)	Length* width(mm)	Cattle	Sheep	Goat	control
Gramineae								
Festuca ovina	Not Round	75	0.0007	3.2*0.74	*	*	*	*
Festuca arundinacea	Not Round	112	0.0017	7.1*1.9				*
Agropyron elongatum	Not Round	66	0.0016	4.1*0.92	*	*	*	*
Agropyron tricophorom	Not Round	70	0.004	9.2*1.8				*
Agropyron cristatum	Not Round	55	0.0013	5.3*1.01				*
Dactylis glomerata	Not Round	112	0.0012	3.02*0.86	*	*		*
Secale montanum	Not Round	95	0.013	7.5*1.7				*
Polygonaceae								
Rumex ponticus	gabled	428	0.034	5.1*4	*	*	*	
Rumex crispus	gabled	1400	0.002	2.4*1	*	*	*	*
Labiatae								
Salvia officinalis	Not Round	350	0.0055	2.6*2.2	*	*	*	*
Umbelliferae								
Conium maculatum	Round	1800	0.0046	3.7*1.5	*	*	*	*
Compositae								
Cynara Scolymus	Linear	200	0.034	7.1*0.1	*	*	*	*
Silybum marianum	Linear	170	0.015	6.3*3.05	*	*	*	*
Plantaginaceae								
Plantago lanceolata	Not Round	65	0.00013	1.4*0.71	*	*	*	*
Papilionaceae								
Trigonella elliptica	Not Round	200	0.01	3.7*2.7				*
Vicia sativa	Round	75	0.062	4.8*4.2	*			*
Ranunculaceae								
Nigella sativa	Not Round	200	0.0017	3.02*1.5				*
Rosaceae								
Sanguisorba minor	Not Round	63	0.0043	3.6*2.4				*

Results of simple linear regression on seed characteristics

Results of simple linear regression between seed germination and their properties showed that there was no significant relationship and morphological characteristics and seed size, seed pass through the digestive system does not affect (Table 6). About two plant species *Rumex ponticus* and *Rumex crispus*, germination percentage between animal treatments, for cattle treatment, is higher than two other treatments. Also, *Rumex ponticus* compare with *Rumex crispus* weight, length and width is greater, and the highest percentage of germination for this species, in cattle treatment is equal to 20 percent. About two plant species *Rumex ponticus* and *Rumex crispus*, there was no clear relationship between seed characteristics and pass through the digestive system.Species *Salvia officinalis, Conium maculatom, Cynara scolymus, Silybum marianum* and *Plantago lanceolata* germinated in all four treatments (3 animal treatments and one control treatment), that the percentage of germination for this species, in cattle treatment more than two other animals treatments and germination percentage in control treatment is more than germination of each animal treatments. Species of *Trigonella elliptica, Nigella sativa* and *Sanguisorba minor* germination was successful only in control treatment, and there was no clear relationship between seed characteristics and pass through the digestive system._Results of simple linear regression for perennial grasses indicate there was no relationship between seed characteristics and pass through the digestive system.

Table 6. significant coefficients simple linear regression between germination percentage of vegetative forms and animal treatments. (ns: Not significant at the 5% level).

Vegetative forms and Animal treatment	Length	R Square	width	R Square	Weight	R Square	Seed shape	R Square
perennial grasses								
Cattle	0.073 ns	0.507	0.174 ns	0.370	0.451 ns	0.119	0.64 ns	0.046
Sheep	0.074	0.503	0.15	0.367	0.452	0.117	0.65	0.044
Goat	0.086	0.476	0.181	0.326	0.481	0.104	0.086	0.476
perennial forbs								
Cattle	0.297 ns	0.197	0.921 ns	0.002	0.435 ns	0.105	0.932 ns	0.001
Sheep	0.173	0.284	0.305	0.173	0.571	0.057	0.581	0.054
Goat	0.521	0.072	0.3	0.177	0.921	0.002	0.988	0
annuals forbs								
Cattle	0.103 ns	0.974	0.684 ns	0.277	0.937 ns	0.010	0.874 ns	0.039
Sheep	0.356	0.706	0.946	0.007	0.801	0.094	0.612	0.328
Goat	0.356	0.706	0.946	0.007	0.801	0.094	0.612	0.328

Results of logarithmic regression on seed characteristics

Logarithmic regression analysis showed, that for perennial grasses between length and germination percentage were significantly related in all animal treatments and with increasing length seed in animal treatments reduced germination (Table 7). The results of this test for perennial forbs showed that, for the treatment of cattle, there was a significant correlation between germination and seed weight and with increasing seed weight, seed germination and through seed of the animal digestive system were reduced. The results of this test for perennial forbs in sheep treatment showed that, between germination percentage, seed weight and seed length there is a direct relationship and with increasing seed length and seed weight_seeds pass through the digestive system successfully reduced. The results of this test for annuals forbs showed that, there was no relationship between seed characteristics and pass through the digestive system.

Table 7. significant coefficients logarithmic regression between germination percentage of vegetative forms and animal treatments. (ns: Not significant at the 5% level, *: Is significant at the 5% level).

Vegetative forms and Animal treatment	Length	R Square	width	R Square	Weight	R Square	Seed shape	R Square
perennial grasses								
Cattle	0.029*	0.496	0.106 ns	0.437	0.197 ns	0.307	0.619 ns	0.053
Sheep	0.039*	0.608	0.139	0.382	0.244	0.278	0.617	0.054
Goat	0.036*	0.608	0.135	0.388	0.223	0.279	0.680	0.037
perennial forbs								
Cattle	0.125 ns	0.345	0.98 ns	0	0.039 *	0.535	0.91	0.003
Sheep	0.038 *	0.540	0.53	0.066	0.021 *	0.618	0.56	0.059
Goat	0.27	0.195	0.36	0.138	0.39	0.123	0.99	0
annuals forbs								
Cattle	0.16 ns	0.933	0.61 ns	0.344	0.66ns	0.258	0.88 ns	0.031
Sheep	0.42	0.612	0.86	0.046	0.92	0.015	0.62	0.308
Goat	0.42	0.612	0.86	0.046	0.92	0.015	0.62	0.308

Germinated density in animal treatment and calculation germination indices in animal and control treatments

Seedling density in animal treatments

The results of Kruskal-Wallis test between two treatment density of seed germination in cattle treatment with sheep treatment for all species showed that, between the two treatments at the 5% level, there are significant differences ($p < 0.05$) so that, average seedling density for cow in all the iterations equal With 3/30 sprouts_and for sheep in all iterations, is equal to germinated seedlings. So that, the results of Kruskal-Wallis test between two treatment density of seed germination in cattle treatment with goat treatment showed that, there was significant difference, so that, average seedling density for goat treatment is equal to 13.3 germinated seedlings all iterations. Also, among goat with sheep treatments, there was no significant difference in the density of germination (Table8).

Table 8. Seedling densities in different treatments based on Kruskal-Wallis. (ns: Not significant at the 5% level , *: Is significant at the 5% level).

Animal treatment	density	Test statistic (chi-Square)
Cow-Sheep	0.036*	4.68
Cow-Goat	0.026*	4.97
Sheep-Goat	0.51 ns	0.429

Table 9. Germinated species list in years 2009 and 2011.

number	species	Number of seedling in year 2011	Number of seedling in year 2009
1	*Achillea wilhelmsii*	43	4
2	*Amaranthus albus*	1	1
3	*Bromus tomentellus*	2	0
4	*Carex stenophylla*	29	17
5	*Chenopodiom albom*	5	0
6	*Crepis kotschyana*	0	4
7	*Dactylis glomerata*	2	0
8	*Discornia sofia*	6	0
9	*Euphorbia sp*	0	8
10	*Festuca arondinace*	10	0
11	*Hordeum bulbosum*	2	1
12	*Plantago lanceolata*	2	0
13	*Sisymbrium irio*	1	0
14	*Ranunculus edulis*	13	18
15	*Taraxacum montanum*	1	1
16	*Trifolium ripens*	0	1

The result of field study conducted in the Karsanak region

Regression results of the relationship between the percentage of area covered by endozoochory Karsanak in 2009 showed no significant relationship (y= 4.044-1.22x, sig= 0.042) also in 2011 there was also a significant relationship between percent cover and endozoochory (y=7.86-2.37x, sig= 0.053).Of the 200 species found in the region, with different growth forms, only 16 plant species were able to germinate and there is no significant relationship between the percentage of area covered germination in years 2009 and 2011 (Table 9)._T-test results of the plant species for the years 2009 and 2011 there was no significant difference (Z=-1.51, sig= 0.131). Some species in year 2009 was able to germinate more higher than in year 2011, and conversely,_also some species only in 2009 or only in 2011 were capable of germination. the results of T-test in level form of vegetative between cover and endozoochory in years 2009 and 2011 showed that, there is not significant relationship (for year 2009:Z= -1.46 , sig= 0.144 & for year 2011: Z= -1.82 , sig= 0.068).

Discussion

The aims of this study passed a few several sample of pasture plants seed and its effect on seed germination by treatment animals (cattle, sheep, goat), was performed. According to App. 1, of the 39 plant species studied that in this experiment, only 11 plants species including: perennial grasses, perennial forbs and annuals forbs (Table 3), were able to germinate.

Seeds pass through the digestive system, causing poor germination of many plant species studied that is in agreement with results of other experiments by feeding different animal species such as: cattle, sheep, goats, rabbits, horses and donkey (Özer, 1979). Research results showed that all the plant species can not germinate after passing through the digestive system due to seed characteristics such as: Seed size and its characteristics animals such as chewing and Jaw system and but small seeds, most are able to pass through the digestive system of livestock (Bruun and Poschlod 2006). Also, the results showed that endozoochory, there has no effected to eliminate dormancy of Plants such as: *Dorema Aucheri*، *Ferula gumosa*، *Ferula Assa-foetida, Ferulago angulata, Artemisia aucheri, Artemisia sieberi, Achillea, Artemisia aucheri, Artemisia sieberi, Eurotia ceratoides* and *Kochia prostrata*.

The results of these experiments for different vegetative forms showed that germination percentage in cow treatment (between all animal treatments), is highest percentage of germination. So, in goat and sheep Treatments, is very low germination percentage. However, germination date in all animal treatments, were recorded faster than in control treatment, because there is food in the stool. This result is may be the result of complex interactions of plant and animal species characterized. For example, the intensity of chewing and digestion food, both of which are different in animal treatments and can be different acts (Simao Neto *et al.*, 1987; Staniforth and Cavers 1977).

Similar findings by researchers in this field, confirm the results of this experiment, So that the seeds in the intestines of sheep, less likely to are germinate than other animal species such as: cattle, horses and donkeys (Simao Neto *et al.* 1987; Shayo and Udén 1998). The researchers showed that, chewing less and swallowing too by cattle and large herbivores, cause is less damage to the seed, but in the case of sheep and goats, is high chewing and damage to seeds and finally, there is an inverse relationship between the animal size and biting animal severity (Lutman *et al.* 2003), that the researchers results in this study, is correct for the plant species: *Festuca ovina, Agropyron elongatum, Dactylis glomerata, Rumex ponticus, Rumex ponticus, Salvia officinalis, Conium maculatum, Cynara Scolymus, Plantago lanceolata* and *Vicia sativa*.

There are many factors in the success of seed to pass through the digestive tract of animal. For example, (Bruun and Poschlod, 2006), features that are associated with endozoochory related to seed traits, characteristics, and characteristics of seed-producing plants, native plants seeds of division. Also (Bruun and Fritzbøger, 2002; Heinken *et al.,* 2002), believe there was the negative relationship between seed size with endozoochory and positive relationship between seed number with endozoochory. (Leishman, 2001) is realized the positive relationship between abundance and population density of plant endozoochory. In a study by (Bakker and Olff, 2003), in the nutrient rich plains between cattle and rabbits, in order to examine different effects of herbivores on scattered plant seeds were in their feces, large herbivores, due to heavy use of plants and seeds, germination density in feces, is higher than the rabbit feces, Also, the researchers also found that despite high germination density in feces of cattle, large herbivores cannot travel great distances, however, small herbivores, because of the traverse greater distance seeds in their stools the seeds are broadcast at a great distance. The scientists also examined in order to investigate density of Germination in feces of herbivores (rabbits and cattle), examples of pasture seeds plants without passing through the digestive system in the stool of these herbivores were cultured. They found that the start of sprouting in the feces of cattle and rabbits than starts the germination of seeds that were planted in the soil is much higher. Also, begin to germinate seeds in the feces of cattle is higher than of rabbit feces that due to the high material such as phosphorus and potassium in cow feces. This scientist stated that small ruminants like rabbit, through the Burrow in soil and relocation the seeds beneath the soil, improves seed germination.

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