EFFECT OF SOURCES AND LEVELS OF NITROGEN ON PARTITIONING OF ANDROGRAPHOLIDE IN KALMEGH [*ANDROGRAPHIS PANICULATA* (BURM. F.) WALL. *EX* NEES.]

S.K. CHAUHAN AND GYANENDRA TIWARI*

AICRP on Medicinal and Aromatic plants, Jawaharlal Nehru Agriculture University, College of Agriculture, Indore–152001 (M.P.) India

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SUMMARY

The impact of different nitrogen sources and levels on andrographolide accumulation and partitioning was investigated in *Andrographis paniculata* (Burm. F.) Wall. *ex.* Nees. Nitrogen application @ 40 kg/ha resulted in maximum active principle (Andrographolide) accumulation in roots and above ground plant parts. Significantly maximum andrographolide yield was noted with 40 kg N/ha in the form of castor cake (67.40kg/ha) and the minimum in control (31.50 kg/ha), while biomass yield was maximum with 40 kg N/ha supplied as urea (2185.19 kg/ha) and minimum in control (1601.85 kg/ha).

***Key words*: Accumulation, active principle, andrographolide.**

INTRODUCTION

Kalmegh [*Andrographis paniculata* (Burm. F.) Wall *ex* Nees.] is an important medicinal plant and useful in liver troubles, malaria and blood purification (Sharma 2001). It is one of the household medicine known 'Alui' extensively used particularly in West Bengal for general debility and certain forms of dyspepsia amongst adults and infants (Kirtikar *et al.* 1980). Andrographolide is extracted in Phramacopy from whole plant. Least is known about the production technology and physiological basis of its active principle partitioning. Andrographolide and Kalmeghin are two bitter organic principles, found in Kalmegh (Sharma 2001). Active principle content of any medicinal plant has its relevance on indigenous and global market price of end products. Export price of kalmegh is based on percentage of active principle in total raw material and other quality parameters. Nitrogen nutrition is known as important factor in controlling metobolism of organic complexes in medicinal and aromatic plant species. In order to boost up bitter principle production and partitioning in Kalmegh, attempt has been made to study the influence of source and levels of nitrogen application on andrographolide content in different plant parts.

MATERIALS AND METHODS

A field experiment was carried out in kharif season 2000-2001 at Research Farm, AICRP (All India Co-ordinated Research Project) on Medicinal and Aromatic Plants, J.N.K.V.V., College of Agriculture, Indore (M.P.) The soil was shallow black with pH 8.0, 9.6 kg/ha N, 9.6 kg/ha P_2O_5 and 663 kg/ha K_2O availablity. Experiment was laid out in factorial randomized block design with eleven treatment combinations in three replications. Five nitrogen sources [F.Y.M. (Farm Yard Manure), Neem cake, Groundnut cake, Castor cake and Urea] and two nitrogen levels (20 and 40 kg/ha) and control (No Nitrogen) were used. The crop was transplanted on 15th July, 2000. Full dose of Nitrogen by organic manures was applied at the time of field preparation and urea was applied after transplanting.

Plants were harvested at the time of physiological maturity for andrographolide analysis. One square meter

*Corresponding author's present address : Department of Plant Physiology, J.N. Agriculture University, Jabalpur-482004, M.P.

randomly selected area per plot was used for obtaining plant samples. Twenty plants were uprooted with intact roots and partitioned into above and underground parts. Biomass production was calculated on the basis of biomass production per plot. Harvesting of crop for biomass yield was done at ground level.

The andrographolide content of all treatment combinations was estimated by solvent extraction method (Honda and Sharma, 1990). Dried and powdered sample (5 gm) was extracted successively with 4x400 ml each of petroleum ether (60-80°C), chloroform and methanol and concentrated to about 500 ml. The solution was treated with charcoal for 24 hrs, filtered and filtrate was concentrated to about 300 ml and kept overnight for crystallization. The crystals were collected by filtration. The recovered charcoal was refluxed with methanol for 2 hrs, filterd and combined the filterate with the mother liquor obtained earlier. It was concentrated to about 500 ml and left overnight for crystallization. The crystals were purified by recrystallization from methanol to get andrographolide. Andrographolide yield of above ground plant parts (kg/ha) was computed by multiplying andrographolide content (%) to biomass production (kg/ha) for each treatment. Andrographolide content as affected by different treatments were analysed statistically as per procedure described by Panse and Sukhatme (1976).

RESULTS AND DISCUSSION

Andrographolide content in dry roots was significantly higher (1.327%) at 40 kg N/ha supplied as castor cake (T_8). It was higher than the same amount of N when supplied in the from of urea, groundnut cake and FYM (Table 1). Andrographolide content was minimum in control *i.e.* no nitrogen application (0.90%). Similarly, andrographolide content in aerial parts (shoot) was maximum (3.11%) at 40 kg N/ha as caster cake (T_8) and minimum in control (1.894%). Thus partitioning of andrographolide differed due to different treatments. Increased nitrogen levels through sources resulted in increased ratio of Andrographolide accumulation in aerial parts as compared to underground parts. Hence there was significant influence of nitrogen nutrition on active principle metabolism. Malnourished plants grown in nitrogen deficit plots have disturbed production and

Table 1. Effects of different treatment on andrographolide content (%) in aerial and edaphic plant parts

Treatment	Andrographolide content in root (% of biomass)	Andrographolide content in shoot (% of biomass)	Ratio of andrographolide content between root and shoot (biomass basis)
20 kg N/ha through f.y.m.	1.037	2.457	0.422
40 kg N/ha through f.y.m.	1.117	2.681	0.417
20 kg N/ha neem cake	1.050	2.393	0.439
40 kg N/ha neem cake	1.060	2.525	0.420
20 kg N/ha groundnut cake	1.080	2.368	0.456
40 kg N/ha groundnut cake	1.200	2.901	0.414
20 kg N/ha Castor cake	1.160	2.380	0.462
40 kg N/ha castor cake	1.327	3.110	0.427
20 kg N/ha urea	1.100	2.402	0.458
40 kg N/ha urea	1.220	2.846	0.428
Control (no application)	0.900	1.894	0.475
S E m ±	0.071	0.105	0.014
CD at 5%	0.209	0.310	0.040

distribution. In addition, the active principle content of roots was comparatively higher than above ground parts probably due to reduced energy levels for translocation (Table 1). A significant effect of different nitrogen levels on andrographolide content was therefore observed. These results corroborate with the findings of Muniramappa *et al.* (1997).

Treatment comprising of 40 kg N/ha through groundnut cake had partitioned maximum active principle towards aerial parts (58.6%) as compared to underground parts *i.e.* roots (41.4%) while in control higher active principle had partitioned towards underground parts i.e. roots (47.5%) as compared to aerial parts (52.5%) (Table 1). When the nitrogen level increased from 20 to 40 kg/ha, the andrographolide partitioning changed towards aerial parts from roots. Groundnut cake enhanced andrographolide accumulation in aerial parts as compared to roots. The content was decreased when nitrogen source was synthetic (urea) but difference due to sources of N supply did not produced significant effect.

A marked difference in biomass production (kg/ha) due to nitrogen doses was observed (Table 2). Both sources and levels of nitrogen application affected andrographolide yield. 40 kg N/ha through urea produced maximum biomass and was *at par* with 40 kg N/ha supplied through any other source of nitrogen. However, 40 kg N/ha showed significant difference in biomass yield as compared to 20 kg N/ha and control. Andrographolide yield (q/ha) showed significant response to both source and level of nitrogen. The maximum andrographolide yield was noted with treatment combination comprised of 40 kg N/ha through castor cake (67.40 kg/ha). The minimum andrographolide yield was noted with control (31.50 kg/ha). It is very interesting to note that sources of nitrogen supply did not produced significant difference in andrographolide content both in root and shoot (Table 1), but produced remarkably significant difference in andrographolide yield (q/ha). These results may be due to the cumulative effects of nitrogen sources in enhancing biomass productivity along with increased andrographolide content (%) in shoot.

Source of nitrogen supply is therefore, very important as for as total andrographolide yield (q/ha) is concerned. The best source of nitrogen application was found to be the castor cake for realization of maximum andrographolide yield. This indicates that organic sources will be the most important input in enhancing the yield of secondary metabolites used for medicinal purpose.

It is proposed that physiology of active principle metabolism and accumulation in medicinal plant species need indepth investigation. This will unfold the posibilities of higher active principle productivity-potential realization through economically profitable, commercially viable, socially acceptable and eco-friendly production technology.

Table 2. Effect of different treatments on biomass productivity (kg/ha) and andrographolide yield (kg/ha) in aerial parts

Treatment	Biomass productivity (kg/ha)	Andrographolide yield (kg/ha)
20 kg N/ha through f.y.m.	1916.67	47.09
40 kg N/ha through f.y.m.	2064.81	55.35
20 kg N/ha neem cake	1888.89	45.20
40 kg N/ha neem cake	2092.59	52.85
20 kg N/ha groundnut cake	1925.93	45.60
40 kg N/ha groundnut cake	2120.37	61.51
20 kg N/ha castor cake	1935.19	47.52
40 kg N/ha castor cake	2166.67	67.40
20 kg N/ha urea	2018.52	48.50
40 kg N/ha urea	2185.19	62.20
Control (no application)	1601.85	31.50
S E m ±	81.66	0.094
CD at 5%	240.83	0.260

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