## INOCULATION AND INTER-CROPPING OF LEGUMES IN ESTABLISHED GRASS FOR INCREASING BIOMASS OF FODDER

### Muhammad Arshad Ullah\*, Nazir Hussain\*\*, Helge Schmeisky\*\*\* and Muhammad Rasheed\*\*\*\*

**ABSTRACT:-** Livestock sector has become very important component of agriculture sector in the world due to variety of dairy and meat products and high income to the farmers. In Pakistan, this vast resource faces many crucial challenges like low quality and high priced feed and fodder and limited chances of increasing area under fodders due to competition for food crops. Intercropping (33%, 50% and 67%) of Panicum maximum grass and legumes (Vicia sativa and cowpeas) coupled with inoculation was studied under rainfed conditions at National Agricultural Research Centre (NARC) Islamabad, Pakistan. Intercropping significantly increased tillering of grass. Seed inoculation of legumes also gave maximum tillers. The grass and legumes biomass without any treatment were recorded as 7.09 and 8.17 t ha<sup>-1</sup>, respectively, during two years of study. Mixed fodder production increased to 11.62, 13.6 and 14.13 t ha<sup>-1</sup> with 33%, 50% and 67% intercropping, respectively. Respective values of biomass were observed as 13.18, 13.70 and 17.87 t  $ha^{-1}$  when combined with inoculation. Intercropping of grass and legumes 67% with inoculation was assessed as the best treatment. The increases were computed as 304%, 230%, 132%, and 60% over grass alone in the first, second, third and fourth crops while respective increases were 101%, 151%, 165% and 74% over monoculture legumes.

Key Words: Panicum maximum; Tillage; Tillering; Legume Crops; Inoculation; Dry Biomass; Pakistan.

#### INTRODUCTION

Livestock sector is developing in Pakistan and its growth rate is the highest among different components of agriculture. Feed and fodder are not only deficient but also very highpriced as well as low in required ingredients. Therefore, the number of animals is far less in comparison of accelerating human population. One of the major problems hindering expansion of ruminant production in the country is the un-availability of good quality fodder in sufficient quantity (Sarwar et al., 2002). The scope of increasing area under fodder crops is not possible because production of cereals and cash crops severely compete with it. The only solution under present situation is to produce more fodder from the

<sup>\*</sup> Land Resources Research Institute, National Agricultural Research Centre, Park Road, Islamabad, Pakistan.

<sup>\*\*</sup> Water Research and Development, Qatar Shell Research and Technology, Doha, Qatar.

<sup>\*\*\*</sup> Department of Landscape Ecology and Nature Conservation, Faculty of Organic Agriculture, University of Kassel, Witzen hausen, Germany. \*\*\*\* Department of Agronomy, PMAS Arid Agriculture University, Rawalpindi, Pakistan

Corresponding author: arshad\_pak786@yahoo.com

presently occupied area. Hence, increase in biomass of fodder per unit area and intercropping of fodder crops are the only alternatives.

Good quality fodder is of great importance for economical animal production. Both quality and quantity of fodder are influenced due to plant species (Kaiser and Piltz, 2002), stage of growth (Kim et al., 2001) and agronomic practices (Rehman and Khan, 2003). Vetches (*Vicia* species) are legumes, which are well adapted to growth in winter and Mediterranean environments throughout the world on a various soil types.

Cowpeas (the legume species *Vigna unguiculata*) is highly palatable, nutritious and rich in protein, calcium and phosphorus than many other summer legumes. It contains higher protein contents, amino acids and vitamins (Bose and Balakarishnan, 2001).

The increasing leguminous portion in animal diet not only increase protein content but also enhance voluntary intake and digestibility of entire diet (Parveen et al., 2001).

Biological  $N_2$  fixation, especially the symbiotic association between legumes and Rhizobia, can provide sufficient amount of N to plants, which reduces the need for industrial fertilizers and the use of nitrogen fertilizers is thus not recommended (Oliveria et al., 2004). The use of inoculation is very low just below 1-3% of the total area under legumes, which is negligible (Aslam et al., 2000). Seed inoculation helps to improve nodulation, N fixation, crop growth and yield of leguminous crops (Zamarud et al., 2006). Biofertilizers (inoculation material) are apparently environment friendly, low cost, nonbulky agricultural inputs, which could play a significant role in plant nutrition as a supplementary and complementary factor to mineral nutrition (Sahai, 2004). Thus, keeping in view the limitations and constraints faced by the farmers busy in livestock production, a comprehensive study was conducted to evaluate the yield performance of grass-legume mixtures under different growing seasons and assess the impact of inoculation of legumes for maximum fodder production.

# MATERIALS AND METHODS

The study was conducted under rainfed conditions for two years and completed in 2007 at experimental area of Rangeland Research Program, National Agricultural Research Centre (NARC) Islamabad, Pakistan. The experimental treatments were;

- $T_1 = Grass 100\%$
- T<sub>2</sub> = Legumes of the season (Cowpeas in summer and Vetch in winter)
- $T_3 = Grass + 33\%$  legumes
- $T_4 = Grass + 50\%$  legumes
- $T_5 = Grass + 67\%$  legumes
- $T_6 = T_2 + inoculation$
- $T_7 = T_3 + inoculation$
- $T_8' = T_4' + inoculation$
- $T_9 = T_5 + inoculation$

The experimental soil has pH 8.4, ECe 0.53 dS m<sup>-1</sup>, total N 0.037%, available P 4.7 mg kg<sup>-1</sup>, extractable K 79.6 mg kg<sup>-1</sup>, O.M 0.53% and textural class was sandy clay loam.

Panicum maximum grass was planted as perennial fodder. Winter legume (Vicia sativa commonly known as vetch) and summer legume (Vigna unguiculata commonly known as cowpeas) were sown as inter crops in the established grass. Summer legume followed winter legume in the next year. Seed of legumes was inoculated with *Rhizobium leguminosarum* before sowing to obtain  $T_6$  to  $T_9$ . The experiment was laid out using randomized complete block designs (RCBD) with four replications.

This study was conducted under rainfed conditions and no irrigations and fertilizer were applied. Grass was harvested at panicle stage, whereas legumes were harvested at 100% flowering. Number of tillers of grass was taken at the panicle stage selecting five plants from each treatment. All the plants in one square meter were clipped close to ground level. Three quadrates were harvested randomly for fresh biomass. Fresh biomass was recorded in each plot of grass and legumes. Then data were calculated on t ha<sup>-1</sup> basis. The fresh samples were oven dried to a constant temperature at 70 °C for 72 h. The dry samples were weighed and dry matter yield (t ha<sup>-1</sup>) was calculated. The meteorological data of three year (2005-07) were collected and divided it by 36 months. This data is the overall average of all the months in three years. Monthly average rainfall was 86 mm, 104 mm and 118 mm, wind speed 60 km day<sup>-1</sup>, 51 km day<sup>-1</sup> and 46 km day<sup>-1</sup>, pan evaporation 4 mm day<sup>-1</sup>,4 mm day<sup>-1</sup> and 4 mm day<sup>-1</sup>, sunshine 8 h day<sup>-1</sup>, each monthly maximum and minimum average temperature  $28^{\circ}$ C,  $3^{\circ}$ C,  $28^{\circ}$ C, 14°C, 28°C, 13°C and average maximum and minimum relative humidity were 83%, 49%, 81%, 51%, 83% and 50% during the study period i.e., 2005, 2006 and 2007, respectively (WRRI, 2007).

All the data were analyzed using one-way analysis of variance with the help of software package of MSTAT-C Microcomputer program, Version 1.3. A least significant difference (LSD) was applied for multiple comparisons (Bicker, 1991).

## **RESULTS AND DISCUSSION**

The growth of grass (Panicum *maximum*) as well as legumes was positively affected when Vicia sativa (vetch) and Vigna unguiculata (cowpeas) forage legumes were inoculated (Table 1). A significant increase was recorded in plant height of *Vicia sativa* in all the three planting geometries (33%, 50% and 67% intercropping). Similar trend was also observed in cowpeas. The intercropping of grass and legumes improved the plant height of grass as well as Vicia sativa forage legume, whereas the height of cowpeas was suppressed in intercropping treatment but positive effect on grass growth was still persistent. Most of these results were also verified in the second year (2007) of the study.

The intercropping treatment results revealed a positive behavior and no negative competition existed for water, nutrients and light up to 67% intercropping. Hence, this planting geometry did not alter the situation significantly, rather proved beneficial. Trannin et al. (2000) reported assimilation of significant amount of nitrogen derived from legume root after harvesting of the crops. They claimed that nitrogen transfer was via decomposing organic matter of legume rather than root exudates or direct mycorhizal hyphae. The positive results have been found in different crops with

intercropping					-	0	0	<b>0</b> (ama)
Treatment	2005-06				(cm) 2006-07			
	Grass	Vicia sativa	Grass	Cowpeas	Grass	Vicia sativa	Grass	Cowpeas
T <sub>1</sub>	114 <sup>b</sup>	-	166 <sup>b</sup>	-	103.0 <sup>d</sup>	-	176.8 <sup>°</sup>	-
$T_2$	-	44 <sup>°</sup>	-	$184^{ab}$	-	$55.0^{\circ}$	-	$115.7^{\circ}$
<b>T</b> <sub>3</sub>	$117^{\text{b}}$	44 <sup>°</sup>	163 <sup>b</sup>	182 <sup>b</sup>	$131.0^{\circ}$	60.3 <sup>b</sup>	$188.0^{\rm b}$	$109.0^{d}$
<b>T</b> <sub>4</sub>	120 <sup>b</sup>	46 <sup>b</sup>	184 <sup>ª</sup>	163 <sup>b</sup>	$143.7^{b}$	$56.0^{\circ}$	199.0 <sup>ab</sup>	130.0 <sup>°</sup>
T <sub>5</sub>	118 <sup>b</sup>	48 <sup>b</sup>	$189^{a}$	$158^{\circ}$	146.0 <sup>b</sup>	61.3 <sup>b</sup>	$208.0^{\mathrm{a}}$	139.3 <sup>b</sup>
T <sub>6</sub>	-	53 <sup>ª</sup>	-	193 <sup>ª</sup>	-	$56.7^{\circ}$	-	132.0 <sup>°</sup>
<b>T</b> <sub>7</sub>	120 <sup>b</sup>	48 <sup>b</sup>	182 <sup>ª</sup>	$180^{ab}$	163.7 <sup>ª</sup>	63.0 <sup>ª</sup>	208.3 <sup>ª</sup>	$107.0^{d}$
T <sub>8</sub>	129 <sup>a</sup>	54 <sup>ª</sup>	191 <sup>ª</sup>	$187^{a}$	146.3 <sup>b</sup>	60.3 <sup>b</sup>	$202.7^{\mathrm{a}}$	149.3 <sup>b</sup>
T <sub>9</sub>	134 <sup>ª</sup>	55ª	196 <sup>ª</sup>	197 <sup>a</sup>	$151.0^{\mathrm{b}}$	65.3ª	214.0 <sup>ª</sup>	$174.3^{a}$
LSD	9.593	2.969	17.78	12.64	11.45	3.05	16.41	15.12

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Assessment of inoculation effect on plant height in grass-legumes

Mean followed by same letter do not differ significantly at 5% level of probability

Table 1.

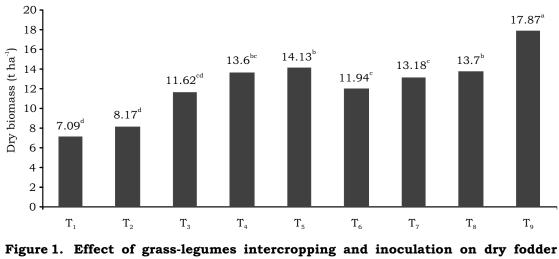
inoculation of legume seeds (Jatish et al., 2000; Stephan et al., 2002; Ampe et al., 2003; Karas et al., 2005). Moreover, Abbas et al., (2001) also reported better growth of barley, pearl millet as well as rhodes, rye and sudan grasses when these were intercropped with legumes. Zhang and Li (2003) recorded the positive effect on growth when wheat/maize was intercropped with soybean. They reported a compensatory growth of non-legume species as well when intercropped with soybean and attributed this effect as the competition-recovery production principle. The improved nutrition of non-legumes especially nitrogen and phosphorus were also found by them when faba bean legume was intercropped. Due to effective  $N_{2}$ fixation, perennial forage legumes have a great potential to increase sustainability in grassland farming based on livestock production

(Carlsson and Huss-Danell, 2003).

Tillering of grass (Panicum *maximum*) as well as forage legumes Vicia sativa and Vigna unguiculata (cowpeas) had favourable effect when legumes were sown after seed inoculation (Table 1). A significant increase was noted in tillering of Vicia sativa in all three planting patterns (33%, 50% and 67%). In cowpeas, tillering responded a normal behaviour. The tillering of grass was increased after every harvest due to its perennial nature. The tillering behaviour of cowpeas was suppressed due to intercropping as well as inoculating treatment because of creeping behaviour. Most of the results of grass and Vicia sativa were also confirmed in second year of the study. Non-significant result was indicated in tillering behaviour of cowpeas during second year as well. However, a small increase was observed in intercropped treatments.

Similar impact was recorded in cowpeas inoculation. The influence of inoculation on tillering of grass and legumes very clearly indicated that well establishment rhizobial population not only favoured the legumes but also the companion grass. The legume Vicia sativa has an erect growing behaviour therefore, did not negatively affect the tillering of grass even when intercropped up to 67%. However, cowpeas have a little erect growing behaviour and subsequent creeping of the crops decreased its tillers due to space competition. Turk (2000) also recorded less tillering of monoculture barley as well as vetch compared with intercropping. as However, the intercropping ratio in that study was 1:3 due to which there was more plant competition for space and light.

Plant height, tillering, branching are ultimately translated into biomass produced in a field. The common finding observed was the increase in weight of fodder biomass of the grass as well as legumes when the later were inoculated (Figure 1). Positive effect of inoculation was consistent in all the four crops over two years. However, biomass increase was more in the later three crops as compared to the first crop that might be due to more rhizobial population established by inoculation with passage of time. The effect of intercropping of legumes up to 67% was not found negatively affecting the biomass production, although plant population increased. Rather a significant increase in biomass production was recorded than grass alone when legumes were intercropped by 67% (Figure 2). Intercropping geometry of 50% was statistically significant in all the four crops except third crop; while, intercropping of 33% remained significant over monoculture grass during the first year. However, inoculation of legumes and intercropping geometry when coupled proved more beneficial and caused significant increase in biomass over monocultures of grass or legumes (Figure 3). The combination of 67% intercropping with inoculation proved the most successful and produced the maximum biomass that was significantly



weight (Means followed by same letter do not differ significantly at P<5%)

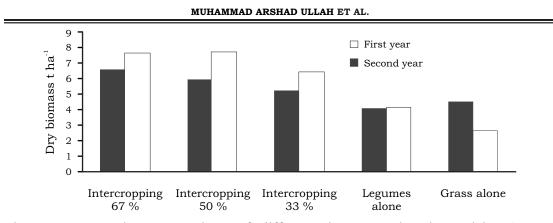


Figure 2. Year wise comparison of different intercropping intensities (grass+ legumes) for fodder production

higher than all the other treatments of the experiment. The increases in biomass production due to this treatment was computed as 304%, 230%, 132%, and 60% over grass alone in the first, second, third and fourth crops, respectively indicating a decrease in increase quantum with time due to establishment and tillering of grass. The increases were calculated as 101%, 151%, 165% and 74% over monoculture legumes in first, second, third and fourth crops, respectively.

Inoculation of both the legumes *Vicia sativa* and *Vigna unguiculata* proved also useful and its coupling with intercropping further enhanced the total fodder yield of four crops in two years (Figure 1). This may be due to symbiotic nitrogen fixation by establishment of rhizobial population, more elemental nitrogen fixation that was also transmitted to the companion grass. Trannin et al. (2000) reported significant assim-

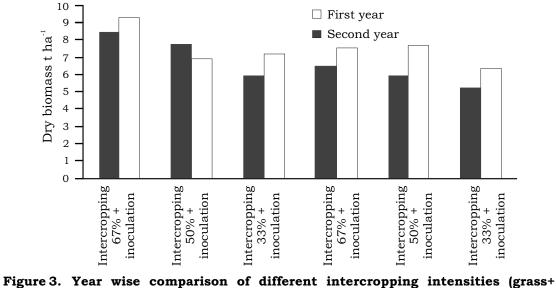


Figure 3. Year wise comparison of different intercropping intensities (grasslegumes) for fodder production

intercropping								
Treatments	2005-06				2006-07			
	Grass	Vicia sativa	Grass	Cowpeas	Grass	Vicia sativa	Grass	Cowpeas
<b>T</b> <sub>1</sub>	$7^{d}$	-	$25^{d}$	-	25.33°	-	24.33ª	-
$T_2$	-	$7^{d}$	-	3.00 <sup>ª</sup>	-	10.00 <sup>e</sup>	-	6.33 N
T <sub>3</sub>	$7^{d}$	8°	$30^{\circ}$	$2.67^{\text{b}}$	$30.13^{\text{bc}}$	$11.00^{\text{de}}$	30.33 <sup>cd</sup>	7.33
<b>T</b> <sub>4</sub>	8°	9 <sup>b</sup>	32 <sup>b</sup>	$2.67^{\text{b}}$	38.33ª	$12.00^{d}$	$35.00^{\text{bc}}$	7.00
<b>T</b> <sub>5</sub>	8 <sup>c.</sup>	$10^{ab}$	34 <sup>ab</sup>	3.00ª	43.33ª	$13.67^{\circ}$	$36.67^{\text{b}}$	7.00
<b>T</b> <sub>6</sub>	-	$10^{ab}$	-	$2.00^{\circ}$	-	$12.67^{\circ}$	-	6.67
<b>T</b> <sub>7</sub>	$9^{\text{b}}$	$10^{ab}$	$28^{\circ}$	$2.00^{\circ}$	$37.67^{\text{b}}$	13.33°	$28.33^{de}$	7.00
T <sub>8</sub>	$10^{ab}$	$11^{a}$	$37^{a}$	$2.00^{\circ}$	39.33ª	15.00 <sup>b</sup>	42.33ª	7.67
<b>T</b> <sub>9</sub>	$11^{a}$	$11^{a}$	38 <sup>ª</sup>	$2.67^{\scriptscriptstyle \mathrm{b}}$	42.00 <sup>ª</sup>	18.00 <sup>ª</sup>	44.67 <sup>ª</sup>	7.33
LSD	0.162	0.1424	2.044	0.1106	6.902	0.4365	4.963	-

INOCULATION AND INTER-CROPPING OF LEGUMES

Assessment of inoculation effect on tillering (per plant) in grass-legume

Means followed by the same letter do not differ significantly at 5% level of probability; NS=Non significant

ilation of nitrogen by coupling crop that was derived from legume roots and decomposition of roots after harvest. It has already been observed that plant height and tillering increased due to different treatments of this study (Tables 1 & 2). Turk (2000), Sleugh et al. (2000), Mohapatra et al. (2001), Abbas et al. (2001), Gubkina (2001), Kuzeev (2002), Zimkova et al. (2002), Daba and Haile (2002), Bergkvist (2003 a & b), Lauk and Lauk (2003), Glacomini et al. (2003), Kuchinda et al. (2003), Kurdali et al. (2003),), Kirilov et al. (2003) and Karadag (2004) also obtained similar results. Geherman and Parol (2004), Hoffman et al. (2004) Malikov (2004), Tamm and Tamm (2005), Sudesh et al. (2006) and Li-Long et al. (2007) also recorded higher dry matter yield when different leguminous crops were intercropped with non-leguminous crops or grasses. They found an

Table 2.

increased growth of a second crop species grown in alternate rows that led to large yield increments on phosphorus-deficient soils.

Biomass production of grass (*Panicum maximum*) can be increased significantly by inter-cropping of legumes (*Vicia sativa* or cowpeas) at 33%, 50% and 67%. The 67% proved the most effective. Seed inoculation also enhanced forage production. These technologies were successful to meet plant nutritional requirements because a good biomass production was obtained in soil highly deficient for supply of nutrients.

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# AUTHORSHIP AND CONTRIBUTION DECLARATION

S. No	Author Name	Contribution to the paper
1.	Dr. Muhammad Arshad Ullah	Conceived the idea, Wrote abstract, Methodology, Did SPSS analysis, Data collection, Results and discussion,
2.	Dr. Nazir Hussain	Introduction Conceived the idea, Data entry in SPSS and analysis
3.	Dr. Helge Schmeisky	Table Figure and Statistical Interpretation
4.	Dr. Muhammad Rasheed	Result and Discussion, References

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