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Response of Spearmint (*Menthaspicata* L.) to Nitrogen and Phosphorus Fertilizers at Koka, Ethiopia

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Spearmint (*Mentha spicata* L.) is perennial aromatic plant and productive up to 15 years, primarily cultivated mainly for its aromatic oil. A field study was conducted to determine optimum nitrogen and phosphorus fertilizer rate for some selected agronomic characteristics, essential oil yield and oil content of spearmint at Koka substation in 2014/2015 cropping season. Four nitrogen levels (0, 30, 60 and 90 kg N ha^{-1}) and four phosphorus levels (0, 10, 20 and 30 kg P ha^{-1}) were used as treatments and laid out in factorial RCBD with three replications. Analysis of variance revealed that, leaf fresh, stem fresh and leaf dry weights of spearmint were not affected significantly ($P > 0.05$) by the main effect of applied rates of inorganic N fertilizer as well as its interaction effect. Pooled mean analysis result showed that, above ground biomass and essential oil yield were significantly ($p>0.05$) influenced by different level of nitrogen, phosphorus and their interaction. The combined application of N and P fertilizers at the rate of 30 kg N and 20 kg P ha^{-1} increased spearmint above ground biomass by 29% and essential oil yield by 12% while compared to the control treatment but statistically at par between them. Generally, a fertilizer dose of 30 kg N ha $^{-1}$ and 20 kg P ha $^{-1}$ could be recommended at koka and other similar area to attain maximum above ground biomass and Essential oil yield of spearmint. Finally, this study provides useful information regarding soil and crop response for further investigation.

Key words: Nitrogen and Phosphorus fertilizer, oil content, oil yield and spearmint

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INTRODUCTION

Spearmint (*Mentha spicata* L.) is perennial aromatic plant and productive up to 15 years and primarily cultivated mainly for its aromatic oil. The essential oil is used for flavoring of different foods, alcoholic and non alcoholic drinks, gum and dental hygiene products, perfumes, hygiene products, pesticides, and pharmaceutical products as medicinal purpose. Moreover, the herbage yield can be used as tea. Spearmint leaves has a synergistic action of antioxidant phyto-chemicals, arotenoids and flavonoids. Consuming spearmint leaves used to combating oxidative stress that causes chronic disease like diabetes (Rajeshwari *et al*., 2012). Growth, development and the quality of aromatic plants are affected by the genetic background, environment and management practices as well as the processing and storage of plant tissues (Clark and Menary, 1980a).

Among many plant growth factors, the nutritional requirements of the crops are considered to be the most important factor (Singh *et al*., 1989). For healthy growth and optimal yield, nutrients must be available to plants in correct quantity, proportion and in a usable form at the right time. To fulfill these requirements, chemical fertilizers and/or organic manures are needed. Fertilization has been reported to have an influence on the phyto-nutritional quality of crops. Inorganic fertilizer is said to reduce the antioxidant levels, while organic fertilizer has been proven to enhance antioxidant content in plants (Dumas, Y.; Dadomo, M.; Di Lucca, G.; Grolier, 2003)

Nitrogen (N) is the most recognized in plants for its presence in the structure of the protein molecule. In addition, N is found in such important molecules as purines, pyrimidines, porphyrines, and coenzymes. Purines and pyrimidines are found in the nucleic acids RNA and DNA, which are essential for protein synthesis. Accordingly, N plays an important role in synthesis of the plant constituents through the action of different enzymes (Saber F.H and Khalid A. K., 2011). A high rate of N application increases leaf area development and increases overall crop assimilation, thus contributing to increased seed yield (Bhardwaj and Kaushal, 1998). Patra *et al*. (1993) reported that straw mulching significantly affected the fertilizer nitrogen use efficiency and essential oil yield in Japanese mint (*Mentha arvensis* L.) Alkire and Simon (1996) and Piccaglia et al. (1993) concluded that N increases essential oil yield of peppermint by influencing a variety of growth parameters such as tillers per plant, the total plant dry weight and the Leaf Area Index (LAI).

Other author reported that, N fertilization was reduced essential oil content in creeping juniper (*Juniperus horizontalis*) (Robert and Francis, 1986), although it has been reported to increase total essential oil yield in thyme (*Thymus vulgaris* L.) (Baranauskienne *et al*., 2003).

Phosphorus also plays an important role in various metabolic processes. It activates coenzymes for amino acid production used in protein synthesis. P also aids in root development, flower initiation, seed and fruit development. According to Mahmoud S. (2009), application of 50kg/ha of P_2O_5 significantly increased total dry matter and essential oil yield of indigenous Mint. From all these results, it can be inferred that it is essential to apply N and P fertilizer to medicinal plants supposed to be grown in soils which are deficient in these nutrients.

Given to the fact that spearmint is a recently cultivated crop in Ethiopia, there is great gap with regard to specific agronomic recommendations for the spearmint producing areas of the country. This urges that a lot has to be done on spearmint agronomic experiments so as to have appropriate spearmint crop fertilizer management recommendations. Hence, the objective of this research was to determine optimum nitrogen and phosphorus fertilizer rate for some selected agronomic characteristics, essential oil yield and oil content of spearmint.

MATERIAL AND METHODS

A field study was conducted at Koka substation in 2014/2015 cropping season. It was geographically located at 08° 26.1' North latitude, 39° 0.1’ East longitude and an altitude of 1617 m.a.s.l. The texture of the top soil (0-20cm) was sandy clay loam with PH 8.01 (1:2.5) soil water suspension. Factorial combinations of four nitrogen level (0, 30, 60 and 90 kg/ha) and four phosphorus level (0, 10, 20, 30 kg/ha) were laid out in a randomized complete block design (RCBD) with three replications. Plot size for each treatment was 4.80 m x 4m.

An auger was used to take soil samples at random from different spots to make a composite sample at depths of 20 cm before fertilizer application. A sizeable quantity of composite soil sample was air-dried and sieved through a 2 mm mesh and subjected to physical and chemical analysis including soil pH, organic carbon, total nitrogen, potassium, phosphorus, and exchangeable K, Na and Ca contents. Urea and TSP were used as source of nitrogen and phosphorous fertilizer, respectively. Full dose of phosphorous was applied as the basal dressing during transplanting. Nitrogen containing Urea fertilizer was applied in split form (1/3 during planting, 1/3 after first harvest and the remaining 1/3 after second harvest). All required manage mental practices were done as and when required.

Plants from the center were harvested by excluding border rows to collect yield and yield contributing characters such as leaf fresh weight, stem fresh weight, leaf dry weight, stem dry weight, above ground fresh biomass, essential oil content (w/w, wet/dry based) and essential oil yield analysis was done using gas chromatography-mass spectrophotometer or modified Clevenger collector apparatus. Then, collected data were statistically analyzed using SAS computer soft ware version 9.0 English and the significance difference between any two treatments means were tested by least significant difference (LSD) at 5% probability level.

RESULT AND DISCUSSION

Soil physico-chemical properties before planting

The results of physicochemical properties of the composite surface soil (0-20 cm) collected before planting showed that the textural class of the soil loam with slightly alkaline (pH 8.01) as presented in (Table 1) indicating that these properties are favorable for Spearmint production. Based on the results obtained for soil analysis, the organic carbon content (1.4%) and total nitrogen content (0.08%) of the experimental soil are low

Table 1:-Soil Physico-Chemical Properties

Location	Particle Size Analysis (%)				pH	ECE microsm^{-1}	OC	Total N	C:N	Avail
	Sand	Silt	Clay	Class	1:25		(%)	(%)	ratio	P (ppm)
Koka	42	32	26	Loam	8.01	170	1.4	0.08	17.6	18

Table 2: Main effect of nitrogen and phosphorus fertilizer rate on Leaf fresh weight, Steam fresh weight, Leaf dry weight and Essential oil content

Treatments	Parameters			
N-level	Leaf fresh weight (kg/ha)	Stem fresh weight (kg/ha)	Leaf dry weight (kg/ha)	Essential oil content
0	19210.8^{ab}	9194.5	7514.4	1.09^{ab}
30	20081.5^{a}	9548.8	7345.5	0.98^{b}
60	19816.6^{ab}	9571.4	7707.5	0.94^{b}
90	18537.7^{b}	9618.3	7036.9	1.16^{a}
LSD	1354.8	NS	NS	0.1
P-level				
0	18868.1^{b}	8867.1^{bc}	7328.1	1.02
10	19456.4^{b}	8646.9^{c}	7554.2	1.04
20	20822.6^{a}	9758.7^{ab}	7671.3	1.07
30	18499.5^{b}	10660.3^{a}	7050.7	1.04
LSD	1354.8	1012.2	NS	NS
N*P	NS	NS	NS	NS
CV	8.37	12.8	10.9	11.77

LSD= Least Significant Deference

(Table 1) in accordance with Landon (1991), who classified the organic carbon content of soil <4%, 4-10%, and >10% as low, medium and high respectively. The same author classified total nitrogen content <0.1, 0.1-0.15, 0.15-0.25 and >0.25 as very low, low, medium, and high respectively. This indicates that, the soil was deficient in nitrogen probably due to continuous cultivation and lack of incorporation of organic materials. Thus, based on the categories of soil characteristics, both nutrient values fall in the low ranges (Charman and Roper (2007). According to Landon (1991) the phosphorus content of the soil is above the critical level (8 ppm) for most crop plants. This was probably due to high amount of P fertilizer received during the past years. This indicated that P is not a limiting nutrient for optimum crop growth and yield in the experimental site. The CEC of the soil for the experimental site was 170 cmolkg^{-1} soil (Table 1), which was very high (Landon, 1991) and appropriate for crop production.

Effects of N and P on Growth Characters

The stem fresh and leaf dry weights of spearmint were not affected significantly ($P > 0.05$) by the main effect of applied rates of inorganic N fertilizer as well as its interaction effect with P (Tables 2). However, application of N had a significant effect on leaf fresh weight (Table 3). The highest leaf fresh weight was recorded from N applied at the rate of 30 kg/ha, which is statistically at par with control and 90 kg/ha application, while the lowest was recorded at highest rate of N application. However, the finding of this investigation was disagreement with most of the previous work of [Saber et al., 2014; Khalid et al., 2015; Minu et al., 2016] they reported that N fertilization in general increased the various growth characters compared with unfertilized plants.

On the contrary, the main effect of P application on spearmint leaf and stem fresh weight were significant ($P \leq 0.05$), but there was no significant effect on leaf dry weight (Table 2). The highest mean leaf fresh weight was recorded from P application at the rate of 20 kg/ha but it was statistically at par with other treatments. Similarly, the highest mean stem fresh weight was recorded from P application at the rate of 30 kg/ha but it was statistically at par with P applied at 20 kg/ha. However, the lowest stem fresh weight was obtained from P applied at 10 kg ha^{-1}, which was statistically at par with no fertilization plot. Leaf dry weight, in this particular study no significant variation were eminent in different levels of nitrogen, phosphorus and their interaction.

Pooled mean analysis result also showed that, above ground biomass was significantly ($p>0.05$) influenced by different level of nitrogen, phosphorus and their interaction. These indicate that the effects of different levels of nitrogen were affected by different levels of phosphorus fertilizer. When P and N fertilizers interaction

Table 3: Pooled mean comparison of above ground biomass and Essential oil yield as affected by the interaction effects of nitrogen and phosphorus fertilizer rates

Nitrogen Level	Above ground biomass				Essential Oil Yield			
	Phosphorus				Phosphorus			
	0	10	20	30	0	10	20	30
0	24504^{cd}	26020^{cd}	25778^{cd}	26562^{bcd}	63.37^{abc}	61.28^{bcd}	54.32^{cdef}	64.30^{ab}
30	25195^{cd}	26315^{cd}	30110^{a}	26245^{cd}	51.13^{efg}	52.97^{defg}	71.07^{a}	45.86^{fg}
60	24965^{cd}	25471^{cd}	26826^{bc}	29572^{ab}	51.53^{efg}	52.73^{defg}	58.56^{bcde}	44.38^{g}
90	25685^{cd}	25690^{cd}	27159^{abc}	23759^{d}	60.77^{bcd}	59.2^{bcde}	63.12^{abc}	63.02^{abc}
LSD	*				**			
	6.92				9.62			

increased from 0N x 0P to 30 kg N x 20 kg P ha^{-1} biomass yield was significantly increased ($P < 0.05$) from 24504 to 30110 kg ha^{-1} (Table 3). However, when it increases further the biomass yield decreases significantly. Besides, compared to the control treatment, the combined application of N and P fertilizers at the rate of 30 kg N and 20 kg P ha^{-1} increased spearmint above ground biomass yield by 29% followed by 21% and 11% which were obtained from the treatment combinations of 90 kg N x 30 kg P ha^{-1} and 90 kg N x 20 kg P ha^{-1}, respectively, while statistically at par between themselves. The lowest biomass yield (24504 kg ha^{-1}) was recorded for control. Previous research finding showed that application of 75/50 kg/ha of NP_2O_5 gave highest total dry matter yield of Indigenous mint (Mahamoud *et.al* 2009).

Effects of N and P fertilizers on Essential oil yield and Essential oil content

As shown in Table (2) P levels had no significant effect on essential oil content (%) of spearmint. On the other hand, N levels had a pronounced effect on essential oil content (%) of spearmint. The highest percent of essential oil content for spearmint was recorded with the application of 90 kg ha^{-1} N fertilizer; while the lowest average essential oil content was recorded from 60 kg ha^{-1} N received plots. Maximum percent of essential oil content (1.09) was observed by application of N fertilizer at the rate of 90 kg ha^{-1} that differed none significantly from control plots. The result also showed that the increasing rate of fertilizer application decreased the essential oil content relative to control. Similar result was reported for mint (Mahamoud *et al.,* 2009). On the other hand, the results of this investigation was disagreement with the works of [Saber et al., 2014; Khalid et al. 2015] they reported that N fertilization increased the vegetative growth and essential oil content of *Nigella sativa* L. and Lovage plants.

N and P fertilizers interaction effect were also highly significant for essential oil yield (Table 3). When N and P fertilizers interaction increased from 0N x 0P to 30N x 20P kg ha^{-1} essential oil yield was significantly increased ($P < 0.01$) from 51.13 to 71.07 kg ha^{-1}. However, when it increases further the essential oil yield decreases significantly. Besides, the maximum essential oil yield increases were observed in 30 kg N ha^{-1} in combination with 20 kg P ha^{-} (71.07 kg ha^{-1}) followed in this order by the combined applications of 0 kg N and 30 P ha^{-1} (64.30 kg ha^{-1}), 0 kg N and 0 kg P ha^{-1} (63.37 kg ha^{-1}), 90 kg N and 20 kg P ha^{-1} (63.12 kg ha^{-1}), and 90 kg N and 30 kg P ha^{-1} (63.02 kg ha^{-1}) whilst no significant differences were observed between themselves.

The lowest essential oil yield (44.38 kg/h) was observed at 60N*30P kg/ha fertilizers application. In general in the present study, no significant difference was observed in essential oil yield from different rates of N fertilizer application compared to control. Similar trend was also observed in essential oil content of spearmint in different fertilizer application. Besides, the maximum essential oil yields of 71.07 kg/ha obtained in the present study is slightly lower than the yields reported by (Mahamoud *et al.,* 2009).

CONCLUSION

Generally, fertilization is an important part of the overall management of mint and other crops. Application of different N and P fertilizer rates at different growth stage in this study did not bring significant effect on leaf fresh, stem fresh and leaf dry weights of spearmint compared with no fertilization. However, above ground biomass and essential oil yield were significantly ($p>0.05$) influenced by different level of nitrogen, phosphorus and their interaction. The combined application of N and P fertilizers at the rate of 30 kg N and 20 kg P ha^{-1} increased spearmint above ground biomass by 29% and essential oil yield by 12% while compared to the control treatment but statistically at par between them.

Hence, detail and further investigation over the factors associated with N availability and responsiveness of the crop such as choice of appropriate N fertilizer, timing and method of application, climatic features, soil characteristics and crop management practices are suggested. Further research in improving soil N and organic matter through addition of different organic fertilizers and evaluation of their economic benefit is required. A fertilizer dose of 30 kg N ha^{-1} and 20 kg P ha^{-1} could be recommended at koka and other similar area to attain maximum above ground biomass and Essential oil yield of spearmint. Finally, this study provides useful information regarding soil and crop response for further investigation.

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