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Intraspecific variation in *Atriplex halimus*: Chemical composition of edible biomass

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SUMMARY – Chemical composition of 45 *Atriplex halimus* accessions from semiarid and arid Mediterranean countries are presented. Edible biomass (leaves and young stems) of 3 shrubs/accession was sampled in June and November of 2000, from *A. halimus* collection maintained in Mazarrón (Murcia), an arid locality in Southeast Spain, near the Mediterranean Sea. Accessions of *A. halimus* belonging to subspecies *halimus* were from Spain and France, and ssp. *schweinfurthii* from Southern and Eastern Mediterranean countries (Morocco, Algeria, Tunis, Syria, Egypt, Israel, etc.). Ash, crude protein (CP), neutral detergent fibre (NDF), acid detergent fibre (ADF), acid detergent lignin (ADL), nitrogen linked to fibre fractions and betaine content were measured in leaves and young stems and the period by vegetal material interaction was analysed. Ash content made up 29% of leaf weight and 6-11% of young stems weight in both samplings dates, hence, leaves contain 3-5 times more minerals than young stems; no significant differences between subspecies were found. However, 67 % of ssp. *halimus* edible biomass was made up of leaves, whereas in ssp. *schweinfurthii* leaves represented only a 50% of it. Crude protein represented 18% of leaf weight, but only 6% of young stems; hence, leaves contained three times more protein than young stems. Leaf protein ranged from 11.4% to 25.7% in ssp. *halimus*, and from 8.1% to 22.3% in ssp. *schweinfurthii*. The CP content of ssp. *halimus* was greater in the spring sampling whereas CP of ssp. *schweinfurthii* was greater in autumn. Mean NDF content (cell wall components) ranged from 31% to 33% in leaves, and from 69% to 84% in young stems. Differences in NDF, ADF and ADL between *A. halimus* subspecies were small. About 30% of leaf nitrogen was linked to cell wall components, values ranging from 23% to 40%. It seems there is enough variability between and within *A. halimus* accessions to select individual shrubs with significantly better values of chemical composition.

Keywords: Chemical composition, salt bush, seasonal variation, leaves, stems.

RESUME – "*Variation intra-spécifique d'*Atriplex halimus *: composition chimique de la biomasse consommable". Une étude de la composition chimique a été menée sur 45 populations d'*Atriplex halimus *des régions arides et semi-arides du bassin méditerranéen. Un échantillonnage a été réalisé en juin et un autre en novembre 2000. La biomasse consommable (feuilles et jeunes tiges) de 3 arbustes/population provenant d'une collection d'*Atriplex halimus *située à "Mazarrón" (Murcie), zone aride située au Sud-Est de l'Espagne a été prélevée. Les populations de* A. halimus *appartenant à la sous-espèce* halimus *provenaient d'Espagne et de France et celles de la ssp.* schweinfurthii *provenaient du Maroc, Algérie, Tunisie, Syrie et Israël. Différents paramètres ont été analysés : cendres, matières azotées totales (MAT), fibre neutre détergente (NDF), fibre acide détergente (ADF), lignine acide détergente (ADL), azote lié aux fractions fibreuses et bétaïne. Le contenu en cendres, pendant les deux dates d'échantillonnage, a été de 29% pour les feuilles et de 6 à 11 % pour les jeunes tiges. Par ailleurs, aucune différence significative n'a été observée entre les sous-espèces. Le contenu en MAT représente 18% dans les feuilles et seulement 6% dans les tiges. Le contenu en MAT dans les feuilles varie entre 11,4 et 25,7% pour la sous-espèce* halimus *et entre 8,1 et 22,3% chez ssp.* schweinfurthii. *Le contenu en MAT de ssp.* halimus *a été élevé dans l'échantillon pris au printemps, tandis que pour la ssp.* schweinfurthii*, le contenu en MAT a été élevé en automne. Le contenu moyen en NDF oscille entre 31 et 33,5% dans les feuilles jusqu'à 69-84% dans les jeunes tiges. De petites différences dans le contenu en NDF, ADF et ADL ont été remarquées entre les sous-espèces d'*Atriplex halimus. *Environ 30% de l'azote sont liés aux fibres. Ce travail a montré l'existence d'une grande variabilité intra et entre sous-espèces d'*Atriplex halimus*, ce qui nous permettra de sélectionner des arbustes en fonction de la composition chimique.*

Mots-clés : *Composition chimique, arbustes halophytes, variations saisonnières, feuilles, tiges.*

Introduction

In the arid and semiarid environments, the grass vegetation withers during the dry season and its

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growth is restricted or limited during the cold season, thus causing forage shortage for livestock. *Atriplex halimus* is a spontaneous shrub of the Mediterranean coast regions, adapted to dry conditions, such as water shortage, high temperatures in summer, and high salinity of the soil (Osmond *et al.,* 1980). On the other hand, *A. halimus* is a perennial shrub presenting outstanding browsing biomass that can cover part of the small ruminant nutritive needs during the forage shortage (Wilson, 1977). This makes *A. halimus* an attractive species for the arid and semiarid Mediterranean areas. There have been differences between several works regarding the chemical composition and *in vitro* digestibility of this shrubs throughout the year (El-Shatnawi and Mohawesh, 2000; Khorchani *et al*., 2000; Haddi *et al*., 2003). Nevertheless, the evolution of the chemical composition of *Atriplex* genera differs depending on the different works (Haddi *et al*., 2003, Muñoz *et al*., 1994; Correal *et al*., 1986). This may be due, in some degree, to the existing differences in the plant material (Muñoz *et al.,* 1994). On the other hand, in recent times two different *A. halimus* subspecies have been found: *schweinfurthii* and *halimus* (Le Houerou, 1992), the variability between them regarding chemical composition not yet being known.

The purpose of the present work is to comparatively study the chemical composition of the two subspecies of *Atriplex halimus* from 45 populations belonging to nine Mediterranean countries in two seasons of the year.

Materials and methods

A collection of 45 accessions of *Atriplex halimus* was planted in 1998 in Mazarrón (Spain), belonging to the subspecies *halimus* and *schweinfurthii*, from nine Mediterranean countries. Fifteen shrubs per population were planted and distributed in lines (one per population) with a planting frame of 2m x 2m. During the year 2000 the plants were sampled in two seasons: spring (7th June) and autumn (29th November). Three shrubs were sampled from each accession. Sampling consisted in picking the edible biomass (leaves and young stems of about 15 cm, which is considered to be the part browsed by animals). Once in the laboratory, the sprouts (about 200 g of green material per shrub) were separated into leaves and stems. The samples were dried at 60ºC in a forced air oven and then, ground to a size of 1 mm. The chemical analyses carried out were ash, crude protein (CP) (AOAC, 1990), neutral detergent fibre (NDF), acid detergent fibre (ADF), acid detergent lignin (ADL) (Goering and Van Soest, 1970), nitrogen linked to neutral detergent fibre (NNDF) and betaine (Grieve and Grattan, 1983). The results obtained, both from leaves and stems, were submitted to an analysis of variance according to a split-plot design where the subspecies and the populations constituted the main plots and the season the split plot.

Results and discussion

The analysis of variance (Table 1) shows significant differences between seasons for all the determinations carried out, both in leaves and stems with the exception of NDF in leaves. The evolution of the chemical composition of *Atriplex halimus* throughout the different seasons has been studied by different authors (Muñoz *et al*., 1994; Correal *et al.,* 1986; El Shatnawi and Mohawesh, 2000; Haddi *et al.,* 2003). The greatest variability in chemical composition of *Atriplex halimus* was found in spring and autumn. At the same time, both factors (subspecie and population) were significant ($P<0.05$).

Even though no joint analysis of variance has been carried out to test the differences between leaves and stems, the content of ashes and CP in the leaves was higher than that in the stems (29.1 vs 8.4 and 17.5 vs 5.9) (Table 2). On the contrary, El-Shatnawi and Turku (2002), in an experiment carried out in Jordan, found no significant differences between leaves and stems of *Atriplex* in the spring and autumn samples, while in winter they observed that CP content in the stems was higher than that in the leaves. Table 2 shows the variation range observed for ashes, CP, NDF and betaine, both in the leaves and stems. All the determinations for both organs show high variability. No significant differences were found between the two subspecies for ADF in leaves (14.2 and 14. 8 for *halimus* and *schweinfurthii* subspecies, respectively). ADL content in leaves for both subspecies was significantly different ($P<0.001$) obtaining a value of 8.3% for the *halimus* subspecies and 7.7% for *schweinfurthii.* The population (subspecie)*season interaction was significant for leaves and stems. Nevertheless, the subspecie*season interaction was only significant in the analysis of variance carried out with leaves (Tables 1 and 2). In Figs 1, 2 and 3 the contents of ash, CP and betaine of the two subspecies in spring and autumn are shown. The phenological differences between some populations of both species may explain the differences between species and populations in both seasons (data not shown). Other authors (Haddi *et al*., 2003; Correal *et al*., 1986) also observed different values for chemical composition in samples of *Atriplex halimus*, and different species of *Atriplex,* respectively picked at different phenological moments.

Table 1. Signification levels of the different factors in the analysis of variance for ash, crude protein (CP), neutral detergent fibre (NDF), acid detergent fibre (ADF), acid detergent lignin (ADL), nitrogen linked to neutral detergent (NNDF) fibre and betaine (BET) carried out on leaves (L) and stems (S)

	Ash		CP		NDF		ADF		ADL		NNDF		BET	
	L	S	L	S	L	S	L	S	L	S	L	S	L	S
Ssp.	ns	***	***	***	***	***	ns	***	***	**	*	ns	ns	-
P (Ssp.)	***	***	***	***	*	***	**	***	***	***	***	***	***	-
Season	**	***	***	***	ns	***	***	***	***	***	**	***	***	-
Ssp.*season	**	ns	***	ns	*	ns	***	ns	***	*	ns	ns	*	-
P (Ssp.)*season	**	***	***	**	***	***	ns	**	ns	**	ns	***	***	-

*P<0.05; **P<0.01; ***P<0.001; ns = P>0.05; Ssp. = Subspecies; P = Population.

Table 2. Variability observed for the determinations of ash, crude protein (CP), neutral detergent fibre (NDF), nitrogen linked to neutral detergent fibre (NNDF) and betaine (BET) in the data obtained for leaves (L) and stems (S)

	Ash			CP			NDF			NNDF			BET		
	Min	Med	Max	Min	Med	Max	Min	Med	Max	Min	Med	Max	Min	Med	Max
L	24.8	29.1	34.1	8.1	17.5	27.3	24.5	31.7	39.8	22.9	30.6	39.7	15.6	28.7	44.0
S	1.4	8.4	15.7	1.2	5.9	12.5	63.0	77.4	92.7	16.5	23.2	27.8	-		-

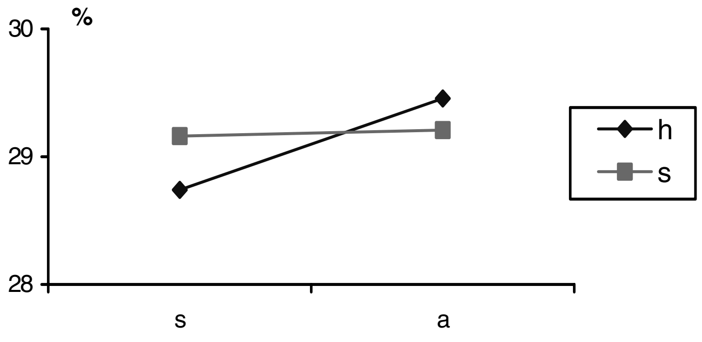

Fig. 1. Evolution of ash content in the leaf fraction of two subspecies of *Atriplex halimus* [*schweinfurthii* (s) and *halimus* (h) in the spring (s) and autumn (a) seasons].

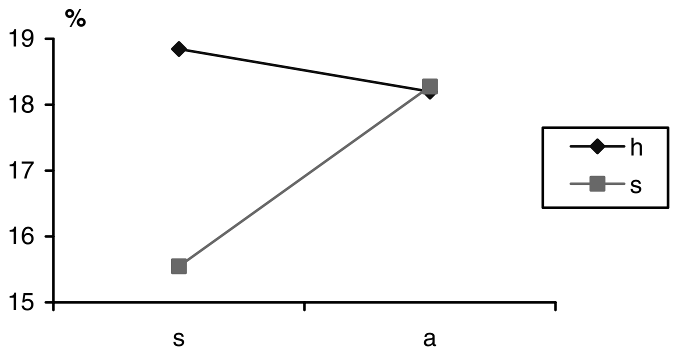

Fig. 2. Evolution of CP content in the leaf fraction of two subspecies of *Atriplex halimus* [*schweinfurthi* (s) and *halimus* (h) in the spring (s) and autumn (a) seasons].

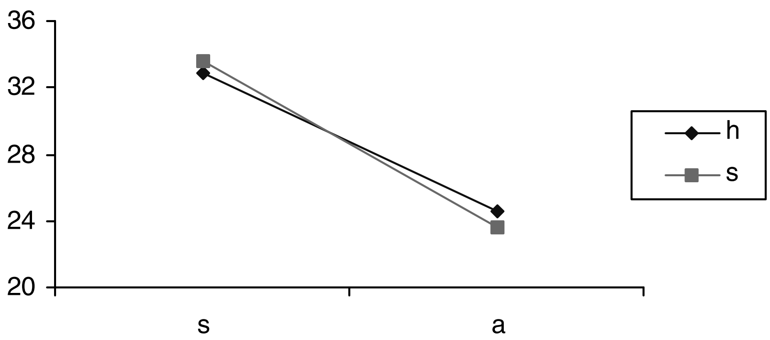

Fig.3. Evolution of betaine content in the leaf fraction of two subspecies of *Atriplex halimus* [*schweinfurthi* (s) and *halimus* (h) in the spring (s) and autumn (a) seasons].

Conclusions

The results obtained show the existence of high variability inter subspecies and intra subspecies of *Atriplex halimus*, and between sampling seasons, thus enabling the selection of the individual shrubs with best chemical composition values, depending on the different needs and exploitation systems.

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