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Impact of inter and intra-annual drought on chemical composition of some Mediterranean shrubs in natural rangelands

H. Ammar^{***}, A.Z.M. Salem^{*****} and S. López^{**}

^{*}Ecole Supérieure d'Agriculture de Mograne, IRESA, Mograne -Zaghouan 1121 (Tunisia)

^{**}Instituto de Ganadería de Montaña (CSIC-Univ. de León), Universidad de León, 24346 León (Spain)

^{***}Universidad Autónoma del Estado de México, Centro Universitario UAEM-Temascaltepec, Estado de México, CP 51300 (Mexico)

^{****}Alexandria University, Department of Animal Production, Faculty of Agriculture, El-Shatby (Egypt)
e-mail: hjr.mmr@gmail.com

Abstract. This study was conducted to determine drought induced nitrogen retranslocation in some Mediterranean shrub leaves as suggested from studies of annual changes in plant nitrogen content. In this regard, crude protein (CP) content and cell wall components in terms of neutral detergent fibre (NDF), acid detergent fibre (ADF) and acid detergent lignin (ADL) were assessed in five Spanish shrub leaves (*Quercus pyrenaica*, *Cytisus scoparius*, *Genista florida*, *Genista scorpius* and *Rosa canina*) harvested during wet (spring) and dry (summer) seasons of 1996 and 1998. Shoot N concentration decreased ($P < 0.0001$) in all species during drought occurred either in 1996 (15-42%) or in 1998 (21-53%). The lowest and the highest values ($P < 0.0001$) were recorded in *G. florida* and *Q. pyrenaica*, respectively. Cell wall components followed an opposite trend. Leaves from *C. scoparius* revealed the largest increase ($P < 0.0001$) of NDF (64%) and ADF (47%) in 1996 and ADL (216%) in 1998. No consistent pattern with respect to drought tolerance was apparent in these chemical composition changes among shrubs. *G. florida* and *G. scorpius* (leguminous) seem to be more tolerant and the magnitude of either CP decrease or cell wall content increase was lower as compared to the remaining species. It was suggested that decreases in leaf nitrogen (N) status during drought is a consequence of retranslocation likely result in lower photosynthetic capacity and decreased whole plant carbon gain following relief of water stress after rain. Drought induced re-translocation may serve to protect plant N from loss of herbivory during periods when soil N uptake and carbon assimilation are limited by water availability.

Keywords. Re-translocation – Nitrogen – Growth – Mitotic activity.

Influence du stade de maturité sur la composition chimique et la digestibilité in vitro de la matière sèche de certaines graminées et légumineuses

Résumé. Cette étude a été menée pour étudier la re-translocation de l'azote dans le feuillage de cinq arbustes fourragers (*Quercus pyrenaica*, *Cytisus scoparius*, *Genista florida*, *Genista scorpius* et *Rosa canina*) collectés de la montagne de León (Nord-ouest de l'Espagne). La collecte a été réalisée en deux saisons (printemps et en été) pendant deux ans (1996 et 1998). Afin d'atteindre notre objectif on a opté à la détermination de la composition chimique de ces espèces arbustives, en termes de protéine brut (PB), fibre détergente neutre (NDF), fibre détergente acide (ADF), lignine détergente acide (ADL). Les concentrations en azote les plus élevées ont été enregistrées au niveau du feuillage de *G. florida* et les plus faibles au niveau de *Q. pyrenaica*. Pour toutes les espèces étudiées, la concentration en azote (N) a suivie une chute durant les périodes de sécheresse (été) aussi bien au cours de la première année (15-42%) que la deuxième année (21-53%). Cependant les composés de la paroi cellulaire ont suivie une allure opposée à celle suivie par le contenu en PB. Les élévations les plus marquées ont été enregistrées par les feuilles de *C. scoparius*, soient 64% et 47% pour NDF et ADF, respectivement, durant la première année et 216% pour ADL durant la deuxième année. *G. florida* et *G. scorpius* (légumineuses) semblent être les plus tolérantes à la sécheresse et par la suite la chute de leurs contenus en PB ou l'augmentation de leurs parois cellulaires ont été plus faibles que celles observées par les autres espèces. La diminution de la concentration de l'azote des feuilles des espèces étudiées durant la sécheresse est la conséquence de la re-translocation du N comme due à une faible capacité photosynthétique et une diminution du gain du carbone par la plante entière après avoir assister au stress hydrique. Cette translocation induite par la sécheresse pourrait servir de protecteur de l'azote de la plante contre l'animal herbivore durant les périodes où l'assimilation du sol en N et en carbone est limitée par la disponibilité en eau.

Mots-clés. Re-translocation – Azote – Activité mitotique.

I – Introduction

In the Mediterranean extensive livestock production systems, woody and herbaceous plants are considered important contributors to grazing animal nutrition (Khazaal *et al.*, 1994). Promotion of suitable and nutritionally better species in these areas could be a practical approach to reduce fodder scarcity during long periods of drought (June-October) and to meet nutritional requirements of many animals. However, the rational use of these species as fodder for ruminants requires the knowledge of their nutritive value. The nutritive value of browse species is highly variable (Papachristou and Papanastasis, 1994) being affected by plant species, plant part, maturity stage (Ammar *et al.*, 2004a,b) and environmental factors such as seasonality, sunlight, temperature and water availability. Seasonal variations are mainly attributed to the physiological changes which occur in plants during their growing season. Moreover, species vary in their response to climatic and physiologic changes (Dann and Low, 1988). In this communication we report the effect of dryness, associated with plant maturity stage, on chemical composition of five Spanish shrub species.

II – Material and methods

1. Studied area and vegetal material

The sampling area is situated in the north of León (Northeast of Spain) at an altitude of 900 m above sea level. The climate is Mediterranean. Mean annual precipitation between 1961 and 1998 was 564 mm and an average temperature of 10.6° C through the year. Leaves from five browse species, namely *Quercus pyrenaica* (hoary oak), *Cytisus scoparius* (L.) Link (scotch broom), *Genista florida* L. (Iberian silver-leaved broom), *Genista scorpius* (L.) DC. (scorpion's thorn) and *Rosa canina* (wild dog rose) were collected in 1996 and 1998 from the uplands of the province of León (Norwest Spain). In every sampling year, and for every shrub species two samples were taken being in spring (leaf flushing) and summer. A total of twenty samples were submitted for this current study. In the laboratory, leaves were handily separated from the original samples, oven-dried at 60°C (samples collected in 1996) or freeze-dried (samples collected in 1998) and finally milled in (1-mm sieve).

2. Chemical analysis

Crude protein (CP) content was determined following the methods of AOAC (1995), whereas neutral detergent fibre (NDF), acid detergent fibre (ADF) and acid detergent lignin (ADL) were determined according to Van Soest *et al.* (1991), adding sodium sulphite to the solution and with the modifications proposed by ANKOM (Ammar *et al.*, 1999).

3. Statistical analysis

Data were analyzed using a randomized complete block design with 2 seasons (spring and summer) × 2 sampling years (1996 and 1998) × 5 browse species in factorial arrangement with 2 repetitions. Differences among means with $P < 0.05$, determined using a least square means test (LSMEANS), were accepted as representing statistically significant differences (Steel and Torrie, 1980).

III – Results and discussion

In this study, the CP contents were high (94-255 g/kg DM, ($P < 0.0001$) (Table 1), clearly superior to those commonly observed in grass species (Carew *et al.*, 1981). Leguminous species such as *C. scoparius*, *G. scorpius* and *G. florida* revealed the highest CP contents as compared with the other species. Similarly, Khanal and Subba (2001) reported high CP contents (>14% DM) in leaves of many leguminous shrubs due to their ability to fix atmospheric

nitrogen. The highest N concentration ($P < 0.0001$) was detected in leaves sampled in spring (April-May), during initial leaf growth. During this season, growth is accompanied by high mitotic activity due to cellular growth and a strong demand for nutrients, in particular N (Ryan and Bormann, 1982). Thereafter, the quantity of this element decreased ($P < 0.0001$), particularly during summer (Table 1). It is therefore evident that re-translocation to perennial tissues occurs before total abscission and it is accentuated under drought conditions.

Table 1. Chemical composition (g/kg DM) of shrubs collected in spring and summer during 1996 and 1998. Differences among means with $P < 0.05$ are reported below

Browse	Season	CP		NDF		ADF		ADL	
		1996	1998	1996	1998	1996	1998	1996	1998
<i>C. scoparius</i>	Spring	255	290	245	228	135	154	39	40
	Summer	152	164	402	299	199	157	103	58
<i>G. florida</i>	Spring	236	272	304	537	191	262	77	103
	Summer	201	216	426	483	247	285	125	135
<i>G. scorpius</i>	Spring	159	188	272	323	151	151	57	61
	Summer	124	123	345	365	167	166	79	82
<i>Q. pyrenaica</i>	Spring	195	251	408	467	217	225	50	32
	Summer	113	118	511	500	272	274	108	101
<i>R. canina</i>	Spring	147	176	343	340	120	117	46	28
	Summer	94	110	296	390	139	149	67	49
SED		0.966		1.285		0.965		0.648	
P value									
	Season	<0.0001		<0.0001		<0.0001		<0.0001	
	Year	<0.0001		<0.0001		<0.0001		<0.0001	
	Browse	<0.0001		<0.0001		<0.0001		<0.0001	
	Season x Year x Browse	<0.0001		<0.0001		<0.0001		<0.0001	

As reflected in Table 1, N content in leaves of all the studied species was higher during the second year of study (1998). The decrease ($P < 0.0001$) of this element from spring to summer was more appreciated in 1998 and reached 53% for *Q. pyrenaica*.

It seems therefore that under a higher temperature and a continuous precipitation among plant growth period, plants tend to accumulate efficiently nutrients in leaves and withdraw them progressively throughout the year (Milia *et al.*, 2004). Moreover, decrease in N concentration could be the result of decrease photosynthetic capacity and a decrease in whole-plant carbon gain. The magnitude of this decrease was higher in *Q. pyrenaica* (53%) and lower in *G. florida* (21%). It seems therefore that factors that affect net N retranslocation include the type and level of internal N reserves, plant species, biomass and N accumulation and partitioning, and other stresses that limit plant growth such as water availability and temperature. Likewise, cell wall contents revealed some seasonal variations, with an opposite trend to that for CP content. These results were reported earlier in the literature (Ammar *et al.*, 2004a,b) and could be explained by plant maturity stage and the progressive senescence of perennial vegetation. It is believed therefore that as plants mature, photosynthetic products are more rapidly converted to structural components, thus having the effect of decreasing protein and soluble carbohydrate and increasing the structural cell wall components (Ammar *et al.*, 2004a,b). On the other hand, high temperatures and water shortage during the summer bring on a more intense lignification of the cell wall. Moreover, the seasonal increase in lignin content associated with the plant maturity was generally more pronounced than the other cell wall fractions and the highest magnitude was observed in leaves of *Q. pyrenaica* sampled in 1998.

IV – Conclusion

On the basis of the results presented herein it can be concluded that browse species contain high levels of N especially in spring, during the initial leaf growth. The relative content of this element decreases after the period of maximum photosynthetic activity owing to the needs of maximum younger plant components; however, as plant mature N retranslocation to perennial tissues occurred. This phenomenon is accentuated under higher temperature and water shortage (summer). Effect of drought on the retranslocation of other elements such as carbon and phosphorus should be studied.

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