

Effects of phenological stages on yield and quality traits of three halophyte plant species as forage sources and combating desertification in Khuzestan province, Iran

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Abstract:

Halophytes have good potential for forage production and combating desertification in saline soils. The present research was aimed to investigate the effects of phenological stages (Vegetative, Flowering & Ripening) on the yield and quality of three halophytes, *Atriplex leuoclada*, *Suaeda fruticosa*, and *Seidlitzia rosmarinus*, in warm steppe rangelands, Khuzestan province, Iran. Data were collected in 2015 – 2016 for Dry Matter (DM) yield. Quality traits included Crude Protein (CP), Dry Matter Digestibility (DMD), Water-Soluble Carbohydrates (WSC), Acid Detergent Fiber (ADF), Crude Fiber (CF), Neutral Detergent Fiber (NDF), and total ash. Data were statistically analyzed using SAS9 software and means were compared using LSD 5% method. Results showed a significant difference between species, phenological stages, and their interaction for all the traits. *S. fruticosa* and *A. leuoclada* with average values of 14.45 and 12.88 t/ha had higher and lower production, respectively. In all species, with some exceptions, the DM yield, WSC, ADF, CF, and NDF increased, and CP, DMD, and ash decreased with the development stages. For *S. rosmarinus*, the lower and higher DM yield with values of 8.11 and 16.90 t/ha were obtained in the vegetative and flowering stages, respectively. The highest and lowest CP in *S. fruticosa* (17.73 and 13.31%), *S. rosmarinus* (13.05 and 9.62%), and *A. leuoclada* (10.19 and 5.29%) were obtained in flowering and seed ripening stages, respectively. In *S. rosmarinus*, DMD decreased in the flowering stage compared to the vegetative stage (from 70% to 65%), but its value increased again in the seed production stage and reached to 90%. However, in *Suaeda* and *Atriplex*, there was no significant variation at different growth stages. Our results clearly showed that *Suaeda fruticosa* having a higher DM yield in the flowering stage (16.20 t/ha) coupled with higher CP (18.04%) ($p < 0.5$) recommended for cultivation as forage sources and combating desertification in Khuzestan province, Iran.

Keywords: Nutritional value; Halophytes; Warm steppe rangelands; Khuzestan

Introduction

The saline land area in Khuzestan province is reported to be approximately 1.493 million hectares in 2002 Raufi (2010). Halophytes are important species of flora in Khuzestan province that grow well in large parts of saline lands, some of which are grazed by livestock, especially sheep and

camels, and also use for desertification programs. Knowledge of the nutritional value of different species that make up the rangeland forage at various times is essential for managing exploitation and achieving production goals (Behnamfar et al., 2018).

Around 349,000 ha of the Khuzestan plain area is the dust

source. Observations showed these areas do not have much plant species richness since they are located on a flat plain and covered with relatively fine-grained sediments or sand dunes with no geographical slopes. However, soil properties change and human activities cause vegetation diversity in different parts of this plain. The vegetation of the region includes four vegetation types: wetland plants, hydro-halophytes, xero-halophytes, and psammophytes. Plants such as *Tamarix passerinoides*, *Lycium depressum*, *Seidlitzia rosmarinus*, and *Atriplex leucoclada* are good adapted to the climatic and soil conditions of the region (Dinarvand et al., 2018).

Seidlitzia rosmarinus is distributed in Isfahan, Fars, Kerman, Khuzestan, Bushehr, Hormozgan, Sistan and Baluchestan, Khorasan, Semnan, Tehran, Qom, Markazi and Yazd provinces. This plant is globally found in Palestine, Jordan, Syria, Iraq, Saudi Arabia, Kuwait, Bahrain, Oman, UAE, Qatar, Afghanistan, and Central Asia (Mozaffarian, 2005). The results published by Maybodi and Zare (2009) showed that the habitat of *S. rosmarinus* is distributed over an area of 600,000 ha located in the deserts of Yazd province, Iran. The high groundwater level and soil salinity are two prominent features of lands suitable for the distribution of this species. *S. rosmarinus* flowers in early autumn, and seed maturity occurs in early December. High seed production and a high germination rate are the positive characteristics of its reproduction. In addition to the role of soil conservation, *S. rosmarinus* habitats located in desert areas can produce suitable forage. This plant is well grazed by camels, and its preference value in open grazing is in second place, followed by *Haloxylon*. *Atriplex leucoclada* is one of the plants adapted to desert areas, which is native to arid and semi-arid rangelands of Iran and has been widely cultivated due to its high resistance to harsh environmental conditions (Ariapour, 2012). This species grows in saline, arid, and semi-arid areas under different environmental conditions (Ardakani, 2001) and is also adapted to cold climate (Masoumi, 2006). It grows in almost flat saline lands with relatively high groundwater, arable land margins, and degraded lands in the Irano-Turanian and Khalij-e-Omani regions. Due to the quick and easy establishment, it can be used in capacity building of saline areas and degraded agricultural lands and thus provide part of the livestock feeding requirement in autumn and winter by producing suitable forage (Moghimi, 2005).

Ball et al. (2001) determined some factors affecting forage quality, and the most important ones were plant genetic, growing season, phenological stages, leaf-to-stem ratio, DM yield, Crude Protein (CP), Crude Fiber (CF), Dry Matter Digestibility (DMD), Total Digestible Nutrients (TDN), and soil nutrient status. In the study of Heady and Pill (1979), CP, DMD, and CF were introduced as the factors affecting the forage nutritional value.

Linn and Martin (1989) divided the factors affecting forage quality and digestibility into two groups: cell wall components (ADL, ADF, NDF, and Pectin) and intracellular contents (starch, fats, soluble proteins, non-protein nitrogen and carbohydrates), among which lignin with zero to 20% had the lowest and intracellular contents with 95 to

100% had the highest digestibility.

Behnamfar et al. (2018) determined the chemical composition and nutritional value of six rangeland species in the sandy area of Khuzestan province including *Stipagrostis plumose*, *Pennisetum divisum*, *Panicum antidotale*, *Asthenatherum forsskalii*, *Convolvulus oxyphyllus*, and *Medicago laciniata* in two phenological stages and announced that there was a significant difference between species in terms of forage quality. They stated that the growth stage highly affects forage quality and nutritional value because by plant growth, the forage DMD in all species decreases. Since forage quality and nutritional values of plants are positively related to CP and DMD and inversely related to ADF and CF, it can be concluded that forage quality is superior in the vegetative stage because the CP and DMD in all species in the vegetative stage are higher than the reproductive stage; moreover, the CF and ADF increased with the development of phenological stages.

Ahmadi et al. (2016) examined the variations in chemical composition and nutritional value of four halophytes, *Atriplex verrucifera*, *Haloxylon persicum*, *Halocnemum strobilaceum*, and *Nitraria schoberi*, in the rangelands of Arak Miqan desert at three phenological growth stages. Their results showed that the forage quality of the four species differed and was affected by phenological stages in each plant species. For most species, the CP decreases, and the NDF and ADF content increases as phenological stages advance. The lowest CP was obtained in *Nitraria schoberi* in the vegetative stage and *Haloxylon persicum* in the seed reopening stage.

Elahi et al. (2014) examined the nutritional value of *Salsola vermiculata* in two vegetative growth and flowering stages in Sistan, Iran. They found a significant difference between the phenological stages for chemical compositions. CP value ranged between 6.05 and 13.03%, ADF content between 16.13 and 36.54%, and NDF content between 30.41 and 47.90%.

Pasandi et al. (2017) studied the chemical composition and digestibility of five range species including *Salsola turcomanica*, *Plantago coronopus*, *Halostachys caspica*, *Halocnemum strobilaceum*, and *Frankenia hirsute*, in saline and alkaline rangelands of Golestan province, Iran in two phenological stages. Their results showed that phenological stages and species had a significant effect on forage quality indices, and by plant maturity, the content of CP, DMD, organic matter, digestible energy, and metabolizable energy decreased while ADF and lignin increased. They concluded that *S. turcomanica* had higher forage quality, and the forage quality of all species in the vegetative stage was much higher than the seed maturity stage.

Abarsaji et al. (2008) determined the forage quality of *Frankenia hirsute* in the saline and alkaline region of Inchebrun in Golestan province, Iran at three phenological stages. Their results showed that the forage quality of this species in the vegetative growth stage was higher than in the flowering and seed ripening stages. The CP content with an average value of 8.73% in the vegetative growth stage was higher than flowering and seed ripening stages, with values of 6.91% and 5.99%, respectively. They suggested this

species for forage production programs.

Adnani et al. (2018) investigated the effect of three phenological stages on the forage quality of three halophytes species (*Nitraria schoberi*, *Halocnemum strobilaceum*, and *Suaeda aegyptiaca*) in the marginal rangelands of the Arak Miqan desert in Iran. The results showed a significant effect of species and phenological stages on forage quality ($p < 0.01$). The highest and lowest forage quality was obtained at the vegetative and Seed ripening stage, respectively. They recommended *Halocnemum strobilaceum* in terms of its nutritional value as a new source of forage in saline lands.

Menke and Steingass (1988) determined the DMD and OMD of *Salsola vermicolata* using the gas production method, and they found significant differences in chemical composition between phenological stages. As the growth stage progressed, the amount of CP, DMD, and ME decreased, and cell fiber content increased. The highest DMD and ME were related to the vegetative stage of *Salsola vermicolata* compared to *Suaeda fruticosa*. They recommended using this plant in the vegetative stage for livestock feeding.

Nejad et al. (2012) examined the chemical compounds (OMD, ash, CP, crude fat, NDF, and ADF) of five halophytes dominant species in the Sistan region, Iran, including *Salsola griffithii*, *Chenopodium album*, *Aeluropus lagopoides*, *Alopecurus textillis* and *Cardaria draba*. Their results showed significant differences between the studied species in terms of chemical composition. The CP, NDF, and ADF content varied between 5.93 to 14.73%, 34.30 to 66.82%, and 13.64 to 41.46%, respectively. Ash content in *Salsola griffithii* was very high (46.87%) and in other species, it ranged between 12.63 to 16.18%. The lowest and highest OMD, with values of 66.13 to 76.24% were related to *Salsola griffithii* and *Cardaria draba*, respectively. The lowest amount of DMD, with a value of 40.51%, was obtained in *Salsola Griffith*. Laudadio et al. (2009) in a study in southern Tunisia found an inverse relationship between the CP

and WSC in halophyte plants.

In another study, the forage quality of *Calligonum comosum* was investigated in three phenological stages in the desert pastures of Kashan, Iran and the forage quality indices including Crude Protein (CP), Acid Detergent Fiber (ADF), Crude Fiber (CF), Dry Matter Digestibility (DMD), Total Energy (TE) and Metabolic Energy (ME) were determined. There was a significant variation in phenological stages for many quality traits ($p < 0.05$). Higher and lower values of CP, DMD, TE and ME were obtained in vegetative growth and seed maturity stages, respectively (Bidgoli, 2018). In the investigations of Zare et al. (2019) on the forage Quality of *Salsola yazdiana* and *S. tomentosa* in different growth stages in the Saline Desert of Yazd Province, Iran, significant differences were found for all quality traits ($p < 0.05$). This study aimed to investigate the effects of phenological stages (Vegetative, Flowering & Ripening) on the yield and forage quality of three halophytes, *Atriplex leuococlada*, *Suaeda fruticosa*, and *Seidlitzia rosmarinus* as forage sources and combating desertification in Khuzestan province, Iran.

Materials and methods

Initially, the key areas of three halophyte habitat were identified. Three salt-tolerant species were selected to determine their chemical composition and nutritional value. The study species *Atriplex leuococlada*, *Suaeda fruticosa*, and *Seidlitzia rosmarinus* were sampled at three phenological stages (vegetative growth, flowering, and seed production) in two consecutive years (Table 1). These three halophytes were selected since they constitute a vegetation type on saline rangelands of Khuzestan province and well grazed by livestock.

The plant samples were shade dried and then, placed at an oven at 70 °C for 48 hours. Due to the woody tissue of the study plants, the samples were ground in two stages (industrial mill and laboratory mill). Afterward, nutritional value

Table 1. Sampling date and location of the study species in different growth stages in two years.

Row	Species name	Phenological stage	Location	Date of collection
1	<i>Atriplex leuococlada</i>	Vegetative	Hamidia	October 14, 2015
2	<i>Atriplex leuococlada</i>	Vegetative	Hamidia	October 30, 2016
3	<i>Atriplex leuococlada</i>	Flowering	Hamidia	November 4, 2015
4	<i>Atriplex leuococlada</i>	Flowering	Hamidia	November 14, 2016
5	<i>Atriplex leuococlada</i>	Ripening	Hamidia	December 27, 2015
6	<i>Atriplex leuococlada</i>	Ripening	Hamidia	December 7, 2016
7	<i>Seidlitzia rosmarinus</i>	Vegetative	Khorramshahr-Jafir	October 13, 2015
8	<i>Seidlitzia rosmarinus</i>	Vegetative	Khorramshahr-Jafir	October 30, 2016
9	<i>Seidlitzia rosmarinus</i>	Flowering	Khorramshahr-Jafir	November 3, 2015
10	<i>Seidlitzia rosmarinus</i>	Flowering	Khorramshahr-Jafir	November 14, 2016
11	<i>Seidlitzia rosmarinus</i>	Ripening	Khorramshahr-Jafir	December 26, 2015
12	<i>Seidlitzia rosmarinus</i>	Ripening	Khorramshahr-Jafir	December 7, 2016
13	<i>Suaeda fruticosa</i>	Vegetative	Khorramshahr-Jafir	October 13, 2015
14	<i>Suaeda fruticosa</i>	Vegetative	Khorramshahr-Jafir	October 30, 2016
15	<i>Suaeda fruticosa</i>	Flowering	Khorramshahr-Jafir	November 3, 2015
16	<i>Suaeda fruticosa</i>	Flowering	Khorramshahr-Jafir	November 14, 2016
17	<i>Suaeda fruticosa</i>	Ripening	Khorramshahr-Jafir	December 26, 2016
18	<i>Suaeda fruticosa</i>	Ripening	Khorramshahr-Jafir	December 7, 2016

Table 2. Results of analysis of variance (MS) for the effect species, phenological traits and their interaction on DM yield and quality traits.

SOV	DF	MS							
		DM yield	CP	DMD	WSC	ADF	CF	NDF	Ash
Species (S)	2	0.15**	3494**	3050.6**	603.8**	1825.6**	1091.1**	2415.3**	52.48**
Stage (P)	2	1.11**	85.6**	298.9**	23.46*	731.2**	19.13**	257.21**	3.27*
S × P	4	0.26**	5.61**	267.9**	8.75	48.1**	38.08**	17.40**	0.29
Year (Y)	1	0.02	1.81	141.4**	142.56**	13.83	0.122	0.680	0.003
S × Y	2	0.025*	2.03	59.2*	93.52**	17.2**	5.93	25.06**	2.31
P × Y	2	0.01	0.26	33.47	5.95	1.87	6.00	6.217	8.43**
P × Y × S	4	0.035	0.79	137.5**	5.10	1.64	6.93	5.49	1.97
Error	24	0.025	0.83	17.21	5.47	1.86	4.65	2.14	0.91
CV		9.41	7.72	6.87	13.31	3.60	5.29	3.32	14.92

*, **= Significant at 5 and 1% probability levels, respectively.

parameters (CP, CF, ADF, NDF, DMD, WSC, and ash) were measured using NIR (Near Infrared Spectroscopy) model INFRAMATIC 8620. Details of the laboratory measurements of quality traits and calibrations of NIR were given by Jafari et al. (2003).

The measurements were performed in three phenological stages (vegetative, flowering, and seed production or maturity) in a split-plot design based on randomized complete design. Data were analyzed using SAS₃ software and the means were compared by the LSD 5% method.

Results

The results of ANOVA and means comparison showed no significant differences between species; however, *Suaeda fruticosa* and *Atriplex leuoclada*, with average values of 14.45 and 12.88 t/ha, had the highest and lowest forage pro-

duction, respectively (Tables 2 and 3). In all three species, the vegetative stage had the lowest DM yield. There was no significant difference between growth stages of species except to *Seidlitzia rosmarinus*. In latter species, the lower and higher DM yield with average values of 8.11 and 16.90 t/ha was obtained in the vegetative and flowering stages, respectively. It was expected the higher DM yield could be obtained in seed ripening stage. However, in all three examined species, during the seed production stage, due to the occurrence of cold (in late autumn), a part of the plant leaves fell causing a decrease in DM yield in this stage (Table 4).

For CP, results showed a decreasing trend with the development of growth stages in studied species. In *Suaeda fruticosa*, the highest and lowest CP, with average values of 18.04 and 13.31%, was obtained in the flowering and seed ripening stages, respectively. It was followed by *Sei-*

Table 3. The means of DM yield and quality traits averaged over both years and phenological stages in three forage species.

Species name	DM yield (t/ha)	CP (%)	DMD (%)	WSC (%)	ADF (%)	CF (%)	NDF (%)	Ash (%)
<i>Seidlitzia rosmarinus</i>	13.88 ^{ab}	11.55 ^b	75.36 ^a	23.89 ^a	26.28 ^c	31.82 ^b	30.67 ^b	7.93 ^a
<i>Suaeda fruticosa</i>	14.45 ^a	16.36 ^a	53.87 ^b	12.50 ^c	44.48 ^a	44.50 ^a	50.42 ^a	6.73 ^a
<i>Atriplex leuoclada</i>	12.88 ^b	7.55 ^c	51.82 ^b	16.34 ^b	34.12 ^b	45.98 ^a	51.03 ^a	4.70 ^b

The means of the column with the same letter are not significantly different ($p < 0.05$) by LSD method.

Table 4. The means of DM yield and quality traits average over two years for each phenological stages in three forage species.

Species name	Growth Stage	DM yield (t/ha)	CP (%)	DMD (%)	WSC (%)	ADF (%)	CF (%)	NDF (%)	Ash (%)
<i>Seidlitzia rosmarinus</i>	Vegetative	8.11 ^c	13.05 ^b	71.41 ^b	23.57 ^b	30.29 ^b	33.63 ^c	35.32 ^c	7.98 ^a
	Flowering	16.90 ^a	11.98 ^b	66.12 ^c	21.65 ^b	27.08 ^c	30.90 ^c	30.87 ^{cd}	8.37 ^a
	Ripening	14.75 ^b	9.62 ^c	88.55 ^a	26.44 ^a	21.47 ^c	30.92 ^c	25.83 ^d	7.45 ^a
<i>Suaeda fruticosa</i>	Vegetative	13.05 ^b	17.73 ^a	52.89 ^d	12.14 ^d	46.29 ^a	43.12 ^b	53.48 ^a	7.11 ^a
	Flowering	16.20 ^a	18.04 ^a	54.93 ^d	11.79 ^d	45.79 ^a	45.42 ^b	52.70 ^a	6.80 ^a
	Ripening	14.10 ^b	13.31 ^b	53.79 ^d	13.59 ^d	41.38 ^a	44.97 ^b	45.08 ^b	6.29 ^a
<i>Atriplex leuoclada</i>	Vegetative	11.85 ^b	10.19 ^b	51.13 ^d	16.72 ^c	33.72 ^b	43.99 ^b	52.84 ^a	5.46 ^b
	Flowering	14.05 ^b	7.17 ^{cd}	51.55 ^d	16.03 ^c	33.97 ^b	43.99 ^b	51.60 ^a	4.62 ^b
	Ripening	12.75 ^b	5.29 ^d	52.78 ^d	16.28 ^c	34.66 ^b	49.97 ^a	48.67 ^{ab}	4.03 ^b

The means of the column with the same letter are not significantly different ($p < 0.05$) by LSD method.

dlitzia rosmarinus with 13, 12, and 9% CP, and *Atriplex leuoclada*, with 10, 7, and 5% in the vegetative, flowering, and seeding stages, respectively. In *Seidlitzia rosmarinus*, DMD decreased in the flowering stage compared to the vegetative stage (from 70% to 65%) but increased again in the seed production stage and reached up to 90%. However, in *Suaeda* and *Atriplex*, changes in this trait at different growth stages did not differ significantly, and it was about 50% in both species (Figure 1).

Discussion and conclusion

Due to the almost similar forage production of all three species studied, the combined uses of *Seidlitzia rosmarinus*, *Suaeda fruticosa*, and *Atriplex leuoclada* are essential for

species diversity and production sustainability in vegetation reclamation of desertification programs. According to the forage quality indices, the CP of all studied species showed a decreasing trend with the development of growth stages. This result is consistent with the findings of Akbarian and Elahi (2015) studies in determining the forage quality of *Salsola vermicolata* and *Suaeda fruticosa* in the Sistan region, Iran at different phenological stages. A similar result was also reported in literature (Abarsaji et al., 2008; Arzani et al., 2013; Kamalak et al., 2005a; Kamalak et al., 2005b; Karabulut et al., 2006). *Suaeda fruticosa*, with about 18% CP in both vegetative and flowering stages, 13% CP in the seed production stage, and 50% DMD was considered a valuable species. Filekesh (1999) investigated

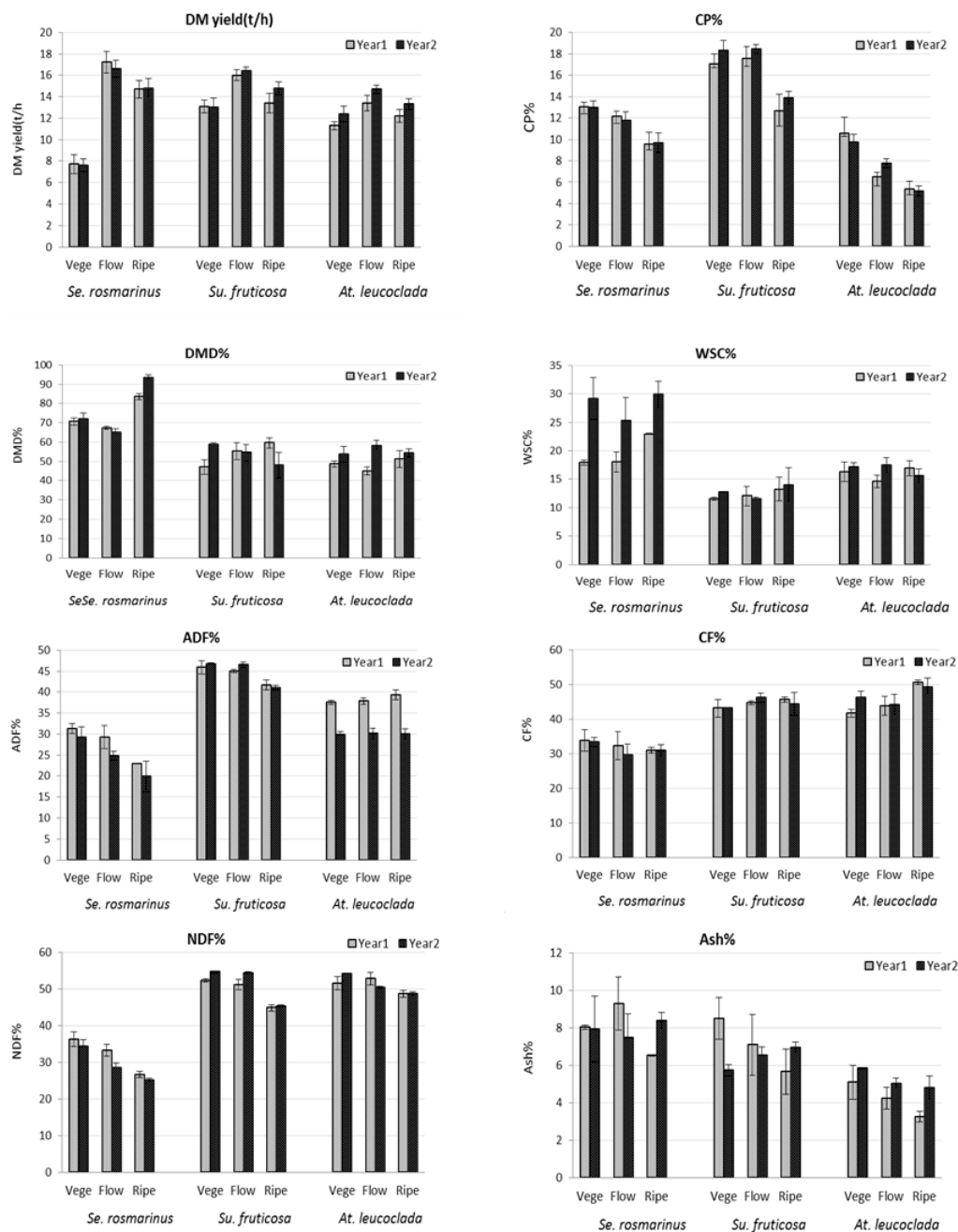


Figure 1. The Mean \pm SE (Standard error) of DM yield and quality traits in three phenological stages in two years in three forage species.

the nutritional value of desert plants in the Sabzevar region and stated that the percentage of the CP content of *Suaeda fruticosa* was 18.72%, which was consistent with the results obtained in our study. Also, *Seidlitzia rosmarinus* with 60–70% DMD in vegetative and flowering stages, which will increase up to 88.5% in the seed production stage due to the high amount of seeds produced at this stage and 13%, 12%, and 9% CP in vegetative, flowering, and seeding ripening stages, respectively. In addition, it is a valuable halophyte for combat desertification purposes in saline soils. This result is consistent with what was reported by Arzani et al. (2013). Similar to our results, Elahi et al. (2014) in examining the nutritional value of five halophytes in the Sistan region announced that *Seidlitzia rosmarinus* had the highest nutritional value among the species studied. Zadeh et al. (2015) and Hadi (2009) also introduced *Seidlitzia rosmarinus* as a halophyte species producing high-quality forage in saline and alkaline soils in terms of protein content and digestibility. *Atriplex leucoclada*, with about 7–10% CP in vegetative and flowering stages and 50–45% DMD is also grazed by livestock, especially sheep and camels on saline lands. Dadvar et al. (2016) studied the DMD of crop and rangeland forage in one-humped camels and found that DMD increased in treatments containing *Atriplex* at the end of the experiment.

It was concluded that *Suaeda fruticosa* having a higher DM yield in flowering stage (16.20 t/h) coupled with higher CP (18.04%) could be recommended for cultivation as forage sources and combating desertification in Khuzestan province Iran.

Authors contributions

All the authors have participated sufficiently in the intellectual content, conception and design of this work or the analysis and interpretation of the data (when applicable), as well as the writing of the manuscript.

Availability of data and materials

Data presented in the manuscript are available via request.

Conflict of interests

The author declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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