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Effect of fertilization and artificial shading on N and various mineral content of herbaceous species

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Abstract. Minerals and N have a significant impact on animal performance. The objective of this study was to investigate the effect of fertilization and artificial shading on N and various mineral contents of the above ground biomass content of the species *Festuca ovina*, *Dactylis glomerata*, *Trifolium repens* and *Trifolium pratense*. Three artificial shade treatments with 90%, 80% and 70% reduction of the total radiation and the control (0%) were tested. Species sowed in pots with soil from a *Pinus brutia* forest understorey. Six pots of each species were placed in each shade treatment and the control. Half of these pots were fertilized with 225 kg/ha of N, 450 kg/ha of P and 225 kg/ha of K after the sowing. Additionally, 395 kg/ha of NH_4NO_3 were top-dressed in April. At the end of the growing season the herbage biomass of each pot was cut at ground level and sampled. The samples were oven-dried at 60°C for 48 h and subsequently analyzed for N, P, Ca, K and Na. Shading treatments increased N content in grasses compared to control ($P < 0.05$). On the other hand, N content increased in legumes only at 90% shading ($P < 0.05$). There was no significant effect of the shade and fertilization on Ca ($P > 0.05$). Fertilization significantly increased the P content in grasses and did not affect it in legumes. The effect of fertilization and shade in K content strongly depended on plant species. Fertilization affects the content of Na only in sun, while the effect of shade depended on plant species. The content of N as well as P, Ca and K content except Na both in grasses and legumes were putatively sufficient to meet small ruminants' maintenance demands. To conclude, a diversity of plant species should be offered to small ruminants grazing pine-based understorey in order to offer them the opportunity to select a balanced diet.

Keywords. Nitrogen – Macrominerals – Grasses – Legumes – Small ruminants' requirements.

Effet de la fertilisation et de l'ombrage artificiel sur la teneur en azote et en minéraux de différentes espèces herbacées

Résumé. *Les minéraux et l'azote ont un impact significatif sur les performances animales. L'objectif de ce travail était d'étudier l'effet de la fertilisation et de l'ombrage artificiel sur les teneurs en azote et en différents minéraux de la partie aérienne de* Festuca ovina*,* Dactylis glomerata*,* Trifolium repens *et* Trifolium pratense*. Trois types d'ombrage artificiel ont été évalués : 90%, 80% et 70% de réduction de l'irradiation totale. Ces traitements ont été comparés à un témoin (0%). Les espèces végétales susmentionnées ont été semées dans des pots remplis de terre provenant d'une forêt de* Pinus brutia*. Six pots de chaque espèce ont été placés sous chaque type d'ombrage artificiel et le témoin. La moitié de ces pots a été fertilisée avec 225 kg/ha d'azote, 450 kg/ha de potassium après semis. En outre, 395 kg/ha de NH_4NO_3 ont été épandus en avril. A la fin de la saison de croissance, la biomasse végétale au niveau de chaque pot a été coupée à ras de sol et échantillonnée. Les échantillons ont été séchés à 60℃ pendant 48 h puis analysés pour leurs teneurs en N, P, Ca, K et Na. Comparé au témoin, l'ombrage artificiel a entraîné une augmentation de la teneur de l'herbe en azote ($P < 0,05$). L'augmentation de la teneur en azote dans les légumineuses a été notée uniquement avec le traitement 90% d'ombrage ($P < 0,05$). La teneur en Ca n'a été affectée ni par l'ombrage ni par la fertilisation. La fertilisation a augmenté la teneur en P uniquement des graminées. L'effet de cette fertilisation et de l'ombrage sur la teneur en K a varié en fonction de l'espèce végétale. La fertilisation affecte la teneur en Na uniquement lorsque la plante est exposée au soleil, alors que l'effet de l'ombre dépend de l'espèce végétale. Excepté le Na, les teneurs en N, P, Ca et K chez les graminées et les légumineuses permettent de couvrir les besoins d'entretien des petits ruminants. En conclusion, la diversification des espèces fourragères destinées aux petits ruminants pâturant dans des forêts à base de pins est recommandée pour leur permettre de sélectionner une ration équilibrée.*

Mots-clés. *Azote – Macro-minéraux – Graminées – Légumineuses – Besoins des petits ruminants.*

I – Introduction

In silvopastoral systems grazing animals coexist with tree production (Rigueiro-Rodríguez *et al.*, 2005). In these systems there is a great competition between woody and herbaceous plants especially for light intensity (Braziotis and Papanastasis, 1995) and soil nutrients (Rao *et al.*, 1998) which affect forage quality. Fertilization has been documented to increase both forage production and tree growth (Sinclair *et al.*, 2000). In addition moderate shade improves the nutritive value of the herbage vegetation as it increases crude protein content of forage (Koukoura and Nastis, 1989; Kyriazopoulos *et al.*, 1999).

Although there are many studies on the effect of shade to crude protein content, there is limited literature concerning minerals, which also influence forage quality (Provenza, 1995) and have a significant impact on animal performance and health (Grunes and Welch, 1989). Mineral contents of forage plants are affected by many factors, such as forage species, stage of maturity, and fertilization (Galdámez-Cabrera *et al.*, 2004). However, limited information exists on how shade and fertilization or their interactions affect mineral content of various plant species. The objective of the present study was to investigate the effect of fertilization and artificial shading on N content and various mineral contents of some perennial grasses and legumes.

II – Materials and methods

The experiment was carried out at the Aristotle University's farm (40° 34' E, 23° 43' N, at sea level) in Macedonia, northern Greece. The climate is semi-arid with a mean annual temperature 16.4°C and a mean annual precipitation 374 mm. Two perennial grass species *Dactylis glomerata* L. (D.gl.) and *Festuca ovina* L. group (F.ov.) and two perennial legume species *Trifolium pratense* L. (T.pr.) and *Trifolium repens* L. (T.re) were seeded in 26 cm diameter and 26 cm height pots. Each species was planted in 24 pots filled with soil from a *Pinus brutia* forest understorey from Chrisopigi, Serres. There were three artificial shade treatments with 90%, 80% and 70% reduction of the total radiation and the control (0%). Six pots of each species were placed in each shade treatment and the control. Half of these pots were fertilized with 225 kg/ha of N, 450 kg/ha of P and 225 kg/ha of K after the sowing. Additionally 395 kg/ha of NH_4NO_3 were topo-dressed in April. At the end of growing season the above ground biomass was harvested and sampled. The samples were oven-dried at 60°C for 48 h and subsequently analyzed for N (AOAC, 1990), for P, Ca, K and Na content using standard methods (Papamichos and Alifragis, 1988). The experimental layout was a split-split plot design (Snedecor and Cochran, 1980) with three replications. Differences among means were determined by the LSD test at $P < 0.05$ level of significance.

III – Results and discussion

Fertilization increased significantly N content of the grasses ($P < 0.05$) only in the sun treatment compare to the control, while there was no significant differences under shade (Table 1). There was a significant interaction among fertilization × shade × species. Concerning the legume, fertilization increased significantly N content of T.pr. in all shade levels and T.re. only in 90% shade level. This is in agreement with findings of Lin *et al.* (2001).

Moreover, F.ov. had significantly higher N content in all shade levels, compared to full sun but D.gl. only in 70%. Generally, CP was higher in shade-grown cool season grasses compared with unshaded treatments (Burner and Brauer, 2003). Papanastasis *et al.* (1995) found also, that CP content of D.gl. was increased with increasing shade level. Our findings indicate that CP content of tested grasses species met only the requirements of small ruminants' maintenance, while CP content of the legumes met and their lactation requirements too (NRC, 1981, 1985).

Table 1. Nitrogen content (%) of DM of the tested species under shading levels and fertilization (F) and non fertilization (NF)

Species	0%		70%		80%		90%	
	F	NF	F	NF	F	NF	F	NF
D.gl.	2.3 aBC	0.9 dC	2.1 abC	2.2 aB	1.6 cC	1.5 cB	1.8 bcC	1.5 cB
F.ov.	2.1 bcC	1.5 dB	2.4 bBC	2.8 aA	2.5 abB	2.8 aB	2.4 bB	2.8 aB
T.re.	2.7 bcA	2.4 cdA	2.7 bcAB	2.2 dB	2.4 cdB	2.4 cdA	3.2 aA	2.8 aA
T.pr.	2.4 cAB	2.5 cA	2.9 abA	2.5 cAB	3.2 aA	2.6 bcA	3.2 aA	3.1 aA

a, b, c, d: means with different lower case letter in the same row differ significantly ($P < 0.05$).
A, B, C: means with different capital letter in the same column differ significantly ($P < 0.05$).

Phosphorus content of D.gl. was lower under 80% and 90% shade compared to full sun and to 70% shade while there were no significant differences for F.ov. However, fertilization increased significantly P content in both grass species in comparison with the control in all treatments (Table 2). On the other hand, there was no significant effect of both fertilization and shade in the legumes species. Phosphorus content in the tested species was higher from that required to meet sheep maintenance demands (0.16-0.20% of DM) suggested by (NRC, 1985; Grace, 1994) except of D.gl. at the 80% and 90% shade level which was lower.

Table 2. Phosphorus content (%) of DM of the tested species under shading levels and fertilization (F) and non fertilization (NF)

Species	0%		70%		80%		90%	
	F	NF	F	NF	F	NF	F	NF
D.gl.	0.38 abA	0.24 dAB	0.33 bcA	0.33 bcA	0.28 cdB	0.13 eC	0.27 dB	0.15 eC
F.ov.	0.32 bcB	0.22 eAB	0.37 abA	0.26 deB	0.41 aA	0.24 deB	0.42 aA	0.28 cdB
T.re.	0.26 abC	0.26 abA	0.27 abB	0.25 abB	0.29 abB	0.30 aA	0.28 abB	0.24 bAB
T.pr.	0.22 bC	0.20 cB	0.26 abB	0.25 abcB	0.24 abcB	0.28 aAB	0.24 abcB	0.20 cBC

a, b, c, d, e: means with different lower case letter in the same row differ significantly ($P < 0.05$).
A, B, C: means with different capital letter in the same column differ significantly ($P < 0.05$).

Fertilization and shade had no significant effect on Ca content of the tested species (Table 3). Similar results were found for D.gl. by Rigueiro-Rodríguez *et al.* (2007) for fertilization and for shade by Papanastasis *et al*. (1995). It is well established that Ca content of legumes species is significantly higher from that of grasses (McDowell, 1992; Juknevicius and Sabiene, 2007). This concurs with our findings. Ca content of the tested species under all treatments was sufficient to meet sheep (NRC, 1985) and goats (Meschy, 2000) demands for maintenance.

In the current study the Ca:P ratio was between 1.4 to 7.1 for the grasses. Although the generally recommended ratio of Ca:P is 2:1, ranges in dietary Ca:P between 1:1 and 7:1 resulted in similar performance of ruminant livestock (Buxton and Fales, 1994) without unfavourable effects if adequate vitamin D is available (Barnes *et al.*, 1990). On the other hand, the Ca:P ratio for the legumes was between 7.7 to 11.8. Low P and high Ca content resulted in this wide Ca:P ratios (Ramírez-Orduña *et al*., 2005).

Table 3. Calcium content (%) of DM of the tested species under shading levels and fertilization (F) and non fertilization (NF)

Species	0%		70%		80%		90%	
	F	NF	F	NF	F	NF	F	NF
D.gl.	0.8 aB	0.8 aB	0.9 aB	1.3 aB	1.2 aB	0.9 aB	1.0 aB	1.0 aB
F.ov.	1.0 aB	0.8 aB	0.7 aB	0.6 aB	0.8 aB	0.8 aB	0.6 aB	0.6 aB
T.re.	2.4 aA	2.0 aA	2.4 aA	2.4 aA	2.7 aA	2.4 aA	2.4 aA	2.4 aA
T.pr.	2.6 aA	2.2 aA	2.4 aA	2.4 aA	2.6 aA	3.0 aA	2.4 aA	2.4 aA

A: means with different lower case letter in the same row differ significantly ($P < 0.05$).
A, B: means with different capital letter in the same column differ significantly ($P < 0.05$).

Potassium (K) content of F.ov. and T.pr. was significantly higher in shade than in full sun (Table 4). Conversely, D.gl. and T.re. had significantly lower K content in shade than in full sun. Fertilization did not affect K content for D.gl. and T.re. in all shade treatments, while, it increased for T.pr. with increasing shade and for F.ov. for all treatments. Potassium content in all treatments was above that required to meet maintenance requirements of small ruminants (NRC, 1985).

Table 4. Potassium content (%) of DM of the tested species under shading levels and fertilization (F) and non fertilization (NF)

Species	0%		70%		80%		90%	
	F	NF	F	NF	F	NF	F	NF
D.gl.	1.9 bB	2.8 aA	2.6 aA	2.7 aA	2.0 bC	1.6 cC	1.6 cB	1.7 cC
F.ov.	1.7 cB	1.4 cC	3.0 aA	2.7 aA	2.8 aA	2.1 bB	2.7 aA	2.3 bAB
T.re.	2.8 abA	2.8 abA	2.9 aA	2.8 abA	2.6 bcAB	2.4 cA	2.7 abA	2.0 dBC
T.pr.	1.6 dC	2.1 cB	2.6 abA	2.1 cB	2.3 bcBC	2.2 cAB	2.9 aA	2.6 aA

a, b, c, d: means with different lower case letter in the same row differ significantly ($P < 0.05$).
A, B, C: means with different capital letter in the same column differ significantly ($P < 0.05$).

Fertilization decreased significantly Na content of tested grasses compared to the control as well as for D.gl. in 70% shade treatment (Table 5), while there was no significant difference in shade in all other cases. In addition, Na content of F.ov. is lower in shade than in sun.

Table 5. Sodium content (%) of DM of the tested species under shading levels and fertilization (F) and non fertilization (NF)

Species	0%		70%		80%		90%	
	F	NF	F	NF	F	NF	F	NF
D.gl.	0.03 cdA	0.06 aB	0.02 dAB	0.05 abA	0.04 bcA	0.05 abA	0.02 dB	0.03 cdB
F.ov.	0.02 bcB	0.04 aB	0.01 cB	0.01 cC	0.01 cC	0.01 cC	0.01 cB	0.01 cBC
T.re.	0.01 cBC	0.03 bA	0.03 bA	0.03 bB	0.04 abA	0.04 abB	0.05 aA	0.04 abA
T.pr.	0.08 aC	0.03 bA	0.02 bB	0.02 bBC	0.02 bBC	0.02 bC	0.03 bB	0.03 bC

a, b, c, d: means with different lower case letter in the same row differ significantly ($P < 0.05$).
A, B, C: means with different capital letter in the same column differ significantly ($P < 0.05$).

Fertilization increased Na content in T.pr. and decreased in T.re. in the sun treatment, while there was no significant difference in shade, despite the fact that T.re. had a progressively higher content in increasing shade level. Content of Na in all treatments was insufficient to meet small ruminant demands (0.09%) (Grace, 1994) indicating the need for a continuous supply of salt to the animals.

IV – Conclusions

It seems that shade treatment increases N content of grass species without any contribution of fertilization. N content of D.gl. was significantly higher in the 70% shade treatment compared to the other treatments. Shade and fertilization combination increased N content of legumes species. Fertilization and shade did not affect Ca content in both grasses and legumes. Fertilization increased P content in grasses and did not affect it in legumes. The effect of fertilization and shade in K content strongly depended on plant species. Fertilization decreased the content of Na only in sun treatment, except T.pr., while the effect of shade depended on plant species. All species contained N and the minerals Ca, K and P in excess of the recommended levels for small ruminants maintenance requirements, expect Na which was insufficient to meet their maintenance requirements.

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