Pak. J. Bot., 41(1): 41-51, 2009.

MINERAL NUTRIENT COMPOSITION OF DIFFERENT ECOTYPES OF WHITE CLOVER AND THEIR NUTRIENT CREDIT TO SOIL AT RAWALAKOT AZAD JAMMU AND KASHMIR

M. KALEEM ABBASI[1*], MAJID MAHMOOD TAHIR[1], ASAD HUSSAIN SHAH[2] AND FARHAT BATOOL[3]

[1]Department of Soil and Environmental Sciences, Faculty of Agriculture, University of Azad Jammu and Kashmir, Rawalakot, Azad Jammu and Kashmir, Pakistan
[2]Department of Plant Breeding and Molecular Genetics, University of Azad Jammu and Kashmir, Faculty of Agriculture, Rawalakot Azad Jammu and Kashmir Pakistan.
[3]Neurochemistry and Biochemical Neuropharmacology Research Laboratory, Department of Biochemistry, University of Karachi, Karachi-75270, Pakistan

Abstract

In comparison with other legumes and grasses, white clover provides forage with high nutritional value and quality. A field experiment was conducted by growing three indigenous and three exotic ecotypes of white clover in the mountain region of Himalayan i.e. the State of Azad Jammu and Kashmir, Pakistan. Plant nutrient content of N, P, K, Ca, Mg, Fe, Mn, Cu and Zn were determined in the forage of white clover while soil nutrient content of N, C, P, K, Ca and Mg were determined in the soil under white clover. Pure grass and soil without white clover were used as control/check. Mineral nutrient contents in white clover were substantially higher than those found in the grass. Among macronutrients, the content of P and K were 0.32 and 1.96%, while the contents of N, Ca and Mg were 2.61, 1.10 and 0.29%, 2−3 fold to the content found in the grass. Micronutrients also showed similar trend and the concentration of Fe, Mn, Cu and Zn were 192, 86, 31 and 49 mg kg^{-1}, almost double than those in the grass. The ratio between Ca/P, Ca/Mg, Ca/K, K/Mg, K/P was 3.9, 4.0, 0.6, 6.8 and 6.2, respectively showing positive balance of mineral nutrient elements in white clover. Growing over two years in the field, white clover increased substantially the concentration of C, N, P and K in soil indicating its potential for improving the nutrient status of the soil. Introduction of white clover in our pastoral ecosystem can provide forage with high nutritional value that can result in increased levels of animal production and allow for high stocking rates.

Introduction

Introduction and establishment of forage legumes in agro-ecosystem are important because they enrich the N content of the soil; mobilize other nutrients and also have a high nutritive value. In addition, the quantity as well as quality of herbage production can be substantially increased. The use of improved legumes, particularly white clover (*Trifolium repens* L.), has long been considered an effective way of improving pasture production and soil N status in many parts of the world (Gibson & Cope, 1985; Ledgard & Steele, 1992). In comparison with other legumes and grasses, white clover provides forage that is very high in nutritive value (Ayres *et al.*, 1998). This superior quality of white clover is due to high levels of crude protein, soluble carbohydrates, minerals (especially calcium, phosphorus, magnesium) and low levels of structural carbohydrates and lignin (Ulyatt *et al.*, 1977; Thomson, 1984). The high nutritive value of white clover-based pastures results in increased levels of animal production and allows for high stocking rates. A number of experiments comparing animal production from white clover v. grass have shown that animals perform significantly better on white clover than grass diets (Rogers *et al.*, 1982; Thomson *et al.*, 1985). A summary of these results indicates an average improvement in live weight gain of about 65% in lambs and about 30% in beef cattle. Milk production in dairy cows was increased substantially with up to 900 kg more milk produced per lactation from cows grazing pure white clover (Wilkins & Munro, 1988).

*Corresponding author E-mail: kaleemabbasi@yahoo.com

There is acute shortage of fodder especially green nutritious fodder in the mountain region of Himalayan (Roy *et al*., 1989) which is the major cause of low productivity of the livestock in this region. Misri (1997) studied the biomass availability of some of the representative pastures of Kashmir Himalaya and found that green herbage availability varied from 4.7−29.1 t ha^{-1}. Ram & Singh (1994) observed that biomass availability varied from 1.62−3.96 t ha^{-1} (green herbage) in Himalayan pasture of Uttar Pradesh, India. Melkania & Singh (1989) have estimated that net above ground biomass varied from 279−1568 gm^{-2} for low elevation Himalaya, 219−285 gm^{-2} for mid elevation Himalaya and 233−372 gm^{-2} for high elevation Himalaya. Indigenous legumes such as clovers (*Trfolium pratens, T. repens)*, have proved successful in Indian Kashmir valley (Gupta, 1977) while successful growth and yield of white clover in the State of Azad Jammu and Kashmir has already been reported in our previous study (Abbasi & Khan, 2004). It was found that the annual herbage yield (fresh fodder) of white clover and clover+grass mixture harvested twice during the season was 7–15 t ha^{-1} while dry matter yield was 3–6 t ha^{-1}. Similarly, the protein content of white clover was 16% as compared to 5% in the indigenous grass species demonstrating the superior feed value of white clover (Abbasi & Khan, 2004). The aim of the present investigation was to examine the mineral nutrient content of indigenous and exotic ecotypes of white clover for their nutritional value. In addition, the mineral nutrient contents of soil under white clover were also determined to examine the nutrient credit of white clover to the soil.

Materials and Methods

1. The study site: The study area (Rawalakot) lies between the altitude of 1800–2000 m above sea level and latitude 33–36^0 in the north–east of Pakistan under the foothills of great Himalayas at Rawalakot district, Poonch division, Azad Jammu and Kashmir (AJK). The topography of the area is mainly hilly and mountainous with valleys and stretches of plains. The study area is characterized by a temperate sub-humid climate with annual rainfall ranging from 500−2000 mm (depending on season), most of which is irregular and falls as intense storms during the monsoon and some times in winter. Mean annual temperature is about 22 ^{0}C (maximum) in summer while winter is fairly cold with temperature ranging even below freezing point. Agriculture is based on rainfed cropping system and maize (*Zea mays* L.) is the favored crop of the region. Vegetables and fruit trees predominates and most important fruits are apple (*Pyrus melus*); pears (*Pyrus communis*); apricots (*Prunus ameriaca*); plums (*Prunus domestica*) and walnuts (*Juglans regia*). A large proportion of the area consists of unculturable waste including forest. The important forest species are deodar (*Cedrus deodara*); kail (*Pinus excelsa*); cheer (*Pinus willichiana*); sufaida (*Populus euramericana*) and spruce (*Abies pindro*). The few important grass species found in the area are *Cenchrus ciliaris* (Buffel grass), *Digitari sanginalis, Setaria pallide, Poa pratensis* and A*runda donax*. Detailed soil survey and soil profile studies in this area have not been done.

2. Establishment of white clover in the field: An area of 23.5 m x 11 m was selected in the research farm, University of Azad Jammu and Kashmir, Faculty of Agriculture, Rawalakot Azad Jammu and Kashmir (AJK). The field was ploughed twice by tractor; weeds and grass were removed manually. There were six ecotypes with five replications. Therefore, the main plot was divided into 30 equal blocks of 2.7 x 4.9 m each. Seeds of indigenous white clover were collected from three different locations of the Poonch division near Rawalakot. The sites were Rawalakot, Banjosa and Tillipir. Seeds of three exotic ecotypes viz., Haifa, NuSiral and Irrigation were collected from New South Wales

Agricultural Research and Advisory Station, PMB, Glen Innes, NSW, Australia. After preparing proper seed beds/sub-plots and making sure the proper moisture level, seeds of each ecotype were sown at about 3-kg ha^{-1} in February 2004. After germination, weeds/grasses were eradicated and only clover was allowed to grow as a monoculture crop.

3. Plant nutrient contents: Four to five healthy and vigorous plants of each ecotype from three central rows of each plot were harvested just before flowering in July 2005. Plants were washed with distilled water, air dried and then oven dried at 65^0C till constant weight. The plant material was milled and thoroughly mixed and stored at 4^0C before analysis. Both macro and micronutrients (Fe, Mn, Cu, Zn) were determined in white clover plants while pure grass collected from the same location was used as control. Total N contents were determined using the micro-Kjeldahl method of Keeney & Nelson (1982). Wet combustion in a 2:1 mixture of HNO_3 and $HClO_4$ was used for mineral element analysis of samples. The phosphorus (P) content was determined by the vanadomolybdate yellow color using spectrophotometer. The calcium (Ca) and magnesium (Mg) contents were determined using an atomic absorption spectrophotometer; potassium (K) content was determined using flame photometer while Fe, Mn, Cu and Zn contents were determined by atomic absorption spectrophotometer. After mineral element contents (elemental form) determined, the K/P, Ca/P, Ca/Mg, Ca/K and K/Mg ratios were calculated.

4. Soil nutrient contents: Soil samples were collected from 0–15 cm depth at random from 3–5 locations from each plot. Composite samples were prepared accordingly. The sample was air dried and sifted (2 mm mesh) to eliminate coarse rock and plant material. Soil sample was stored in cool and dry place until used. Samples (triplicate) were analyzed for total soil organic carbon (C), total N and pH. Soil organic carbon (SOC) was determined using a modified Mebius method (Nelson & Sommers, 1982). Total N was determined by the Kjeldahl's digestion, distillation and titration method (Bremmer & Mulvaney, 1982), available P by the Olsen extraction method (Olsen & Sommers, 1982) and available K was extracted with 1N Ammonium acetate, adjusted to pH 7 and was determined flamephotometrically. Soil pH was determined by a glass electrode on 1:1 (v/v) soil:water suspension after a 1 hour equilibration period with occasional stirring. The AB-DTPA test was used to determine Ca, Mg, Fe, Mn, Cu and Zn using Atomic absorption spectrophotometer model AANALYST-700.

The results were analyzed and comparison was made on the basis of Standard Error of Means (SEM) using the Microsoft Office Excel 2003.

Results

Mineral nutrient contents of white clover were significantly higher than those found in pure grass. The N content of different ecotypes of white clover varied between 2.21% to 3.0% while N content in grass was 0.85% (Table 1). The average N content in white clover (indigenous and exotic ecotypes) was 2.61% (Fig. 1) showing that white clover had three times more N than the N content of grass. The P and K content of white clover ecotype varied between 0.21 to 0.44% and 1.44 to 2.9%, respectively relative to 0.18 and 0.88% of grass (Table 2). On average, P and K content of white clover were 0.32% and 1.96% (Fig. 1) indicating a 75 and 123% more P and K in white clover than in the grass. Likewise, Ca and Mg also showed similar trend observed for P and K. However, the level of increase in both the nutrients was very high than that found for P and K. Indigenous ecotypes exhibited higher values i.e., 30, 42, 33 and 22% for N, P, K, Mg than the exotic ecotypes. However, the Ca content of exotic ecotypes was 23% higher than the Indigenous ecotypes.

Table 1. Concentration of different mineral elements in exotic and indigenous ecotypes of white clover (plant samples) grown over two years (after final harvest).

Treatments	N (%)	P (%)	K (%)	Ca (%)	Mg (%)	Fe (mg kg^{-1})	Mn (mg kg^{-1})	Cu (mg kg^{-1})	Zn (mg kg^{-1})
Control (Grass)	0.85	0.18	0.88	0.36	0.11	158	52	14	25
Rawalakot	2.9	0.38	1.96	1.35	0.33	156	92	40	50
Banjosa	3.0	0.29	1.86	0.99	0.28	250	70	34	43
Tollipir	2.94	0.44	2.90	0.53	0.34	180	76	28	49
Irrigation	2.32	0.21	1.44	1.51	0.27	192	110	28	48
NuSiral	2.29	0.26	1.82	1.15	0.31	192	92	23	51
Haifa	2.21	0.31	1.78	1.08	0.21	180	74	30	50
SEM	0.30	0.04	0.25	0.17	0.14	16.33	7.67	3.35	3.78

*SEM = Standard Error of Means (n=6)

Table 2. The Ca/P, Ca/Mg, Ca/K, K/Mg and K/P ratios in forages of grass and white clover.

Treatments	Ca/P	Ca/Mg	Ca/K	K/Mg	K/P
Grass	2.0	3.3	0.4	8.0	4.9
Rawalakot	3.6	4.1	0.7	5.9	5.2
Banjosa	3.4	3.5	0.5	6.6	6.4
Tollipir	1.2	1.6	0.2	8.5	6.6
Irrigation	7.2	5.6	1.0	5.3	6.9
NuSiral	4.4	3.8	0.6	6.1	7.0
Haifa	3.5	5.1	0.6	8.5	5.4

Table 3. Concentration of different mineral elements and pH of the soils under exotic and indigenous ecotypes of white clover grown over two years (after final harvest).

Treatments	C (g kg^{-1})	N (g kg^{-1})	C:N ratio	P (mg kg^{-1})	K (mg kg^{-1})	pH
Control	4.93	0.66	7.47	1.12	21.6	7.82
Rawalakot	7.60	1.05	7.26	2.36	41.0	7.67
Banjosa	7.66	0.92	8.35	1.92	38.0	7.45
Tollipir	7.80	0.99	7.85	1.65	37.3	7.48
Irrigation	7.95	0.86	9.24	1.59	34.7	7.45
NuSiral	7.02	0.85	8.22	1.33	35.9	7.55
Haifa	6.82	0.82	8.32	1.23	32.7	7.46
SEM	0.64	0.05	0.90	0.23	1.84	0.21

*SEM = Standard Error of Means (n=6)

The micronutrient contents in white clover and pure grass are also presented in Table 1. Iron content in white clover varied between 156 to 250 mg kg^{-1} while Mn content was in the range between 70 to 110 mg kg^{-1}. Similarly, Cu and Zn content were 23 to 40 mg kg^{-1} and 43 to 51 mg kg^{-1}, respectively. The corresponding values for grass were 118, 52, 14 and 24 mg kg^{-1} respectively. The average values of micronutrients between grass and white clover showing that micronutrient contents in white clover were significantly higher than those found in the grass (Fig. 2).

Mineral elements present in the forage of white clover and grass have certain ratios. The ratios calculated in the present investigation were Ca/P, Ca/Mg, Ca/K, K/Mg and K/P (Table 2). The Ca/P, Ca/Mg, Ca/K, K/Mg and K/P for grass were 2.0, 3.3, 0.4, 8.0 and 4.9. The Ca/P for different ecotypes of white clover varied between 1.2 to 7.2, Ca/Mg 1.6 to 5.6, Ca/K 0.2 to 1.0, K/Mg 5.3 to 8.5 and K/P 5.2 to 7.0. Exotic ecotypes exhibited higher ratios than the indigenous ecotypes except for K/P. The average ratios between different elements showed that white clover exhibited higher values than the grass except for K/Mg (Fig. 3). Percent increase in Ca/P, Ca/Mg, Ca/K, and K/P in white clover over grass was 95, 22, 37, 27 and 75% while the ratio of K/Mg in grass was 15% higher than the white clover.

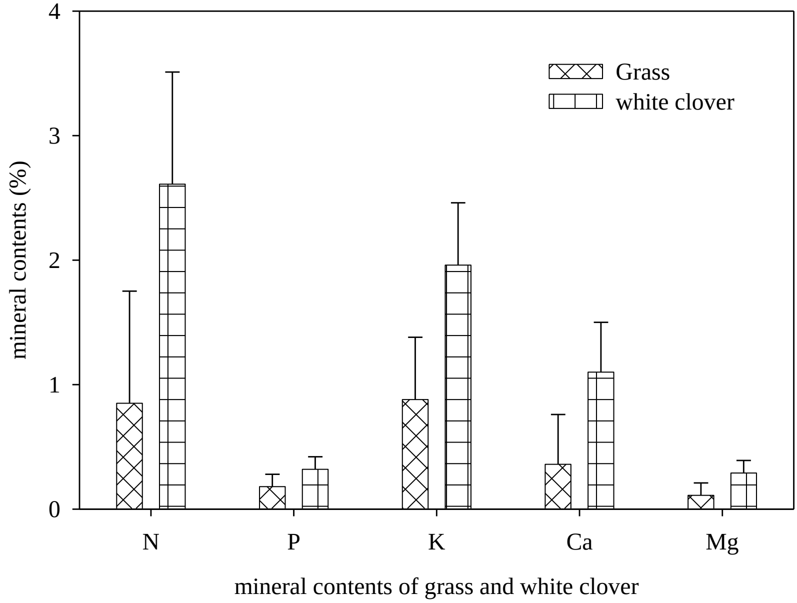

Fig. 1. Mineral nutrient content i.e., macronutrients in the forage of white clover (average of 05 ecotypes on dry weight basis) and grass grown over two years. Vertical lines in each bar indicate Standard Error of Means (SEM).

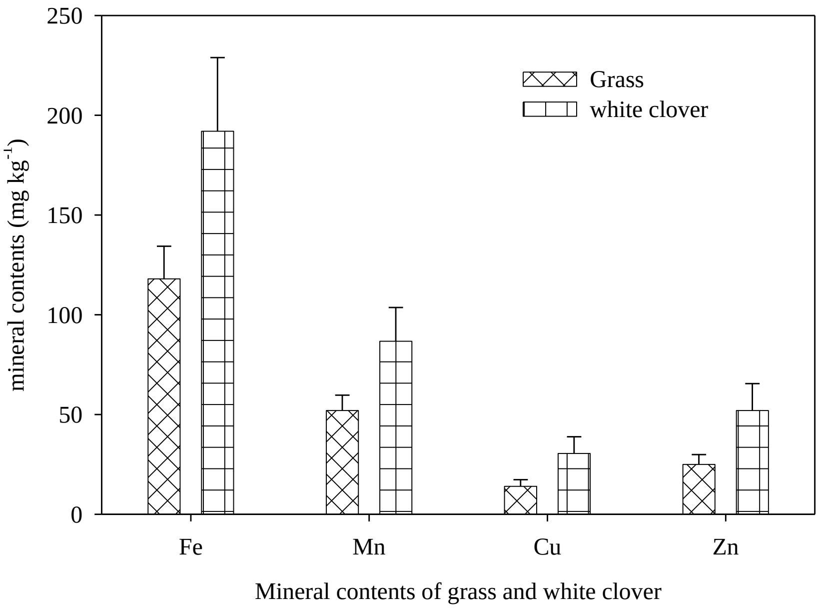

Fig. 2. Mineral nutrient contents i.e., micronutrient in the forage of white clover (average of 05 ecotypeson dry weight basis) and grass grown over two years. Vertical lines in each bar indicate Standard Error of Means (SEM).

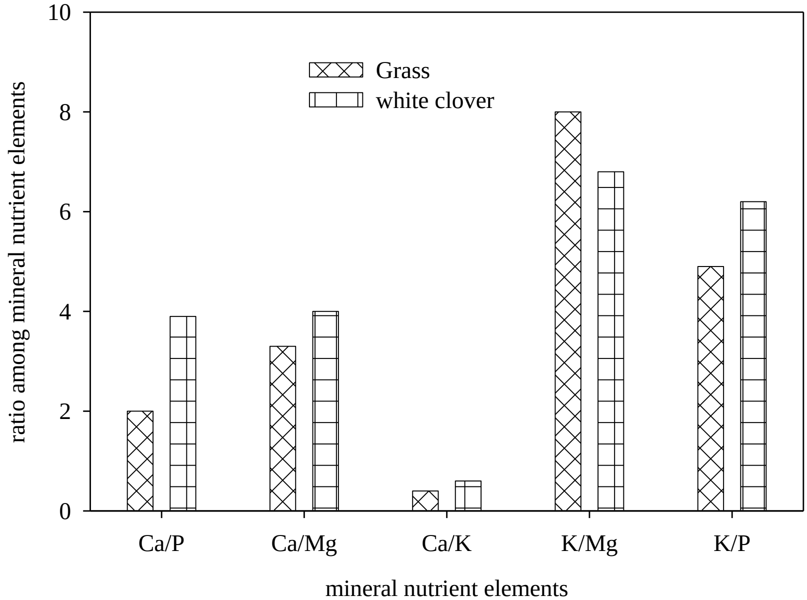

Fig. 3. The Ca/P, Ca/Mg, Ca/K, K/Mg and K/P ratios in forages of white clover (average of 05 ecotypes on dry weight basis) and grass.

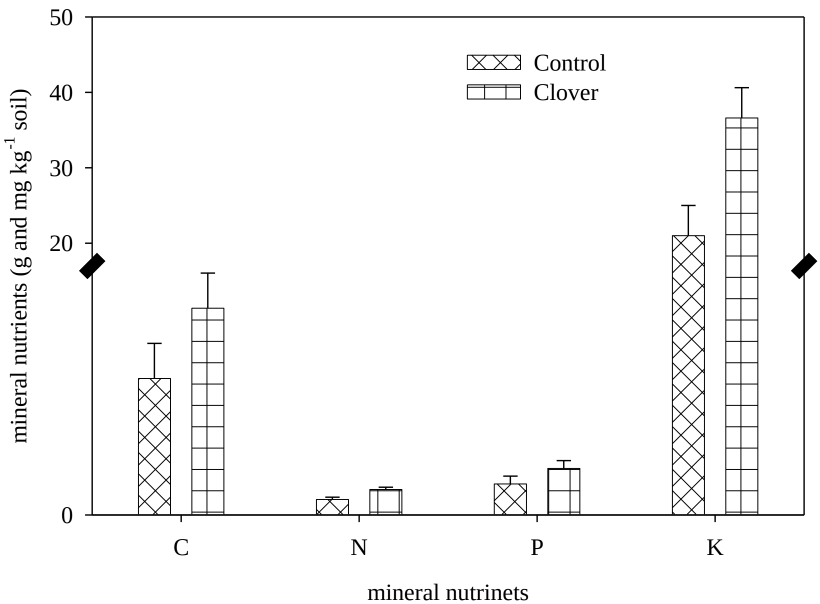

Fig. 4. Changes in mineral nutrient content i.e., macronutrients of soil following the cultivation of white clover (average of 05 ecotypes) and in soil without white clover (control). Vertical lines in each bar indicate Standard Error of Means (SEM, *n*=06).

Both indigenous and exotic ecotypes of white clover significantly increased the mineral nutrient content of the soil. Table 3 indicates the changes in the concentrations of different mineral nutrient elements i.e., C, N, C:N ratio, P, K including pH following the introduction of white clover in soil. The C and N content in the control soil without white clover were 4.93 and 0.56 g kg^{-1}, respectively. Growing of white clover over two years increased the content of C and N to a maximum of 7.95 and 1.05 g kg^{-1}. Different ecotypes of white clover did not show any significant difference for C but had significant difference for N. Soil under the ecotypes collected from Rawalakot and Tollipir had significantly more N than the others. By taking the average values (soils under six ecotypes of white clover), the C and N content of white clover growing soils were 7.47 and 0.92 g kg^{-1}, respectively (Fig. 4) indicating 52 and 62% increase over control. The C:N ratio for control soil was 6.54 while soils under white clover had C:N ratios in the range of 7.85 to 9.24 with an average value of 8.2 i.e., the increase in C:N ratio over control was 25%. Phosphorus and potassium concentration in the soil without white clover were 1.12 and 21.6 mg kg^{-1} while soils with white clover had significantly more P and K ranging from 1.23 to 2.36 mg kg^{-1} for P and 32.7 to 41.0 mg kg^{-1} for K concentration. Among different ecotypes, P and K concentration was highest (significant) in soil with ecotype Rawalakot while no significant difference was observed among other ecotypes. Figure 4 indicate the comparison between the average values of white clover soils with soil without white clover. The mean concentration of P and K for soils with white clover was 1.68 and 36.6 mg kg^{-1} indicating an increase of 50 and 69% over control.

Soil under white clover showed a significant decrease in soil pH. The pH of the control soil was 7.82 while soils with white clover had pH in the range of 7.67 to 7.45 indicating 0.2 to 0.4 unit decline in pH. Among white clover ecotypes, soil with ecotype Rawalakot had relatively high pH while the remaining soils did not show much difference. The average pH value for different soils covered with white clover was 7.51 compared to pH of 7.82 of the control soil showing a 0.3 unit decrease in pH due to the presence of white clover.

Discussion

The study confirmed that the white cover has better chemical composition and higher nutritional value in comparison with pure grass. The N, P, K, Ca and Mg contents of white clover were 2.61, 0.31, 1.96, 1.10 and 0.29%, substantially higher than those found in the grass. Our results were similar to those reported by Hoveland (2000) who found 0.4, 1.5 and 0.4% of P, Ca and Mg in white clover compared to 0.2, 0.3 and 0.2% in Bermudagrass. The higher content of N in white clover reflected higher protein content that can increase animal performance on pasture simply due to increased nutrient density (Hoveland, 2000). Similarly, the higher P and K content will increase the forage quality that would affect the meat and milk quantity and quality. In addition, the higher level of magnesium in clovers decreases the potential risk of grass tetany in the spring months (Tekeli & Ates, 2005). Likewise, the micronutrient contents were also significantly higher in white clover than pure grass. The Fe, Mn, Cu and Zn content in white clover (average) were 192, 86, 31 and 49 mg kg^{-1} relative to 118, 42, 14 and 25 mg kg^{-1} in the grass showing that micronutrients in white clover were almost double than the grass. Relatively low content of Cu than the other micronutrients is attributed to the possible retention of Cu in the roots. Jarvis (1980) reported that roots of ryegrass and white clover retained more than 71% of applied Cu when the plants were grown in flowing nutrient solution. The critical levels of these mineral elements in white clover reported by Dunlop & Hart (1987) were; 4.5−5.5%, 0.1−0.25%, 0.8−1.1%, 0.5−1.0%, 0.12−0.14% for N, P, K, Ca and Mg, respectively while the critical levels for Fe, Mn, Cu and Zn were 50−70, 20−35, 4−6 and 12−18 mg kg^{-1}. Except N, all mineral nutrient elements in white clover were 2−6 times higher than the critical levels reported above; showing high quality and nutritional value of white clover grown under local conditions of Rawalakot AJK.

In general, higher mineral nutrient content in white clover would have two-fold effect, i) affect on the mineral nutrient content of soil following its addition to the soil as root exudates, dead leaves and fragments of roots or when it is used as green manuring, ii) effect on animal health and production when it is used as pasture. In a pasture, nutrients are taken up by plants from litter and soil, and are subsequently transferred to grazing animals. It was found that animals perform significantly better when animal production was compared from white clover v. grass (Rogers *et al.*, 1982; Thomson *et al.*, 1985). A summary of these results indicates an average improvement in live weight gain of about 65% in lambs and about 30% in beef cattle (Wilkins & Munro, 1988). Wilkins & Munro (1988) reported milk production in dairy cows was increased substantially with up to 900 kg more milk produced per lactation from cows grazing pure white clover. Animal body usually contained about 1.5−5% of mineral nutrient elements (Tekeli *et al.*, 2003). Tekeli & Ates (2005) reported that the requirement for major mineral nutrients for gestating beef cows or lactating beef cows is 0.60−0.80% (w/w) for K, 0.18−0.44% for Ca, 0.18−0.39% for P and 0.04−-0.10 % for Mg. Our results indicated that white clover grown in local conditions had mineral nutrients sufficient enough to fulfill the above requirement of the animals.

In order to keep good animal health, the balance of mineral nutrient elements in forage or animal diet is very important. A lack of one mineral element content can not be balanced by the others. These elements could be in certain ratio. In the present study, the Ca/P, Ca/Mg, Ca/K, K/Mg, K/P of white clover was 3.9, 4.0, 0.6, 6.8 and 6.2, respectively. Rodriguez Julià (1991) reported 6.25 K/P, 2.64 Ca/P and 0.45 Ca/K ratios from white clover-grass mixtures, almost similar to the present findings. The Ca and P are closely related to animal health and metabolism (Tekeli & Ates, 2005). It is very important to keep a proper balance of Ca and P in relation to vitamin D. A desirable ratio of Ca/P is between 2:1 and 1:1 (Miller & Reetz, 1995). Allison (2003) suggested that when concentrations of K and nitrogen (N) are high, 0.25 %, Mg in the forage may be required to prevent grass tetany.

The widely acknowledged benefit from introducing white clover into the pasture sward is enhanced soil fertility due to fixation of atmospheric nitrogen (Lane *et al.*, 2000). A symbiotic relationship with the bacterium *Rhizobium trifolii* allows white clover to utilize atmospheric nitrogen where soil nitrogen is insufficient. The present study demonstrated the positive influence of white clover on the nutrient status of soil. Presence of white clover in soil increased the organic C and N because of the addition of plant N and C throughout the growing season as root exudates, dead leaves and fragments of roots. The debris will decompose and be added to the humus or biomass organic matter pools and after mineralization can provide a low but continuous supply of nutrients to plants in the long run. In addition to the accumulation of OM through plant material, white clover has the potential to add N in soil *via* atmospheric N_2 fixation. Among different mineral nutrients, accumulation of N in white clover growing plots was the maximum i.e., 62% over control showing that part of this N may be added *via* N_2

fixation. The C:N ratio of soils was below 10 and the increase in the C:N ratio of soil with white clover over control was relatively small (10%). The low C:N ratio of the soil indicating that added C or N by white clover could easily be mineralized to NH_4^+–N and then into NO_3^-–N, forms of N available to plants. It is generally recognized that clover decreased the C:N ratio of soil OM (Elgersma & Hassink, 1997). In addition to C and N, soil P and K were also increased substantially following the introduction of white clover. The percent increase in P and K over control soil was 50 and 28%, respectively. This increase in P and K is attributed to the addition of OM in soil *via* plant materials. Organic matter is generally recognized as the primary P and K supplying source in the soil and loss and addition of P and K has been closely associated with the OM in the soil (Abbasi & Rasool, 2005; Liu *et al*., 2002). Furthermore, beside supplying N, forage legumes and grasses are reported to mobilize large quantities of other nutrients (Atta-Krah, 1990), which led to increase their uptake in plants

Presence of white clover in soil decreased the pH and decline in pH is attributed to the addition of OM *via* white clover. Soils under vegetation typically become acidic, the effect usually being ascribed to the release of acidic organic compounds soil during decomposition. Under temperate-climate conditions, accelerated soil acidification has been observed where improved pastures of annual grasses in mixture with legumes (e.g., *Trifolium subterraneum*) have been grown continuously for 25 to 50 years (Haynes, 1983; Bolan *et al*., 1991). In contrast to temperate leguminous plants, the growth of grasses absorbing nitrate (by exchange of OH^- or HCO_3^-) generally tends to raise soil pH (Boonman, 1993).

Conclusions

This study was carried out to determine the mineral nutrient content of indigenous and exotic ecotypes of white clover grown in the soil with low inherent fertility. White clover provided forage that is very high in nutritive value and would therefore improve the pasture value if introduce in the pastoral ecosystem. The positive effect of white clover on quality of companion grasses would be of great significance if white clove/grass mixture is established in our rangelands. The high forage value of white clover would increase the meat and milk production potential of dairy animals. Residual effect of white clover was also studied during the investigation. Beside supplying N, white clover also mobilized other nutrients which led to increase their concentration in soil. White clover would enhance the nutrient status of soil if introduced in the agriculture ecosystem. Indigenous ecotypes found more nutrient efficient than exotic ecotypes having higher mineral nutrient contents.

Acknowledgements

This work was funded by Pakistan Science Foundation, Islamabad through Project/grant No. PSF/Res/AJK-UCR/Agr (275). Thanks to Dr. L. A. Lane, of New South Wales Agricultural Research and Advisory Station, PMB, Glen Innes, NSW, Australia for providing seeds of indigenous ecotypes of white clover and improvements in the manuscript.

References

Abbasi, M.K. and G. Rasool. 2005. Effects of different land-use types on soil quality in the hilly area of Rawalakot Azad Jammu ad Kashmir. *Acta Agric. Scand. Sec. B-Soil Plant Sci.,* 55: 221-228.

Abbasi, M.K. and M.N. Khan. 2004. Introduction of white clover for herbage production and nitrogen fixation in the hilly areas of Azad Jammu and Kashmir. *Mount. Res. Develop.,* 24: 234-240.

Allison, C. 2003. *Controlling Grass Tetany in Livestock. Guide B-809, 2*. New Mexico State University Press, Mexico.

Atta-Krah, A.N. 1990. Alley farming with *Leucaena*: Effect of short grazed fallows on soil fertility and crop yields. *Exp. Agric*., 26: 1-10.

Ayres, J.F., K.S. Nandra and A.D. Turner. 1998. A study of the nutritive value of white clover (*Trifolium repens* L.) in relation to different stages of phonological maturity in the primary growth phase in spring. *Grass Forage Sci.,* 53: 250-258.

Bolan, N.S., M.J. Hedley and R.E. White. 1991. Processes of soil acidification during nitrogen cycling with emphasis on legume based pastures. *Dev. Plant Soil Sci.,* 45: 169-179.

Boonman, J.G. 1993. From shifting cultivation to crop-grass rotations. In*: East Africa's Grasses and Fodders: Ecology and Husbandry*. (Ed.): J.G. Boonman. Kluwer Academic Publishers, Dordrecht, pp. 65-85.

Bremner, J.M. and C.S. Mulvaney. 1982. Nitrogen–Total. In: *Methods of Soil Analysis, Part 2. Chemical and Microbiological Properties Part 2*. (Eds.): A.L. Page, R.H. Miller & D.R. Keeney. ASA-SSSA, Madison, WI, pp. 595-624.

Dunlop, J. and A.L. Hart. 1987. Mineral nutrition. In: *White clover*. (Eds.): M.J. Baker, W.M. Williams. CAB International, Wallingford, Oxon, UK, pp. 153-181.

Elgersma, A. and J. Hassink. 1997. Effects of white clover (*Trifolium repens* L.) on plant and soil nitrogen and soil organic matter in mixture with perennial ryegrass (*Lolium perenne* L.). *Plant Soil,* 197: 177-186.

Gibson, P.B. and W.A. Cope.1985. *White clover*. In: *Clover Science and Technology*. (Ed.): N.L. Taylor. Soil Science Society of America, Madison, WI, USA, pp. 471-490.

Gupta, R.K. 1977. Energy forests- Farm and Community lands. Indian Farming.

Haynes, R.J. 1983. Soil acidification induced by leguminous crops. *Grass Forage Sci.,* 38: 1-11.

Hoveland, C.S. 2000. Achievements in management and utilization of southern grasslands. *J. Range Manage*., 53: 17-22.

Jarvis, S.C. 1980. The uptake and distribution of copper in perennial ryegrass and white clover grown in flowing solution culture with a controlled supply of copper. *J. Sci. Food Agric.,* 31: 870-876.

Keeney, D.R. and D.W. Nelson. 1982. Nitrogen-Inorganic forms. In: *Methods of Soil Analysis, Part 2. Chemical and Microbiological Properties Part 2*. (Eds.): A.L. Page, R.H. Miller & D.R. Keeney.ASA-SSSA, Madison, WI, USA, pp. 643-693.

Lal, R., G.F. Wilson and F. Okigbo. 1979. Changes in properties of an Alfisol produced by various crop covers. *Soil Sci. Soc. Am. J.,* 127: 377-382.

Lane, L.A., J.F. Ayres and J.V. Lovett. 2000. The pastoral significance, adaptive characteristics and grazing value of white clover (*Trifolium repens* L.) in dryland environments in Australia: a review. *Aus. J. Exp. Agric*., 40: 1033-1046.

Ledgard, S.F. and K.W. Steele.1992. Biological nitrogen fixation in mixed legume/grass pastures. *Plant Soil,* 141: 137-153.

Liu, S.L., B.J. Fu, Y.H. Lü and L.D. Chen. 2002. Effects of reforestation and deforestation on soil properties in humid mountainous areas: a case study in Wolong Nature Reserve, Sichuan province, China. *Soil Use Managt.,* 18: 376-380.

Melkania, N.P. and J.S. Singh. 1989. Ecology of Indian grasslands. In: *Perspective in Ecology.* (Eds.): J. S. Singh and B. Gopal. Jagmonder Book Agency, New Delhi.

Miller, D.A. and H.F. Reetz-Jr. 1995. Forage fertilization. In*: Forages Vol. I: An Introduction to Grassland Agriculture.* (Eds.): R.F. Barnes, D.A. Miller and C.J. Nelson.Iowa State Univ. Press, Ames, Iowa, pp. 79-91.

Misri, B.K. 1997. Important fodder trees/shrubs of temperate Himalaya: Distribution pattern and habitat affinities. In *Training Course on Management and Utilization of fodder trees and shrubs in sub-tropical and temperate Himalaya*, Sponsored by FAO Rome, organized by IGFRI, Jhansi, (September 22-30, 1997), pp. 18-23.

Nelson, D.W. and L.E. Sommers. 1982. Total carbon, organic carbon and organic matter. In: *Methods of Soil Analysis, Chemical and Microbiological Properties, Part 2* (Eds.): A.L. Page, R.H. Miller and D.R. Keeney. ASA-SSSA, Madison, WI, USA, pp. 539-579.

Olsen, S.R. and L.E. Sommers. 1982. Phosphorus. In: *Methods of Soil Analysis, Chemical and Microbiological Properties, Part 2.* (Eds.): A.L. Page, R.H. Miller & D.R. Keeney. ASA-SSSA, Madison, WI, USA, pp. 403-430.

Ram, J. and S.P. Singh. 1994. Ecology and conservation of alpine meadows in central Himalaya, India. In: *High altitudes of the Himalaya*. (Eds): Y.P.S. Rangtey and R.S Rawal. Nainital Gyanodaya Prakashan, India, pp. 221-276.

Rodriguez Julià, M. 1991. Desarollo y evaluación del sistema integrado de diagnostico y recomendación (DRIS) para la fertilización de las praderas permanentes. University of Basque State, Spain, pp. 3-25.

Rogers, G.L., R.H.O. Porter and I. Robinson. 1982. Comparison of perennial ryegrass and white clover for milk production. Proceedings of a conference on dairy production research organized by the new Zealand and Australian Societies of Animal production, Ruakura Animal Research Station, Hamilton, New Zealand, 2-5 February, 1982.

Roy, D., K.A. Shankaranarayan and R.S. Pathak. 1989. The fodder trees of India and their importance. *Indian Forester,* 106: 306-311.

Tekeli A.S., R. Avcýoðlu and E.Ates. 2003. Changes in some morphological and chemical properties of Persian clover (*T. resupinatum* L.) in relation to time and above-ground biomass. *Ankara Uni. J. Agric. Sci*., 9: 352-357.

Tekely, A.S. and E. Ates. 2005. Yield potential and mineral composition of white clover (*Trifolium repens* l.)-tall fescue (*Festuca arundinacea* schreb.) mixtures. *J. Cent. Eur. Agric.,* 6: 27-34.

Thomson, D.J. 1984. The Nutritive value of white clover. In: *Forage Legumes Occasional Symposium No. 16.* (Eds): D.J. Thomson. British Grassland Society, pp. 78-92.

Thomson, D.J., D.E. Beever, M.J. Haines, S.B. Cammell, R.T. Evans, M.S. Dhanoa and A.R. Austin. 1985. Yield and composition of milk from Friesian cows, grazing either perennial ryegrass or white clover in lactation. *J. Dairy Res*., 52: 17-21.

Ulyatt, M.J., J.A. Lancaster and W.T. Jones. 1977. The nutritive value of legumes. *Proc. New Zeal Grassland Assoc*., 38: 107-118.

Wilkins, R. and H. Munro. 1988. The prospects for livestock production from white clover. In: *Science and Change in Agriculture*. (Ed.): J. Hardcastle. Agriculture and Food Research Council: London, pp. 12-13.

(Received for publication 20 August 2008)