Chapter 6

Macaranga denticulata and AM-fungi Association

6.1 Introduction

In the previous chapter the beneficial effect of a pioneer species, *Macaranga denticulate*, on rice yields was established with a series of crop cutting survey from 2000 to 2004. However, the effect could be detected with high stand density of *Macaranga* of some 4,000 plants ha^{-1}. The patchiness of *Macaranga* in the landscape of shifting cultivation fields, resulted in variable rice yields from 0.71 to 4.53 ton ha^{-1}. Understanding the key factors and mechanisms determining survival and establishment of *Macaranga* may lead to increase number of survival in fallow periods and stabilize rice yields across the landscape. In this situation, the soil is fairly poor with variable soil pH an average of about 4.8 and available phosphorus 3-4 ppm. Some 30 species of arbuscular mycorrhizal (AM) fungi was found in the rhizosphere of *Macaranga denticulata* growing in the farmers' fields (Youpensuk et al., 2004).

The purpose of this experiment was to test whether AM-fungi could influence growth and nutrient uptake of *Macaranga* seedlings

6.2 Materials and Methods

A pot experiment was set up for a period of 6 months from September 2001 to February 2002 at the Plant Genetic Resource and Nutrition Laboratory of the Faculty of Agriculture, Chiang Mai University. The soil for the experiment was collected from Mae Hia experiment station on upland fields with a depth between 0-50 cm below soil surface (Table 6.1). The soil was acidified to pH 4.9 with $Al_2(SO_4)_3.18H_2O$ 1g/kg soil. This was to simulate acidity of the soil of the study site at Tee Cha soils were sterilized by steaming at 95 -100 °C for 4 hrs before 17 kg of soil was placed in each 30 cm in diameter, 30 cm deep, pots. The seeds (imbibed overnight in water) were planted on 1 September 2001 and thinned to one plant per pot at 7 days after seedling emergence. All pots received water once a day to meet optimum requirement. Seedlings were sprayed with insecticide once a month to control aphids. The treatments were factorial combination with two levels each of AM inoculation (M_0 = nil; M_+ = inoculated), N (N_0 = nil; N_+ = 120 kg N ha^{-1}) and P (P_0 = nil; P_+ = 120 kg P ha^{-1}), in three replications. The AM fungi inoculum consisted of 70 gsoil mixed from *Macaranga* root zone from farmer's field in Tee Cha, placed in each hole in the pot before sowing *Macaranga* seed. The N_+ treatment consisted of a total of 4.23 g pot^{-1} of urea and P_+ treatment 0.46 g pot^{-1} of triple super phosphate, both in 2 equal applications, the first 2 weeks after emergence, and every 2 weeks after that.

Plant height was measured every month for six months. Harvesting was undertaken after six months after emergence to determine shoot and root weight. Shoots were cut off at the ground level and roots were recovered by washing. Both shoots and roots were dried at 80 ^{0}c for 48 hr for dry weight determination and the materials were finely ground for laboratory analysis of N, P, K, Ca and Mg.

Table 6.1 Soil characteristics from Mae Hia research station and Tee Cha village.

Soil characteristics	Mae Hia research station	Tee Cha village
Soil texture	Sandy loam	Sandy loam
Soil pH	5.5-5.7	4.83-4.93
Organic mater (%)	1.27-1.31	3.43-3.84
Nitrogen (%)	0.064-0.066	0.21-0.29
Phosphorus (ppm)	3.60-4.59	3.00-4.29
Potassium (ppm)	48-50	156-196
Calcium (ppm)	2.51-2.58	0.47-0.75
Magnesium (ppm)	0.98-1.15	0.48-0.67

6.3 Results

6.3.1 Height of *Macaranga* seedling

Plant height was increased by AM fungi inoculation and N and P application through out the study period (Figure 6.1). The plant grew most slowly in $M_0N_0P_0$. Growth rate was increased with the application of M, N or P, but most strongly in $M_+N_+P_+$, $M_+N_+P_0$ or $M_0N_+P_+$. There was significant interaction between the effect of M and P ($P < 0.05$) from one month after seedling emergence (Table 6.2). The effects of M and P on height were generally interchangeable (Figure 6.2). With or without N, plant height was increased to about the same extent by P or M, and applying M and P together had about the same effect as P or M alone.

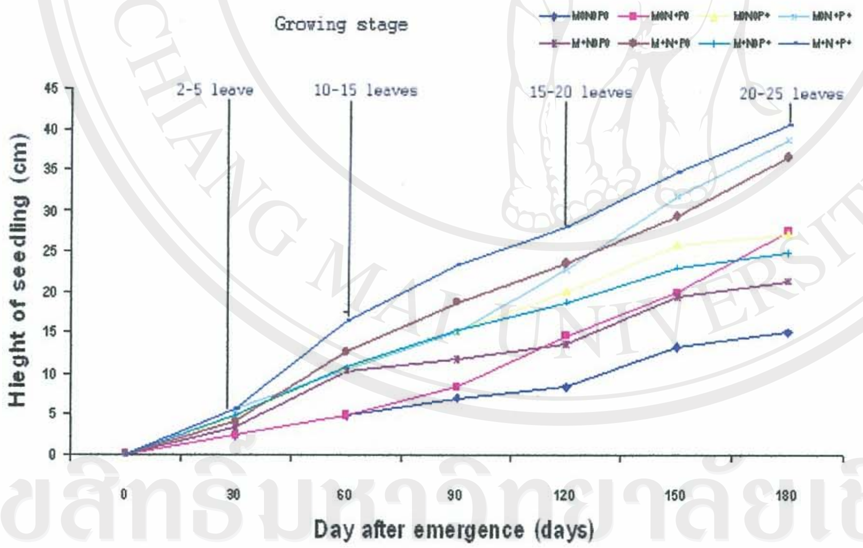


Figure 6.1 Changes in seedling height of *Macaranga*, AM inoculation (M_0 = nil; M_+ = inoculated), N (N_0 = nil; N_+ = 120 kg N ha^{-1}) and P (P_0 = nil; P_+ = 120 kg P ha^{-1})

Table 6.2 Height (cm) of *Macaranga* seedling after emergence on effect of mycorrhiza fungi, N and P.

Treatment	Day after emergence (days)					
	30	60	90	120	150	180
$M_0N_0P_0$	1.8	4.3	6.0	8.7	13.3	15.0
$M_0N_+P_0$	4.2	12.0	16.7	19.3	25.7	27.0
$M_0N_0P_+$	2.3	5.7	10.0	14.3	19.7	27.3
$M_0N_+P_+$	5.5	14.7	20.7	22.3	31.7	38.7
$M_+N_0P_0$	3.2	9.3	12.0	13.7	19.7	21.3
$M_+N_+P_0$	4.5	14.7	19.0	23.0	29.3	36
$M_+N_0P_+$	4.5	10.3	14.7	18.7	23.0	25.0
$M_+N_+P_+$	5.8	16.2	22.0	23.0	34.7	40.7
Significant difference by F-test						
M	*	*	*	*	*	*
N	*	*	*	*	*	*
P	*	*	*	*	*	*
M x N	NS	NS	NS	NS	NS	NS
M x P	*	*	*	*	*	*
N x P	NS	NS	NS	*	NS	NS
M x N x P	NS	NS	NS	NS	NS	NS

Note: M=Mycorrhiza, N=Nitrogen, and P=Phosphorus

* = significant difference at $p < 0.05$, NS= No significant

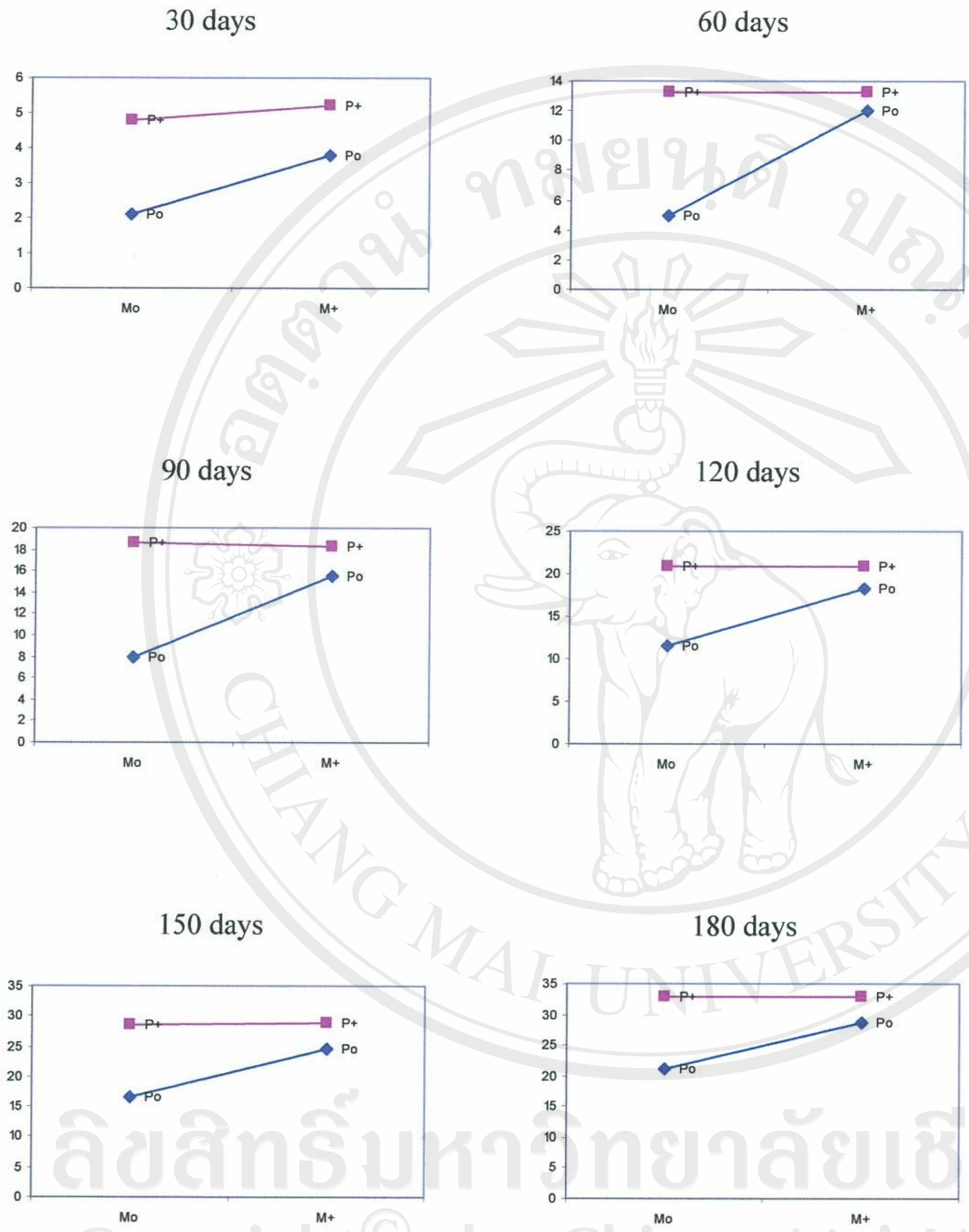


Figure 6.2 Effects of interaction between Mycorrhyza (M_0 = nil; M_+ = inoculated) and P (P_0 = nil; P_+ = 120 kg P ha^{-1}) on height (cm) of *Macaranga* seedling.

6.3.2 Shoot, root and total dry weight

Shoot, root and total dry weight were increased by AM fungi inoculation and N and P application (Table 6.3). The effects of N and P on the plant dry weights were dependent on the AM fungi inoculation; and there was also an interaction between the effect of N and P (N x P, M x N and M x P significant P<0.05). The interaction between M and N was positive. Mycorrhiza had little effect in N_0. While dry weight was increased with N_+, N application had bigger effect on dry weight with M_+. Similar positive interaction was found between the effect of N and P on plant dry weights. Application of N and P together had larger effects on plant dry weights than when N and P were applied separately. On other hand, there was negative interaction between M and P. Plant dry weights were increased by either M or P, and each having about the same effect as when both M and P were applied together. These effects were generally similar on dry weight of shoot (Figure 6.3a) and root (Figure 6.3b), and when shoot and root were combined into total dry weight (Figure 6.3c).

In general M and P tended to increase shoot:root ratio, whereas N tended to depress shoot:root ratio (Table 6.3). There was significant interaction between M and P ($P < 0.05$) on their effects on the shoot:root ratio, and also between and N and P ($P < 0.05$), but different direction; but there was no interaction between M and N. Shoot: root ratio was increased to about the same level, i.e. shoot was increased more compared with root, by M or P separately, and MP together had the same effect as M or P separately. On the other hand, shoot:root ratio was slightly depressed when P was applied without N and slightly increased when N and P were applied together. The shoot:root ratio was increased by M and depressed by N, without significant interaction between the two factors (NS at $P < 0.05$).

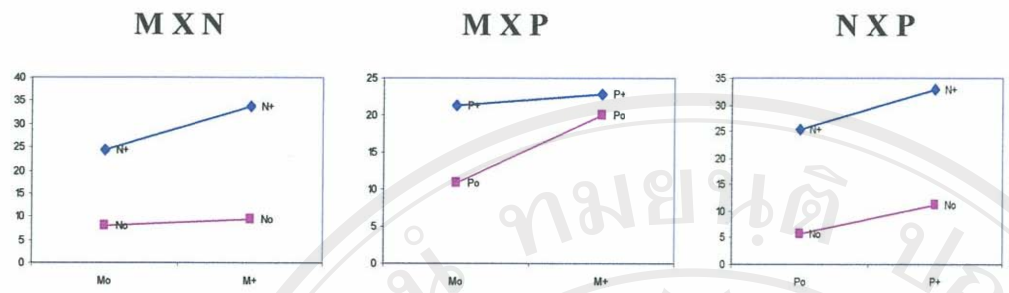
Table 6.3 Effects of AM fungi inoculation and N and P application on dry weight of *Macaranga* seedling at 6 months.

Treatment	Dry weight (g plant^{-1})			Root : Shoot ratio
	Shoot	Root	Total	
$M_0N_0P_0$	3.71	2.00	5.71	0.54
$M_0N_+P_0$	18.16	7.77	25.93	0.43
$M_0N_0P_+$	11.94	9.64	21.58	0.81
$M_0N_+P_+$	30.48	19.85	50.33	0.65
$M_+N_0P_0$	7.95	6.84	14.79	0.86
$M_+N_+P_0$	32.24	18.91	51.15	0.59
$M_+N_0P_+$	10.58	7.96	18.54	0.75
$M_+N_+P_+$	35.01	25.25	60.26	0.72
Significant difference by F-test				
M	*	*	*	*
N	*	*	*	*
P	*	*	*	*
M x N	*	*	*	NS
M x P	*	*	*	*
N x P	NS	*	*	*
M x N x P	NS	NS	NS	NS

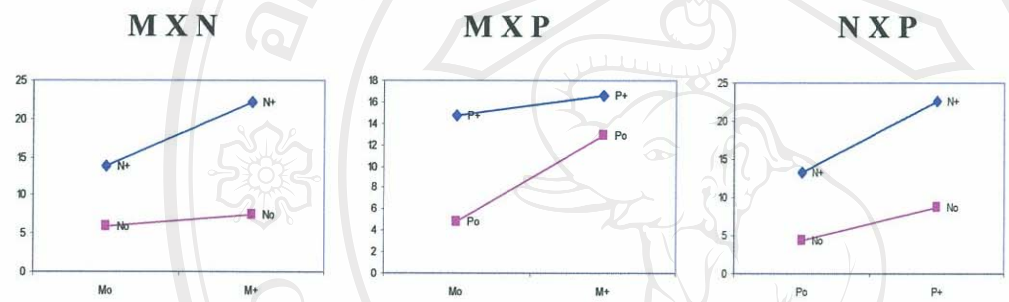
Note: M=Mycorrhiza, N=Nitrogen, and P=Phosphorus

* = significant difference at $p < 0.05$, NS= No significant

a) Shoot dry weight



b) Root dry weight



c) Total dry weight

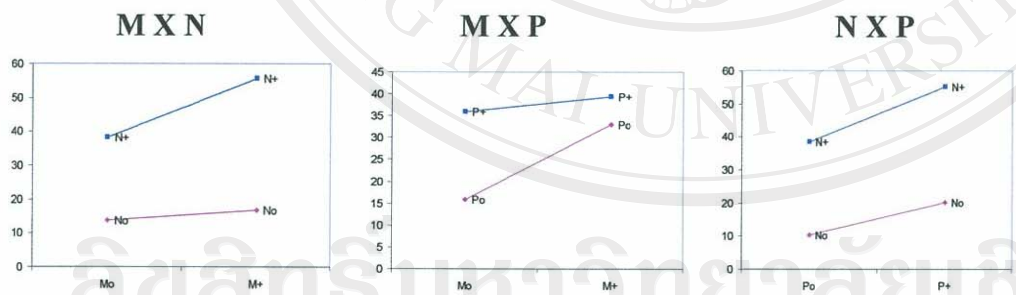


Figure 6.3 Interaction effect of Mycorrhiza (M_0 = nil; M_+ = inoculated) and fertilizer (N_0 = nil; N_+ = 120 kg N ha^{-1}; P_0 = nil; and P_+ = 120 kg P ha^{-1}) on shoot(a), root(b) and total(c) dry weight of *Macaranga* seedling.

6.3.3 Nutrient accumulation

Analysis of shoot dry matter for N, P, K, Ca and Mg found that M, N and P increased accumulation of all these nutrients in *Macaranga denticulata.* Shoot contents of N, P, K, Ca and Mg were highest in $M_+N_+P_+$, with 53.7 mg N, 9.00 mg P, 92.76 mg K, 53.42 mg Ca, and 12.23 mg Mg $plant^{-1}$. Shoot of plant in the nil treatment had only 7.40 % N, 8.87 % P, 13.31 % K, 10.48 % Ca and 13.57 % Mg of $M_+N_+P_+$ (Table 6.4). There was significant interaction ($P<0.05$) between the effect of M and P, and also between M and N but in different direction, on the shoot contents of N, K, Ca and Mg. Shoot contents of these nutrients were increased to about the same extent by M or P or both together, while they were increased by N application in M0, but more strongly with M+. Nitrogen application increased shoot contents of N, K, Ca and Mg, independently of P (P x N not significant at $P < 0.05$) (Table 6.4 and Figure 6.4). Phosphorus content of the shoot was affected differently, only P x N was significant ($P < 0.05$). The shoot P content was increased very slightly by P, more strongly by N, with the largest increase when P and N were applied together.

Table 6.4 Shoot nutrient content of *Macaranga* seedling 6 months after germination affected by mycorrhiza fungi inoculation and N and P fertilizer.

Treatment	Nutrient content (mg plant^{-1})				
	N	P	K	Ca	Mg
$M_0N_0P_0$	3.98	0.80	4.36	5.60	1.66
$M_0N_+P_0$	31.05	3.02	22.20	24.26	6.36
$M_0N_0P_+$	10.02	3.04	10.26	20.63	5.64
$M_0N_+P_+$	49.45	8.15	30.98	41.47	11.81
$M_+N_0P_0$	7.32	1.88	7.25	12.94	3.60
$M_+N_+P_0$	50.30	5.15	30.84	42.32	12.48
$M_+N_0P_+$	10.20	2.94	8.80	16.34	4.97
$M_+N_+P_+$	53.78	9.02	32.76	53.42	12.23
Significant difference by F-test					
M	*	*	*	*	*
N	*	*	*	*	*
P	*	*	*	*	*
M x N	*	NS	*	*	*
M x P	*	NS	*	*	*
N x P	NS	*	NS	NS	NS
M x N x P	NS	NS	NS	NS	NS

Note: M=Mycorrhiza, N=Nitrogen, and P=Phosphorus

* = significant difference at $p < 0.05$, NS= No significant.

Furthermore in case of root part, it was found that all treatment effected on nutrient accumulation significantly different at $P<0.05$. Root nutrient accumulated in treatment with $M_+N_+P_+$ highest were 21.05 mg N, 6.49 mg P, 29.53 mg K, 28.35 mg Ca and 14.49 mg mg Mg $plant^{-1}$ (Table 6.5). There was significant interaction ($P<0.05$) between the effect of M and N, on the root contents of N, P, K, Ca and Mg and between M and P but in different direction, on the shoot contents of K, Ca and Mg. Root contents of these nutrients were increased to about the same extent by M or P or both together, while they were increased by N application in M0, but more strongly with M+. The interaction of P x N are significant ($P < 0.05$) on the root content of N, P and Ca but not significant on K and Mg (Figure 6.5).

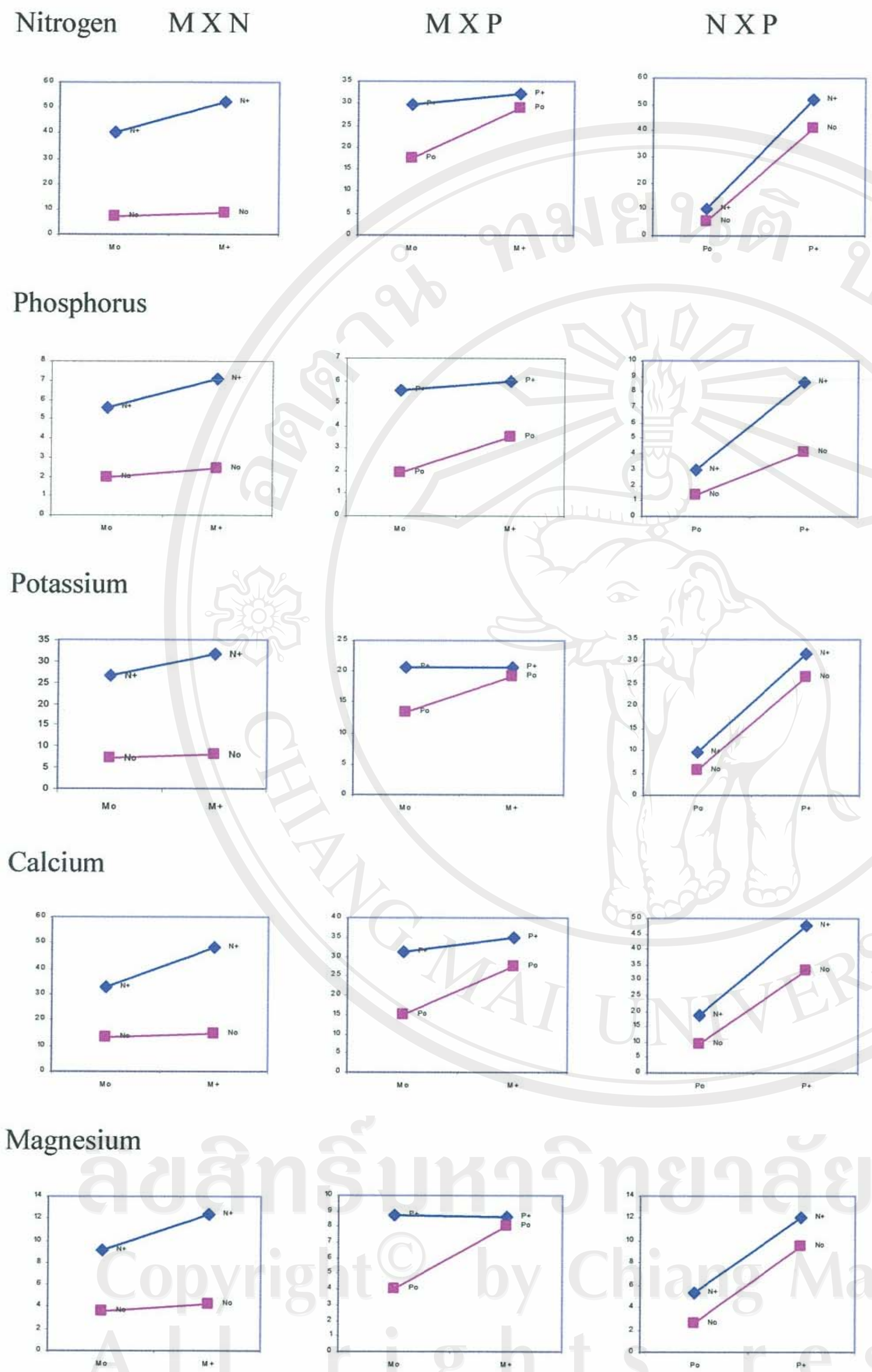


Figure 6.4 Interaction effects of Mycorrhiza (M_0 = nil; M_+ = inoculated) and fertilizer (N_0 = nil; N_+ = 120 kg N ha^{-1}; P_0 = nil; and P_+ = 120 kg P ha^{-1}) on nutrient contents in shoot.

Table 6.5 Root nutrient contents of *Macaranga* seedling 6 months after germination affected by mycorrhiza fungi inoculation and N and P fertilizer.

Treatment	Nutrient content (mg plant^{-1})				
	N	P	K	Ca	Mg
$M_0N_0P_0$	1.53	0.41	2.69	1.87	1.47
$M_0N_+P_0$	7.28	1.26	11.26	8.18	5.56
$M_0N_0P_+$	4.85	4.05	13.35	9.22	7.33
$M_0N_+P_+$	16.31	6.87	25.46	26.83	13.02
$M_+N_0P_0$	3.89	1.93	9.31	7.12	4.61
$M_+N_+P_0$	15.39	2.39	24.50	21.25	12.47
$M_+N_0P_+$	5.22	4.28	12.34	8.20	5.09
$M_+N_+P_+$	21.05	6.49	29.53	28.35	14.49
Significant difference by F-test					
M	*	*	*	*	*
N	*	*	*	*	*
P	*	*	*	*	*
M x N	*	*	*	*	*
M x P	NS	NS	*	*	*
N x P	*	*	NS	*	NS
M x N x P	NS	NS	NS	NS	NS

Note: M=Mycorrhiza, N=Nitrogen, and P=Phosphorus

* = significant difference at $p < 0.05$, NS= No significant

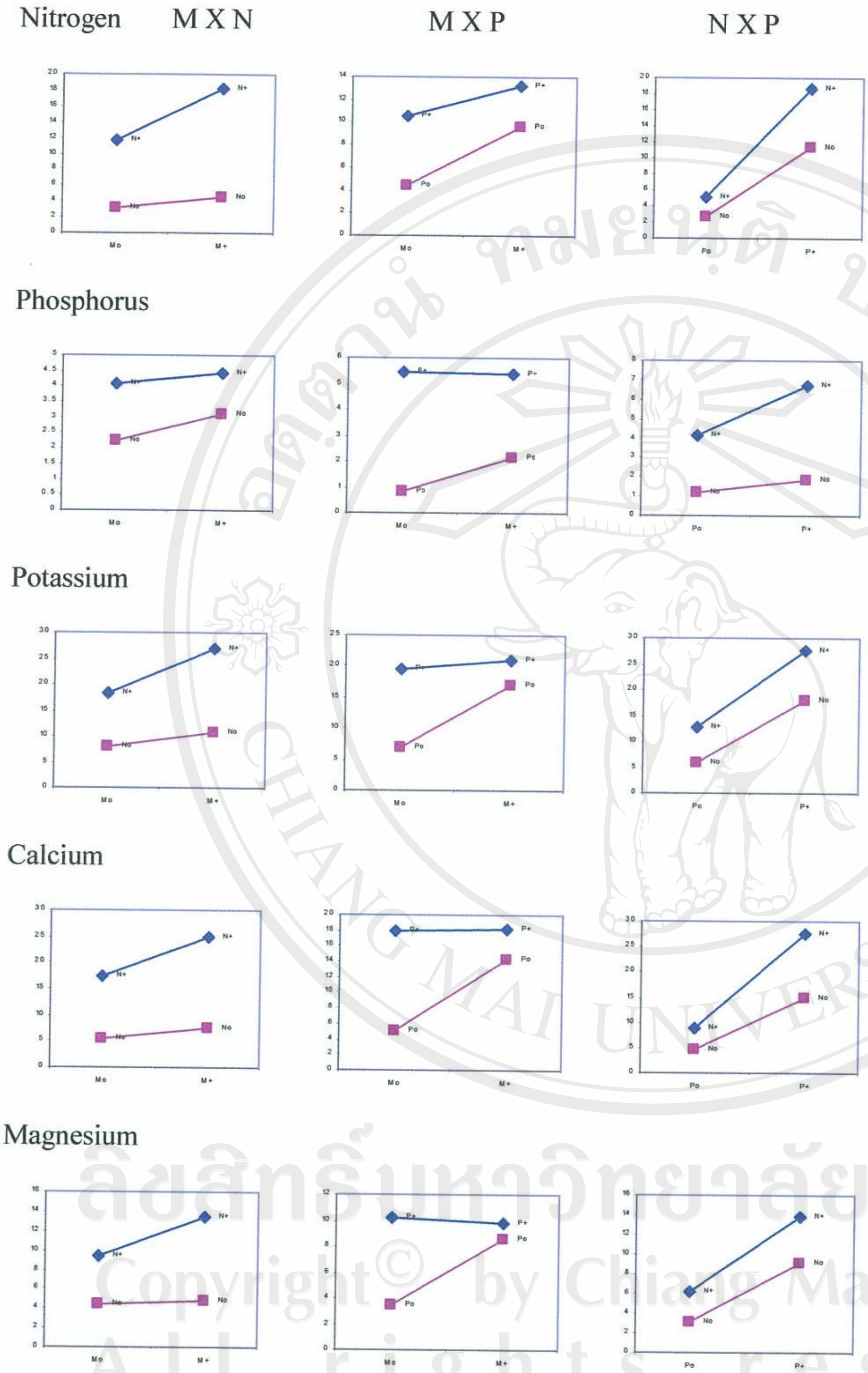


Figure 6.5 Interaction effect of Mycorrhiza (M_0 = nil; M_+ = inoculated) and fertilizer (N_0 = nil; N_+ = 120 kg N ha^{-1}; P_0 = nil; and P_+ = 120 kg P ha^{-1}) on nutrient content in root.

For analysis of total plants dry matter for N, P, K, Ca and Mg found that M, N and P increased accumulation of all these nutrients in *Macaranga denticulata.* Total plant nutrient contents of N, P, K, Ca and Mg were highest in $M_+N_+P_+$, with 74.83 mg N, 15.51 mg P, 62.29 mg K, 81.77 mg Ca, and 26.72 mg Mg plant^{-1}. Total of plant in the nil treatment had only 7.36 % N, 7.80 % P, 11.32 % K, 9.14 % Ca and 11.71 % Mg of $M_+N_+P_+$ (Table 6.6). There was significant interaction ($P<0.05$) between the effect of M and P, on the total plant contents of N, P, K, Ca and Mg and interaction between M and N was significant, on the plant contents of N, K, Ca and Mg but in different direction. Total plant contents of these nutrients were increased to about the same extent by M or P or both together, while they were increased by N application in M0, but more strongly with M+. For N and P interaction was significant ($P< 0.05$), on the total plant content of P and Ca, But N, K and Mg not significant (Figure 6.6).

Table 6.6 Total nutrient content of *Macaranga* seedling 6 months after germination affected by mycorrhiza fungi inoculation and N and P fertilizer.

Treatment	Nutrient content (mg plant^{-1})				
	N	P	K	Ca	Mg
$M_0N_0P_0$	5.51	1.21	7.05	7.47	3.13
$M_0N_+P_0$	38.33	4.28	33.46	32.44	11.92
$M_0N_0P_+$	14.87	7.09	23.61	29.85	12.97
$M_0N_+P_+$	65.76	15.02	56.44	68.3	24.83
$M_+N_0P_0$	11.21	3.81	16.56	20.06	8.21
$M_+N_+P_0$	65.69	7.54	55.34	63.57	24.95
$M_+N_0P_+$	15.42	7.22	21.14	24.54	10.06
$M_+N_+P_+$	74.83	15.51	62.29	81.77	26.72
Significant difference by F-test					
M	*	*	*	*	*
N	*	*	*	*	*
P	*	*	*	*	*
M x N	*	NS	*	*	*
M x P	*	*	*	*	*
N x P	NS	*	NS	*	NS
M x N x P	NS	NS	NS	NS	NS

Note: M=Mycorrhiza, N=Nitrogen, and P=Phosphorus

* = significant difference at $p < 0.05$, NS= No significant

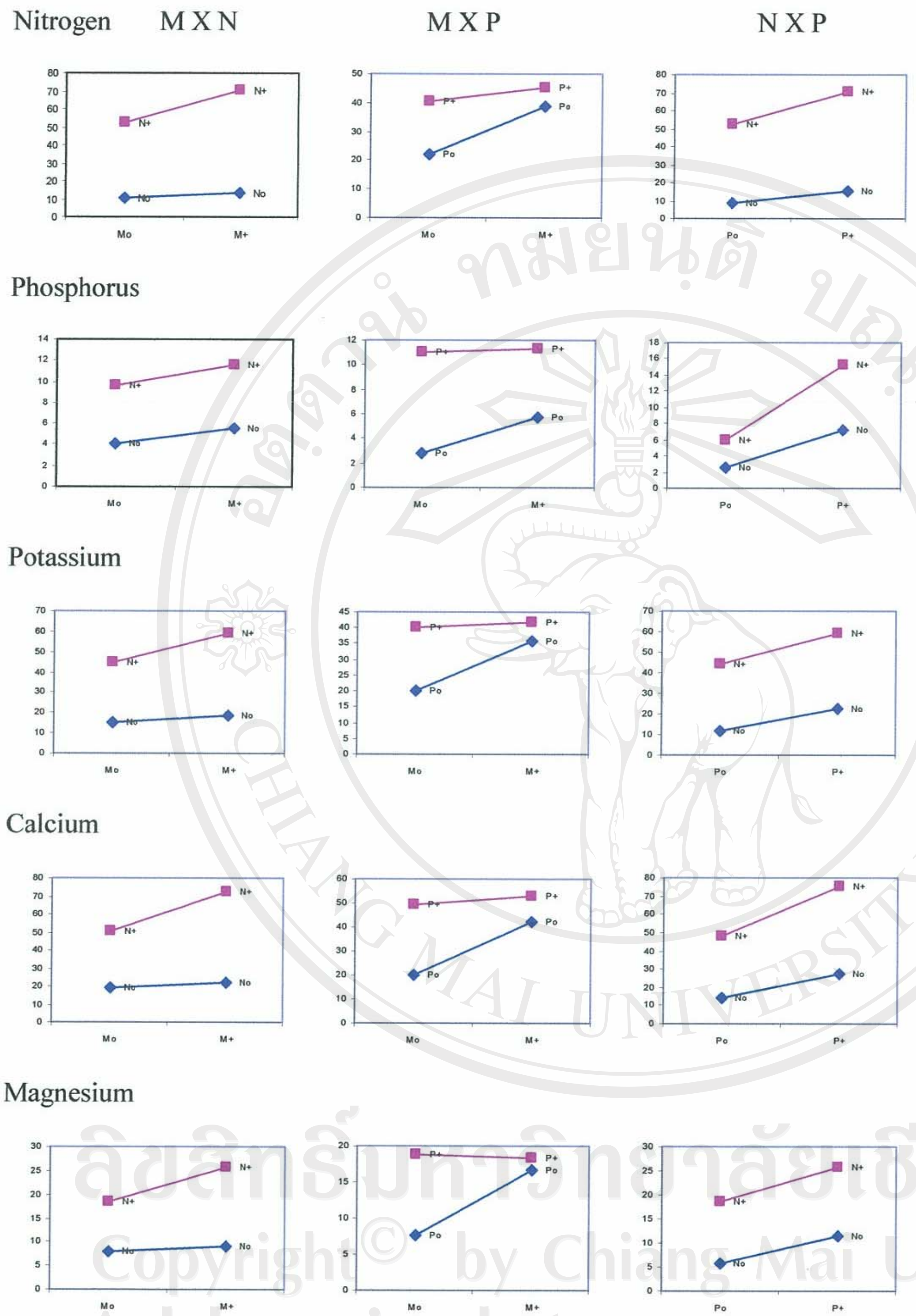


Figure 6.6 Interaction effect of Mycorrhiza (M_0 = nil; M_+ = inoculated) and fertilizer (N_0 = nil; N_+ = 120 kg N ha^{-1}; P_0 = nil; and P_+ = 120 kg P ha^{-1}) on nutrient content in total plant.

6.4 Discussion

This experiment has clearly established the strong effects of AM fungi on growth and nutrient uptake of *Macaranga* an acidic, low P soil. In both plant height and dry weight, *Macaranga* responded strongly when P was limiting (P_0) but N was not limiting (N_+). The abundance of AM fungi, some 30 species in 6 genera, found in the fallow at this site (Youpensuk *et al*., 2004) would have provided sufficient inoculants for the establishment for the new regeneration cycle of the fallow.

That growth and nutrient uptake of some plant species can be enhanced by AM fungi is well known. Smith and Read (1997) found that Mycorrhiza increased plant growth, nutrient accumulation and to the use of soil nutrients. Others (e.g. Marschner and Dell 1994; Taylor and Harrier 2001) reported that, AM fungi improved the nutritional status of plants (e.g., N, P, K, Ca, Mg, Mu, Cu and Zn) resulting in increased growth. In addition to the effect of AM fungi in enhancing nutrient uptake, the partitioning of considerable proportion of nutrients below ground may have also contributed to the value of *Macaranga* as a fallow enriching species, is supported by 2 pieces of evidence. One of these is the result from this chapter, showing partitioning of some 30% to 50% of nutrients to the roots. The other process is suggested by data set presented in chapter 5: (a) below ground biomass that accounted for one quarter of the total biomass accumulated in the fully cycle fallow, and (b) dry season leaf fall, in which the *Macaranga* shed all of its leaves except young leaves on new meristems, contributing to some 36% more litter under the fallow with dense than sparse *Macaranga*.