

## NITROGEN FIXATION AND TRANSFER IN TROPICAL LEGUME-GRASS SWARDS IN SOUTH-EASTERN QUEENSLAND

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### ABSTRACT

The amount of fixed N in tops of tropical legume-grass swards and transfer of fixed N to the grass have been estimated for swards at three sites in south-eastern Queensland over a five year period. Swards were sampled for dry matter and nitrogen content usually twice each growing season and all plant material above 10 cm was removed from plots after each sampling. The amount of fixed N in tops was in the range 100–140 kg N ha<sup>-1</sup> for *Macroptilium atropurpureum* cv. Siratro, *Glycine wightii* cv. Cooper and *Desmodium intortum* cv. Greenleaf while values of 51 and 74 kg N ha<sup>-1</sup> were estimated for *Lotononis bainesii* cv. Miles. Estimated transfer of fixed N below cutting height to the tops of the companion grass, relative to total fixed N in tops, was 12–15% for Siratro and lotononis, 16% for glycine and 17% for desmodium.

### INTRODUCTION

One of the main criteria in assessing pasture legumes is their capacity to increase the N available to grazing animals. Estimates of N fixed by apparently well grown tropical pasture legumes range from < 50 to 900 kg N ha<sup>-1</sup> year<sup>-1</sup> (Nutman 1976). Most of these values were derived from N yields of tops of pure legume swards. In mixed legume-grass swards, fixed N in tops can be estimated from the total N yield of legume and grass tops in the sward minus the uptake of mineral N from sources other than fixation by the legume. This mineral N can be estimated from the N yield of the grass growing alone in adjacent plots (Nutman 1976). Long-term N yields of tropical legume-grass mixtures, together with those of the grasses growing alone, have been measured at different sites in Queensland (Jones, Davies and Waite 1967; Miller and List 1977). However, the actual yields of fixed N in tops could not be estimated as a major portion of the unsampled material would have been returned by the grazing and topping technique used; after each sampling plots were grazed for up to one week and then topped with a mower. This would have resulted in a recycling of fixed N already accounted for in the yield harvests.

An important consequence of legume growth in mixed swards is transfer of fixed N to the companion grass. This can be estimated from the difference between N yield of the grass grown with the legume and that when grown alone. When top residues are returned to the soil, as in the above experiments, N transfer can be considerable: viz. 29–39% of legume N yield for *Macroptilium atropurpureum* cv. Siratro, 20% for *Desmodium intortum* cv. Greenleaf and *Glycine wightii* cv. Tinaroo and 13% for *Lotononis bainesii* (Jones, Davies and Waite 1967, Miller and List 1977). However, studies in pots and micro-plots suggest that the contribution of fixed N below cutting height to N transfer is proportionately small: viz. < 2% of the N fixed by *D. uncinatum* and *Indigofera spicata* over two years (Henzell 1962) and up to 5% by *D. intortum* and 6–11% by *Centrosema pubescens* over one year (Whitney, Kanehiro and Sherman 1967). The importance of long-term, direct N transfer (below cutting height) in tropical legume-grass swards in the field cannot be assessed from the above studies.

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A cut and removal trial, measuring the response of various tropical legume-grass swards to Mo at six sites over five years in south-eastern Queensland (Johansen *et al.* 1977, 1978) provided an opportunity to estimate long-term yield of fixed N in tops and N transfer to the grass.

## METHODS

Characteristics of the sites and experimental details have been given by Johansen *et al.* (1977). Soil type (Stace *et al.* 1968) and average annual rainfall for each site is as follows: A—xanthozem at Cooroy, 1627 mm; B—red podzolic at North Deep Creek, 1211 mm; C—prairie soil at Glastonbury, 1151 mm; D—yellow podzolic at Ormeau, 1499 mm; E—solodized solonetz at Ripley, 894 mm; F—yellow podzolic at Eskdale, 872 mm. Treatments consisted of several rates and sources of Mo applied to legumes grown with a companion grass, *Panicum maximum* cv. Gatton.

Measurements of fixed N in tops and transfer of fixed N to the grass were determined for all those legumes which grew well in the plus Mo treatment but only at sites where there was negligible growth of at least one legume species in the nil Mo treatment. This occurred at sites A, B and C and for the legumes, *Desmodium intortum* cv. Greenleaf, *Glycine wightii* cv. Tinaroo, *Lotononis bainseii* cv. Miles and *Macroptilium atropurpureum* cv. Siratro. At the nil Mo treatment, there was negligible growth of desmodium at site A after the first harvest of the second year, of Siratro at site B after the first harvest of the third year, and of glycine at site C from the first year (Johansen *et al.* 1978). The N yield of the grass in these nil Mo treatments was assumed to be from uptake of mineral N from sources other than fixation by the legume.

Dry matter yield was determined by cutting two 0.5 m<sup>2</sup> quadrats at 10 cm height from each 5 m × 5 m plot after periods of vigorous growth, usually twice each season. The remaining plant material above 10 cm was mown and removed from the experimental area.

At each harvest, samples of the legumes and Gatton panic were taken from each plot for N analysis. Legume samples comprised ten shoot tip samples, back to the third expanded leaf for Siratro, desmodium and glycine and the distal 10–15 cm for lotononis. For Gatton panic, approximately 20 youngest expanded leaves (ligule apparent) were bulked for each sample. During the fifth growing season (1976), whole shoot samples (above 10 cm) of legume and grass were also taken from each plot for N analysis. Samples were oven dried, ground and analysed for N after Kjeldahl digestion (Williams and Twine 1967).

At the final harvest of the fifth growing season, soil samples for N analysis were taken at sites A, B and C from treatments receiving either no Mo, or high Mo with annual maintenance applications. Fifteen cores each 2 cm diameter and to a depth of 10 cm, were bulked for each of the four replicate plots. The soil was air dried and analysed for total N (Bremner 1965), nitrate-N (Bremner and Keeney 1966) and boiling water extractable N (Method 2 of Keeney and Bremner 1966). Boiling water extractable N gives a measure of organic N that is easily mineralized to nitrate and was found to correlate well with plant N uptake (Keeney and Bremner 1966). Replicates were analysed separately for site C but were bulked for sites A and B.

## RESULTS AND DISCUSSION

### *Nitrogen concentrations in topgrowth*

Nitrogen concentrations in whole shoots for the first four years were calculated from the ratios of N concentration in whole shoot to that in the corresponding tip (legume) or first expanded leaf (grass) samples established from the fifth year's data (Table 1), as whole shoot samples for N analysis were not taken during the first four growing seasons. Harvests from all sites and for all legumes were included in this

analysis. These ratios were not significantly affected by Mo treatment, even though there was a strong N response, and they did not differ significantly, within harvest and site, for the four legumes studied. However, ratios for grasses were usually higher when grown with lotononis or desmodium than with Siratro or glycine (Tables 1 and 2). There were also some significant site and sampling time differences for both legumes and grasses (Table 2), probably due to differences in stage of growth at harvest. For example, at site E the physiologically younger material at the April 1976 harvest had higher ratios than the older material harvested in January 1976. Notwithstanding these differences the average ratios (Table 1) have been used for calculating N concentration in whole tops.

TABLE 1

*Nitrogen concentration in whole shoot samples as a percentage of that in tip samples (legumes) or youngest leaves (grass). Means and standard deviations over all sites and Mo treatments.*

	Legume species			
	Siratro	Lotononis	Desmodium	Glycine
Legume	(n=76) 74±11	(n=51) 74±14	(n=10) 75±7	(n=11) 75±6
Grass	43±7	50±8	48±8	44±8

TABLE 2

*Site, harvest date and species effects on the ratio of N concentration in whole shoot to tip (legume) or leaf (grass) samples (%)*

Site	Harvest date	Legume		Grass (with)	
		Siratro	Lotononis	Siratro	Lotononis
A	Jan. 1976	79.3	72.3	41.2	49.1
B	Feb. 1976	80.4	85.2	45.1	49.1
D	Jan. 1976	65.7	59.5	43.8	46.4
E	Jan. 1976	69.1	70.8	44.4	55.3
E	Apr. 1976	87.7	87.9	48.0	50.8

LSD (P=0.05) between:

species means	n.s.	2.3
site means	4.4	3.6

Nitrogen yields in tops of legumes and the companion grass were calculated as the product of N concentration in whole shoots and dry weight of tops (Johansen *et al.* 1978) for each harvest. Errors in this calculation would be due to variability of dry matter yield and N concentration. The data suggests yield was estimated within 10 to 15 per cent (Johansen *et al.* 1978). It appears that estimation of N concentration was of the same order of magnitude due to the average values used for the ratios of N in tip and whole shoot.

#### Nitrogen fixation

The amount of fixed N in tops in legume-grass swards averaged over five years ( $N_F$ ) was calculated as follows ( $\text{kg N ha}^{-1} \text{ year}^{-1}$ ):

$$N_F = N_{L+} + N_{G+} - N_{G-}$$

where  $N_{L+}$  is average annual yield of N in legume tops where legume growth was not limited by Mo deficiency,  $N_{G+}$  is average annual yield of N in tops of the companion grass and  $N_{G-}$  is average annual yield of N in grass tops in plots where Mo deficiency severely limited legume growth (i.e. the estimate of native soil mineral N uptake).

Average annual N yields of legumes and grasses in plots showing maximum legume growth at Mo responsive sites are shown in Table 3. High N yields were

obtained with Siratro at all sites, desmodium at site A and glycine at site C. The Siratro N yields were higher than the 55-75 kg N ha<sup>-1</sup> year<sup>-1</sup> obtained in mixed swards under a 1070 mm rainfall at Samford in south-eastern Queensland (Jones, Davies and Waite 1967) and similar to the 100 kg N ha<sup>-1</sup> year<sup>-1</sup> under irrigation in north Queensland (Miller and List 1977), where the Siratro was not well adapted. The N yields of the better adapted desmodium and glycine in north Queensland, namely 174 and 135 kg N ha<sup>-1</sup> year<sup>-1</sup>, respectively, were higher than obtained by these legumes in this study. Nitrogen yields of lotononis were similar to the 66 kg N ha<sup>-1</sup> year<sup>-1</sup> obtained at Samford (Jones, Davies and Waite 1967).

TABLE 3

*Nitrogen yields of legume and grass tops ( $N_{L+}$ ,  $N_{G+}$ ,  $N_{G-}$ ), fixed nitrogen in tops ( $N_F$ ) and nitrogen transfer ( $N_T$ ) in legume-grass swards averaged over five years (kg N ha<sup>-1</sup> year<sup>-1</sup>). See text for full definition of symbols.*

Site	Legume	$N_{L+}$	$N_{G+}$	$N_{G-}$	$N_F$	$N_T$	$\frac{N_T}{N_F} \times 100$
A	Siratro	85	53		97	12	12
	Desmodium	86	58	41	103	17	17
	Lotononis	63	52		74	11	15
B	Siratro	108	42	23	127	19	15
	Lotononis	45	29		51	6	12
C	Siratro	119	37		137	18	13
	Glycine	106	39	19	126	20	16

The N yields obtained in this study would be underestimates where all material was not harvested. This occurred after the final harvest in each season where there was some regrowth before winter and in specific instances of failure to harvest (Johansen *et al.* 1978). However, from growth observations, true dry matter yield would have been underestimated by less than 10 per cent.

The calculated values for fixed N in tops ( $N_F$ ) shown in Table 3 will depend on the degree to which  $N_{G-}$ , the N yield of the grass in nil Mo treatments of the most Mo responsive legumes, correctly reflects the uptake of mineral N not fixed by the legume.  $N_F$  would be underestimated if there was some fixation by legumes in nil Mo treatments. There was some early growth of desmodium at site A and Siratro at site B on the nil Mo plots. However this may have been a result of high availability of soil N due to mineralization promoted by cultivation rather than availability of Mo for N fixation, as there was virtually no growth after two years.  $N_F$  would be overestimated where  $N_{G-}$  does not account for the uptake of native mineral N in mixed swards. This could occur where better growth of grass and legume in Mo sufficient treatments allowed better access to native mineral N. Associative nitrogen fixation in the root systems of grasses has not been taken into account. Most likely it would be of low magnitude and occur in both the nil Mo and the Mo sufficient plots as the available N status of soils under rapidly growing tropical grasses is usually low (Henzell 1968).

#### *Transfer of Nitrogen*

Transfer of fixed N below cutting height to the grass ( $N_T$ ) was calculated as follows (kg N ha<sup>-1</sup> year<sup>-1</sup>):

$$N_T = N_{G+} - N_{G-}$$

Values for  $N_T$  will depend on  $N_{G-}$  as mentioned above, and also on the validity of  $N_{G+}$ . It would be an underestimation where the legume in the Mo-sufficient mixed swards takes up a significant proportion of the native soil mineral N. This underestimation is likely to be small as Vallis, Henzell and Evans (1977) found that, using

$^{15}\text{N}$  to label mineral N, the proportion of mineral N in Siratro, desmodium and lotononis grown with *Digitaria decumbens* or *Chloris gayana* in the field was nearly always less than 25% of annual uptake of mineral N. A factor resulting in over-estimation of  $\text{N}_{\text{G}+}$ , and hence  $\text{N}_{\text{T}}$ , would be return of some plant tops above cutting height when harvests were not taken or all material not removed. However, as mentioned previously, this is considered to be minimal.

In this study it is not possible to distinguish the relative contribution to  $\text{N}_{\text{T}}$  of N mineralization from leaf litter and decomposition of legume roots and nodules as estimates of leaf litter were not made. However, Whitney and Kanehiro (1967) estimated that at least half of the N available for transfer from *D. intortum* and *C. pubescens* to an associated grass derived from leaf fall rather than roots and nodules or leaching from attached leaves.

The amounts of N transferred to the grass relative to fixed N in tops in this study (Table 3—Siratro and lotononis 12-15%, glycine 16% and desmodium 17%) are considerably greater than those obtained in short term experiments with pots and microplots (Henzell 1962, Whitney, Kanehiro and Sherman 1967), whether percentages are calculated relative to either total fixed N or fixed N in tops alone. Indeed the present values approach the 20% obtained for a mixed sward with an annual legume, *Trifolium subterraneum* (Simpson 1976). In tropical legume-grass swards where much of the above-ground material was returned to the soil transfer of fixed N was 13-40% (Jones, Davies and Waite 1967, Miller and List 1977). The results of the present study suggest that a major proportion of this N transfer could come from material below cutting height.

#### *Increase in soil nitrogen*

In order to get some indication of the changes in soil N associated with legume growth over five years, measurements of total and "available" soil N were made in plots with optimum levels of Mo for legume growth and plots without Mo (Table 4). Optimum legume growth significantly increased all soil N parameters at site C. Although statistical significance was not tested, there appeared to be an increase in total N and boiling water extractable N for Siratro and desmodium at site A and an increase in total N and nitrate-N for Siratro at site B.

TABLE 4  
*Soil N in plots with optimum Mo and without Mo after five years.*

Site	Legume	Total N (%)		Nitrate -N (ppm)		Boiling Water N (ppm)	
		-Mo	+Mo	-Mo	+Mo	-Mo	+Mo
A	Siratro	0.186	0.234	1	2	120	150
	Desmodium	0.198	0.211	2	2	130	173
	Lotononis	0.180	0.136	1	2	177	164
B	Siratro	0.189	0.227	4	19	128	111
C	Siratro	0.225	0.270	4	25	138	166
	Glycine	0.219	0.297	2	34	160	222
LSD*		0.021		8		20	

\*Least significant differences (LSD,  $P=0.05$ ) refer to site C only and for comparison of N levels within species.

Calculation of the total amount of fixed N, above and below ground, would require intensive measurements of soil N and N in roots at the final harvest. As the present study was primarily concerned with estimating the amount of fixed N available for grazing animals the necessary detailed below-ground measurements were not

done. However, from the limited measurements available it appears that in some cases vigorous legume growth over five years contributed significantly to the total N content of the soil (Table 4). Measurement errors preclude detailed extrapolation but at site C growth of glycine appears to have caused an increase in the 0–10 cm horizon of about 700 kg N ha<sup>-1</sup> (bulk density = 0.9 g cc<sup>-1</sup>), slightly more than the five year total for fixed N in tops. The increases in nitrate in plots with vigorous legume growth are probably transient, depending on microbial activity, as suggested by the negligible effects at site A (Table 4).

Estimations of fixation and transfer will depend on the type and intensity of management imposed on a pasture. That is, the results will reflect the management system used. In this system of intermittent removal of above ground herbage it has been demonstrated that even though the major portion of fixed N is not returned there is considerable increase in soil N and transfer of fixed N to the grass from leaf fall and plant material below cutting height.

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