Effects of Nitrogen and Phosphorus Fertilization on Micro Nutrient Contents of
Trifolium angustifolium and Lotus suaveolens from Fabaceae on a Grassland
Ecosystem: the Case of Kahramanmaraş, Eastern Mediterranean Region of Turkey

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Abstract: This study examined the effectiveness of various grass plants as Trifolium angustifolium and Lotus suaveolens from Fabaceae to fertilizer treatments (N+P fertilization) to encourage the micro nutrition of this vegetation. Micro nutrient contents of this vegetation were determined annually in 2002 and 2003. Experimental plots received rains of 606.3 and 857.5 mm during the 2 study years compared to mean growing season rainfall of 729.8 mm. Micro nutrient contents of these grass plants were increased significantly by nitrogen and phosphorus fertilizers application. Contents of micro nutrients after nitrogen and phosphorus fertilizer treatments, it is determined and ordered for Lotus suaveolens Cu>Fe>Zn>Mn and for Trifolium angustifolium Cu>Fe=Mn>Zn. Nitrogen and phosphorus chemical fertilizers influence both micro nutrient mobility and bioavailability and the effects to these plants may be different.

Key words: Trifolium angustifolium, Lotus suaveolens, fabaceae, fertilization, micro nutrient, grass plants

INTRODUCTION

The Mediterranean area is characterized by it is climate and soil nature presents a great extent of natural spaces where grows a varied pastoral vegetation containing considerable forage varieties for sheep and goat livestock. So in area represented by Morocco, Algeria, Tunisia, Libya, Egypt, Palestine, Lebanon, Jordania, Syria and Turkey number of sheep and goats represents, respectively 200.219 and 25.359 million heads (Medagri, 2001).

Most Mediterranean grasslands are on soils deficient in one or more plant nutrients (Seligman, 1996). Pasture production is poor in areas of low soil phosphorus availability (Henkin et al., 1996). Whereas phosphorus addition can cause a burst of productivity with an increase in plant cover of legume species (Osman et al., 1991, 1999) little is known on how changes in productivity affect species richness, particularly in Mediterranean environments (Dabaschasche et al., 1996).

As a rule, high productivity tends to reduce species diversity (Tilman and Pacala, 1993; Janssens et al., 1998), especially when it is caused by nitrogen enrichment (Foster and Gross, 1998) but the effects of phosphorus enrichment are less well understood (Henkin et al., 1996). Turkey is located at the intersection of two important gene centers like Mediterranean and near East and in the ninth order among all continental countries in terms of biodiversity. Natural races of most grass plants cultivated as animal and human food grow on grass ecosystems (Vavilov, 1951; Ture and Becuk, 2007).


Competition may be more severe between similar species than between species with contrasting growth patterns and nutritional needs. Even so, all plants compete for the same resources (light, water and nutrients). There is an overlapping of resource requirement with nitrogen fixing plants as an exception (Rahef, 2007).

Biodiversity is a key factor in ecosystem structure and function. Conservation of biodiversity is now a legal obligation for the member states of the European Community (Baker, 2002; Barbrecheck and Neher, 1999). Natural grasslands belong to the most valuable ecosystems and are a result of stable agricultural management over centuries by using the grasslands as
As a result of this stable management, the grassland ecosystem is well developed and characteristic for the bio-geographical region. Also, typical for natural grasslands is the low input of nutrients in the grassland ecosystems.

As a result of this, less competitive grassland species are able to survive in these grasslands. Last but not least, natural grasslands need management by continuation of farming traditions like fertilizing, pasturing and cutting of grasslands (Meshinev et al., 2005). On the other hand, the native pasture ability to sustain animal production and nutritive value of pasture (Syman, 2002).

In the pasturelands and grasslands, it is important to find out alternative methods by which production and nutrition can be increased without causing unfavorable fertilizer doses. The Mediterranean region of Turkey is quite well-known from the floristic and vegetational points of view. No fertilizer study on grassland has previously been carried out in this area.

The micro nutrient content of plants presents type of approach in determining the nutrient availability of a soil. When the nutrient content or concentration in the plant tissue is very low the rate of growth is also low. The nutrient content of a plant tissue not only reflects soil availability.

It is also affected by other factors such as the kind of plant or tissue, the age of plant and the supply of the plant with other plant nutrients (Mengel and Kirkby, 1978). A survey of the sufficient levels of micro nutrients, especially Fe, Cu, Zn and Mn in different feed plants or materials is shown in Table 1.

The objective of this study was to determine the effects of level N and P fertilizations of various grass plants as *Trifolium angustifolium* and *Lotus suaveolens*, on micro nutrition contents of these plants in Kahramanmaras, Turkey.

Within the general goal of increasing grassland productivity, the particular aim of the present study was to examine the responses of a mixed community of plants in this grassland to micro nutrient enrichment as expressed in the effects on these species.

![Table 1: Sufficient levels (ppm) of the micro nutrients in different feed plants with references](image)

<table>
<thead>
<tr>
<th>Elements</th>
<th>Trifolium</th>
<th>Bean</th>
<th>Meadow grass</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fe</td>
<td>214-261</td>
<td>-</td>
<td>-</td>
<td>Bear, 1954</td>
</tr>
<tr>
<td>Cu</td>
<td>14-112</td>
<td>11</td>
<td>-</td>
<td>Beseon, 1941</td>
</tr>
<tr>
<td>Zn</td>
<td>14-112</td>
<td>-</td>
<td>360</td>
<td>Beseon, 1941</td>
</tr>
<tr>
<td></td>
<td>20-128</td>
<td>11</td>
<td>-</td>
<td>Prince et al., 1955</td>
</tr>
<tr>
<td>Mn</td>
<td>High</td>
<td>-</td>
<td>-</td>
<td>Scharrer and Monk, 1956</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>McHargue, 1945</td>
</tr>
</tbody>
</table>

Characteristics of plants: *Trifolium angustifolium*. *Trifolium angustifolium* is an annual herb of Fabaceae, Leguminosae family, *Trifolium angustifolium* L. variety. *Lotus suaveolens*: *Lotus suaveolens* is an annual herb of Fabaceae, Leguminosae family, *Lotus suaveolens* Pers. variety. Each of *Trifolium angustifolium* and *Lotus suaveolens* were collected during the reproductive stage of growth for micro nutrient analysis. Each plant was collected from random sites from the interior of each subplot.

Analyses of soil: The soil sample was air-dried and passed through a 2 mm sieve before analysis. The soil pH
Fertilizer application rates were 0, 50, 75, 100 and 150 kg ha$^{-1}$ N and 0, 40, 60, 80 and 100 kg ha$^{-1}$ P$_2$O$_5$ in a factorial treatments arrangement. A commercial N and P fertilizers were used the ammonium nitrate (33% N) and triple super phosphate (45% P$_2$O$_5$), respectively. Phosphorus and nitrogen fertilizers were broadcast by hand in 5 November 2002 and 2003 and in 12 March 2002 and 2003, respectively.

**Analyses of micro nutrients in the plants:** Herbaceous vegetation was harvested annually within each plots, 0.5 m$^2$ located within each plot when plants were flowering. After taking flowering plant material, all of the plant materials were dried in oven under 60°C and then grinded. The sample of 0.5 g grinded plant material was exposed to burning by using the mixing of nitric acid and perchloric acid. After the completion of burning, the sample was diluted and then the instrumental analysis was conducted. The elements of Fe, Cu, Zn and Mn were determined by atomic absorption spectrophotometer, AAS 3110 Perkin Elmer®.

**Statistical analyses:** ANOVA was performed for each response variable (Fe, Cu, Zn and Mn) using the SPSS. The LSD test was used to assess differences among treatment means ($p<0.05$) (SPSS, 2003; Steel and Torrie, 1980).

**RESULTS AND DISCUSSION**

The grassland situated experiment has red brown Mediterranean soils. Some chemical and physical analyses of the major soil groups in the area are given in
Table 2. Soils of experiment area are non-calcareous with acidic, insufficiently provided in organic matter N, P and K. Micro nutrients contents of *Trifolium angustifolium* and *Lotus suaveolens* were found to accumulate in the plants growing in this field study. As well as increasing micro nutrient mobility through these plants, nitrogen and phosphorus fertilizers application may also bioavailability of micro nutrients to plants. To quantify amounts of micro nutrients in the plants after nitrogen and phosphorus fertilizer treatments, it was determined micro nutrient contents of these plants. During two years (2002 and 2003) of nutrient enrichment, generally concentrations of all four micro nutrients (Fe, Cu, Zn and Mn) were higher in nitrogen and phosphorus fertilizer application than control (no nitrogen and phosphorus, NOPO).

### Micro nutrients contents of *Trifolium angustifolium* plant:

Application of nitrogen and phosphorus at different concentrations had significantly influenced on the micro nutrient contents of *Trifolium angustifolium* plant (Table 3). Micro nutrients concentrations of Fe, Cu, Zn and Mn were determined in 2002 and 2003 for *Trifolium angustifolium*. Mean Fe, Cu, Zn and Mn concentrations in *Trifolium angustifolium* ranged from 92.36 -551.41 ppm, 7.22-31.03 ppm, 18.03-43.48 ppm, 221.4-90-2231.48 ppm, respectively.

With nitrogen application, Fe and Cu contents in *Trifolium angustifolium* were significantly greater in N15 (150 kg N ha⁻¹) and N5 (50 kg N ha⁻¹) applications, whereas Zn and Mn contents in *Trifolium angustifolium* was significantly greater in N0 (control) and N7.5 (75 kg N ha⁻¹) applications (Table 3).

### Micro nutrients contents of *Lotus suaveolens* plant:

Application of nitrogen and phosphorus at different concentrations had significantly influenced on the micro nutrient contents of *Lotus suaveolens* plant (Table 4). Micro nutrients concentrations of Fe, Cu, Zn and Mn were determined in 2002 and 2003 for *Lotus suaveolens*. Mean Fe, Cu, Zn and Mn concentrations in *Lotus suaveolens* ranged from 123.55-583.30 ppm, 4.99-31.57, 2.54-69.66 ppm, 221.47-2231.48 ppm, respectively.

With nitrogen application, Fe and Zn ppm contents in *Lotus suaveolens* were significantly greater in N15 (150 kg N/ha) application, whereas Cu and Mn contents in *Lotus suaveolens* was significantly greater in N10 (100 kg N ha⁻¹) and N7.5 (75 kg N ha⁻¹) applications (Table 4).

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### Table 2: Soil general characteristics of study area

<table>
<thead>
<tr>
<th>Soil properties</th>
<th>2002</th>
<th>2003</th>
</tr>
</thead>
<tbody>
<tr>
<td>Texture</td>
<td>Loam</td>
<td>Loam</td>
</tr>
<tr>
<td>Organic matter</td>
<td>2.180</td>
<td>2.190</td>
</tr>
<tr>
<td>pH</td>
<td>5.280</td>
<td>5.250</td>
</tr>
<tr>
<td>EC (µS/cm)</td>
<td>0.010</td>
<td>0.010</td>
</tr>
<tr>
<td>CaCO₃ (°%)</td>
<td>0.390</td>
<td>0.420</td>
</tr>
<tr>
<td>N (%)</td>
<td>0.110</td>
<td>0.120</td>
</tr>
<tr>
<td>P₂O₅ (kg ha⁻¹)</td>
<td>11.00</td>
<td>12.50</td>
</tr>
<tr>
<td>K₂O (kg ha⁻¹)</td>
<td>69.90</td>
<td>72.65</td>
</tr>
</tbody>
</table>

### Table 3: Micro nutrients (Fe, Cu, Zn and Mn) contents of *Trifolium angustifolium* plant in 2002 and 2003

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Fe contents (ppm)</th>
<th>Cu contents (ppm)</th>
<th>Zn contents (ppm)</th>
<th>Mn contents (ppm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>NOPO</td>
<td>111.97</td>
<td>119.31</td>
<td>8.05</td>
<td>8.28</td>
</tr>
<tr>
<td>NOPO4</td>
<td>307.48</td>
<td>332.23</td>
<td>7.62</td>
<td>7.65</td>
</tr>
<tr>
<td>NOPO6</td>
<td>226.47</td>
<td>257.39</td>
<td>12.12</td>
<td>11.08</td>
</tr>
<tr>
<td>NOPO8</td>
<td>184.07</td>
<td>195.42</td>
<td>13.26</td>
<td>12.22</td>
</tr>
<tr>
<td>NOPO10</td>
<td>251.56</td>
<td>258.91</td>
<td>11.85</td>
<td>13.18</td>
</tr>
<tr>
<td>N5PO</td>
<td>254.82</td>
<td>258.83</td>
<td>7.36</td>
<td>7.39</td>
</tr>
<tr>
<td>N5P4</td>
<td>97.42</td>
<td>113.91</td>
<td>9.55</td>
<td>7.95</td>
</tr>
<tr>
<td>N5P6</td>
<td>98.60</td>
<td>101.63</td>
<td>11.43</td>
<td>9.06</td>
</tr>
<tr>
<td>N5P8</td>
<td>92.36</td>
<td>97.87</td>
<td>31.03</td>
<td>11.25</td>
</tr>
<tr>
<td>N5P10</td>
<td>154.07</td>
<td>145.22</td>
<td>14.14</td>
<td>15.40</td>
</tr>
<tr>
<td>N7.5P10</td>
<td>245.48</td>
<td>244.53</td>
<td>15.23</td>
<td>11.95</td>
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<tr>
<td>N7.5P4</td>
<td>333.94</td>
<td>271.51</td>
<td>10.58</td>
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<td>239.14</td>
<td>250.00</td>
<td>7.72</td>
<td>11.09</td>
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<td>N7.5P8</td>
<td>173.49</td>
<td>180.25</td>
<td>7.45</td>
<td>9.38</td>
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<tr>
<td>N7P10</td>
<td>147.38</td>
<td>146.58</td>
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<td>8.92</td>
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<tr>
<td>N10PO</td>
<td>135.44</td>
<td>142.94</td>
<td>13.52</td>
<td>13.01</td>
</tr>
<tr>
<td>N10P4</td>
<td>158.90</td>
<td>159.13</td>
<td>11.85</td>
<td>11.85</td>
</tr>
<tr>
<td>N10P6</td>
<td>392.26</td>
<td>286.37</td>
<td>11.32</td>
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<tr>
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<td>194.12</td>
<td>195.62</td>
<td>14.59</td>
<td>14.09</td>
</tr>
<tr>
<td>N10P10</td>
<td>407.84</td>
<td>337.65</td>
<td>14.52</td>
<td>14.99</td>
</tr>
<tr>
<td>N1P5O</td>
<td>228.48</td>
<td>224.97</td>
<td>10.71</td>
<td>12.41</td>
</tr>
<tr>
<td>N1P4</td>
<td>147.42</td>
<td>148.25</td>
<td>16.38</td>
<td>13.61</td>
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<tr>
<td>N1P6</td>
<td>319.05</td>
<td>261.63</td>
<td>7.25</td>
<td>8.88</td>
</tr>
<tr>
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<td>280.52</td>
<td>274.21</td>
<td>9.12</td>
<td>8.38</td>
</tr>
<tr>
<td>N1P10</td>
<td>551.41</td>
<td>281.01</td>
<td>10.79</td>
<td>10.87</td>
</tr>
<tr>
<td>LSD</td>
<td>(0.01)</td>
<td>139.36</td>
<td>53.03</td>
<td>5.34</td>
</tr>
</tbody>
</table>

**Fertilizer applications are presented significant value as p<0.01. *Fertilizer applications are presented significant value as p<0.05. PNon-significant value**

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Early studies recorded that sufficient micro nutrient contents in the different feed plant contained from 214-261 ppm of Fe uptake by a range of the different feed crops.

The Beeson found that for Cu and Zn, sufficient levels occurred at ranged from 11-112 ppm and from 13-300 ppm in the feed crops, especially the highest accumulation in the leguminous plants. Prince et al. and Schorr and Munk also indicated that for Zn, sufficient levels were around 11-128 ppm and 34 ppm, respectively. As suggested by McHargue Mn concentration in the grass plants was in the highest levels.

In this study, it was appeared that each plant species has a different affinity for uptake and accumulation of different micro nutrient content. Trends were observed between plants of different uptake. The gradual increase in micro nutrient identified in Trifolium angustifolium and Lotus suaveolens in Kahramanmaras, Turkey. The increasing order effects of nitrogen and phosphorus fertilizer applications to micro nutrient contents of these grassland plants were followed as according to the iron, copper, zinc and manganese contents of plants:

Fe, Cu and Zn: Lotus suaveolens > Trifolium angustifolium
Mn: Trifolium angustifolium > Lotus suaveolens

Quantified amounts of micro nutrients after nitrogen and phosphorus fertilizer treatments, it is determined and ordered effectiveness of added fertilizers in increasing micro nutrient contents to these plants followed the order also:

Lotus suaveolens: Cu > Fe > Zn > Mn
Trifolium angustifolium: Cu = Fe = Mn > Zn

Since, added nitrogen and phosphorus chemical fertilizers influence both micro nutrient mobility and bioavailability and the effects to these plants may be different. The selectivity and availability of micro nutrients by adding nitrogen and phosphorus nutrients depends on the plants species. We found that the availability of Cu with adding fertilizers increased in Lotus suaveolens and Trifolium angustifolium plants, Fe increased in Lotus suaveolens plant and Trifolium angustifolium with Mn. In addition to these effects, there is clear evidence that Trifolium angustifolium plant species show non-differential Fe and Mn uptakes in selectivity.

CONCLUSION

As we found in this study, nutrient application especially nitrogen and phosphorus is increased generally, micro nutrients contents of plants. It is important to vegetation and animals in ecosystems. In addition to these effects, there is evidence that species,
genera and families of plants and animals show different
effect to nutrient uptake. By adding nitrogen and
phosphorus to investigated four types of plants,
Trifolium angustifolium and Lotus suaveolens, were
effected to accumulate micro nutrients in different. This
relative selectivity of plants may be used on
physiological, animal nutrition and environmental
pollution studies and of these plants.

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