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Measurement of Nitrogen Fixed by *Pueraria Phaseloides* by N-15 Dilution Technique

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Key words: Nitrogen fixation; N-15 dilution technique, percent N-15 atom excess.

ABSTRAK

Satu kajian ladang telah dijalankan untuk menentukan jumlah N yang dapat diikat oleh **Peuraria phaseloides** yang ditanam sebagai tanaman penutup bumi di celah-celah barisan kelapa sawit (**Elaeis guineensis**) di Ladang Seafield, Batu Tiga, Selangor. Jenis rumput yang didapati tumbuh bersama ialah **Paspalum conjugatum** dan **Ottochloa nodosa**. Plot-plot berukuran 1 m \times 1 m dipilih secara rawak di antara barisan-barisan kelapa sawit untuk memberikan beberapa julat peratus tanaman kekacang dan peratus rumput. Plot-plot ini dipotong sehingga 5 cm dari paras bumi dan disiramkan dengan baja ammonium sulfat yang telah diperkayakan dengan 10% N-15 "atom excess" pada kadar 2 g N/m². Plot-plot ini dituai selepas 3 bulan. Analisis jumlah N dan perkayaan N-15 dalam tisu dijalankan ke atas rumput dan kekacang. Hasil penyelidikan menunjukkan perbandingan yang bermakna di antara jumlah N yang diikat oleh Peuraria apabila ia tumbuh bersaingan dengan beberapa julat peratus rumput. Apabila julat Peuraria berada di antara 0 – 40%, ia dapat mengikat N sejumlah 9.20 kg/ha/3 bulan. Ini meningkat ke 22.34 dan 37.80 kg/ha/3 bulan apabila julat Peuraria ialah 41 – 60 dan 61 – 80%. Jumlah N yang diikat oleh Peuraria ialah apabila komposisi sesuatu blot itu mengandungi lebih kurang 80% Peuraria.

ABSTRACT

A field experiment was carried out to determine the amount of nitrogen fixed by **Pueraria phaseloides** which was grown as a cover in the interrow of two-year old oil palms (**Elaeis guineensis**) in Seafield Estate, Batu Tiga, Selangor. The grasses that were found growing together with the legume cover were **Paspalum congugatum** and **Ottochloa nodosa**. Plots of 1 m by 1 m were randomly selected within the interrows of the palms to give various mixtures of grass and legume. These plots were cut at 5 cm above the ground, and applied with 2 g N/m² as ammonium sulphate enriched with 10% atom excess N-15. The regrowth of the legume and grass were harvested after three months, and analysed for total N and N-15 enrichment of the tissues. The results show that there were significant differences in total N fixed by Peuraria mixed with different percentages of grass. At 0 – 40% legume composition, the total N fixed by Peuraria was 9.20 kg/ha/3 months and increased to 22.34 and 37.80 kg/ha/3 months at 41 – 60% and 61 – 80% respectively. Total N fixed at 81 – 100% legume composition was 22.73 kg/ha/3 months. The maximum amount of N fixed by Peuraria was when the mixture contained about 80% legume standing.

INTRODUCTION

Nitrogen fertilizer is currently one of the most important nutrients in estate crop production. In oil palm plantations, the cost of the nitrogen fertilizer amounts to half of the total fertilizer budget. It has been observed that crops grown without leguminous covers established between the planting rows require much more chemical fertilizer additions for optimum growth and production compared to crops grown with leguminous ground covers (Pushparajah and Tan, 1976; Yeow *et al.*, 1982).

Various workers have measured the contribution of leguminous ground covers to the soil and crops grown on that area, but quantitative measurements using the isotope dilution technique has not been attempted. This is due to the high cost of labelled nitrogen fertilizer and the lack of instrumentation for the measurement of labelled N (N-15).

This study is the first to be conducted in this field in an attempt to measure the amount of nitrogen fixed by a legume cover crop (*Pueraria phaseloides*) in an oil palm plantation.

MATERIALS AND METHODS

This experiment was conducted in an oil palm field at Seafield Estate, Batu Tiga, Selangor. The oil palm trees were 2 years old with the inter rows covered with *Pueraria phaseoloides* mixed with indiginous grasses, *Paspalum conjugatum* and *Ottochloa nodosa*. Twenty plots of $1 \text{ m} \times 1 \text{ m}$ were randomly choosen along the inter rows of the oil palm trees, such that the mixtures between grass and legumes were varied. These plots were cut to about 5 cm above the ground.

Nitrogen fertilizer as ammonium sulphate labelled with 10% atom excess N-15 was applied in solution form at the rate of 2 g N/m² (20 kg N/ha). The plants in these plots were harvested after 3 months.

The centre $0.75 \text{ m} \times 0.75 \text{ m}$ were harvested and samples divided into legume and grass. These were dried in an oven at 65° C until constant weight is achieved and then ground for analysis. Some of the samples were sent to FAO/ IAEA Biotechnology Laboratory at Seibersdorf, Austria for N-15 analysis using mass spectrometer. Total nitrogen was analysed using the modified Kjeldahl procedure as described by Cottenie *et al.*, 1982.

RESULTS AND DISCUSSION

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Dry Matter Yield

The dry matter yield of legume was found to be higher than the grass (Table 1). This may be due to the rate of growth of the legume being faster than that of the grass (Whiteman, 1974). From the dry matter yield obtained, the different percentages of legume and grass mixtures of each plot were calculated and these were divided into plots with 0 - 40%, 41 - 60%, 61 - 80% and 81 - 100% legume standing. The number of plots for each legume to grass ratio were 5, 3, 4 and 8 respectively.

TABLE 1 Dry matter yield of legume and grass (g/m²)

Plot no.	Legume	Grass	
put, Plat		Paspalum	
1	214.56	64.66	seamnan d via k adar i
2	42.72		
3	222.43	12.62	48.48
4	52.66	0.48	vortunio Put
5	118.49	67.34	and notpusos
6	180.67	57.01	they wales
7	76.89	8.55	net o m one
8	128.60	19.57	and grad a
9	94.31	70.31	lot t <u>en</u> mon
10	146.36	23.89	-
11	103.32	1.69	-
12	60.67	2.13	96.19
13	97.03	100.64	
14	59.00	41.37	58.03
15	153.06	17.49	r platfings i
16	59.07	were Parpa	117.76
17	86.10	in the intern	78.20
18	173.01	39.16	ere mind S
19	5.28	2.36	192.87
20	18.18	total + and	126.27

Nitrogen Concentration

Table 2 shows the N concentrations in Pueraria, Paspalum and Ottochloa. The highest N concentration in Pueraria was observed in plots where there were 81 - 100% legume, but these were found to be not significantly different from the N concentrations in other plots. There

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Percent	Percent N		
Legume	Pueraria	Paspalum	Ottochloa
81 - 100	2.84 ^a	1.80ª	tro <u>ce</u> n fixi
61 - 80	2.59 ^a	1.49 ^a	Puteraria
41 - 60	2.64 ^a	1.75 ^a	1.55 ^a
0 - 40	2.68 ^a	1.92 ^a	1.81 ª

TABLE 2 Average N concentration (%) in legume and grass

Means in the same column followed by the same letter are not significantly different at P = 0.05.

was also no significant difference between the N concentrations in the grasses in the different plots. However, the average N concentration of Pueraria was found to be significantly higher than that in the grasses.

Total Nitrogen

The total N content in Pueraria was also found to be higher than that in the grasses. This is due to the higher % N and dry matter yield of Pueraria present in all the plots. The same observation was made by Watson et al., (1964a, 1964b) and Gibson, et al., (1977). The N content in Pueraria present in the plots with 61 - 80% legume was significantly higher (P 0.05) than those of all the other plots (Table 3).

TABLE 3 Total N content of Pueraria, Paspalum and Ottochloa

Percent	Total N content (g/m ²)			
Legume	Pueraria	Paspalum	Ottochloa	
81 - 100	2.98 ^b	0.32 ^a	ISSON, A.B	
61 - 80	4.65 °	0.94 ^a	inod s a i	
41 - 60	2.46 ab	1.52 ^a	1.21 ^a	
0-40	1.30 ^a	0.42 ^a	2.00 ^a	

Means in the same column followed by the same letter are not significantly different at P = 0.05.

% N-15 Atom Excess

The % N-15 atom excess in the Pueraria was found to be lower than in the grasses (Table

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4). This is to be expected since the N obtained by Pueraria from the soil and fixation from the atmosphere has reduced the N-14: N-15 ratio in the tissues (Hardason et al., 1983). The percent N-15 atom excess found in Pueraria grown in the different legume: grass composition was found to be not significantly different. The same observation was found in the % N-15 atom excess of Paspalum and Ottochloa.

TABLE 4 Average % N-15 excess present in the legume and grass

Percent	% N-15 atom excess			
Legume	Pueraria	Paspalum	Ottochloa	
81 - 100	0.136 ^a	0.615 ^a		
61 - 79	0.077 ^a	0.474 ^a	0.472 ^a	
41 - 60	0.061 ^a	0.677 ^a	0.789 ^a	
0 - 40	0.127 ^a	0.325 ª	0.429 ^a	

Means in the same column followed by the same letter are not significantly different at P = 0.05.

Percent Nitrogen Fixed by Pueraria

The percentage of nitrogen fixed by Pueraria was calculated using the formula proposed by Fried and Middelboe (1977) and Fried and Broeshart (1981). From the results obtained, a plot was made following the Cate and Nelson (1971) procedure to obtain the critical Pueraria: grass ratio required to obtain > 80% nitrogen fixed by Pueraria (Fig. 1). It was found that in order to obtain > 80% nitrogen fixation by Pueraria, the mixture should contain $\leq 80\%$ legume.

Total Nitrogen Fixed by Pueraria

The total nitrogen fixed by the Pueraria was obtained by multiplying the % N fixed by total nitrogen in Pueraria. The highest amount of nitrogen fixed was found to be in the plots with 61-80% legume, which amounted to 37.80 kg N/ha/3 months. This is due to the high dry matter produced which influenced the number of nodules present on the legume roots. Thus

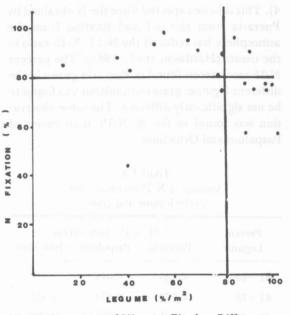
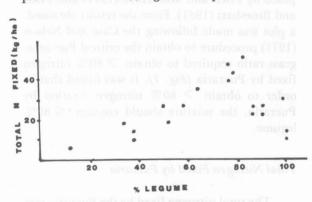


Fig. 1: Percentage of Nitrogen Fixed at Different Percentages of Legume – grass mixture.

leading to an increase in nitrogen obtained from fixation (Hardy et al., 1977).

When the values of the amount of nitrogen fixed by Pueraria was plotted against percentage of legume present in all the plots (*Fig. 2*), it was shown that the highest nitrogen fixed was from plots with about 80% legume composition.





CONCLUSION

The highest total nitrogen derived from fixation by Pueraria phaseloides was calculated

to be 37.80 kg Na/ha/3 months in the plots with 61 - 80% legume stand. This is equivalent to 151.2 kg N per hectare per year. This value is lower than those found by other workers who have used other indirect methods for calculating nitrogen fixed by legume cover crops. It was also found that the highest amount of nitrogen fixed by Pueraria was when the mixture contained about 80% legume.

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