

Effects of Nitrogen, Phosphorus and Potassium Levels on Kenaf (*Hibiscus cannabinus* L.) Growth and Photosynthesis under Nutrient Solution

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Abstract

To date, little is known about the effect of levels of nitrogen, phosphorus and potassium on kenaf grown in nutrient solution culture. The objective of the present study was to examine the effects of different nitrogen, phosphorus and potassium levels on kenaf growth such as diameter, plant height, leaf number, root dry weight, stem dry weight and leaf dry weight and physiology like chlorophyll content, photosynthesis and stomatal conductance. Treatment consisted of 5 different levels of nitrogen viz. 0, 50, 100, 200 and 400 mg/L and 5 different levels of phosphorus and potassium viz. 0, 25, 50, 100 and 200 mg/L replicated thrice in a completely randomized design in a shade house. Growth (diameter, plant height and leaf number), chlorophyll content and photosynthesis were measured once weekly and plant components biomass was measured, 28 DAT. Different levels of nitrogen, phosphorus and potassium had significant effects on all the parameters studied. The highest values for diameter, plant height, leaf number, root dry weight, stem dry weight, leaf dry weight, photosynthesis and stomatal conductance were obtained from 200N, 100P and 100K whereas values decreased with further increase in levels of nutrient concentration. All the growth rates, chlorophyll content and photosynthesis declined with lower level of nitrogen, phosphorus and potassium. Among the plant components, leaf dry weight had the greatest decrease while root/shoot ratio increased under N deficiency. The results of this study provide new knowledge to produce kenaf with better nutrient management in the field.

Keywords: Kenaf, Growth, Chlorophyll content, Photosynthesis, Nutrition

1. Introduction

There has been an increase in interest in growing kenaf throughout the world for its high biomass yield and the elevated fibre content. Kenaf (*Hibiscus cannabinus* L.), a fast growing C₃ plant native of tropical Africa, is being

investigated as new source of bioenergy as well as an industrial crop and has high potential to be used in Malaysia. The high biomass yield and the elevated fibre content of kenaf justify the growing interest on this multipurpose crop for its potential role in agroecosystems involving biomass production as substitute of non-renewable resources (Cosentino and Copani, 2003).

Nitrogen, phosphorus, potassium and water are considered as the major limiting factors in crop growth, development and finally economic yield (Glass, 2003; Parry, *et al.*, 2005). To grow kenaf the responses of plants to N, P and K fertilization are of considerable importance in agriculture. Although N, P and K frequently limits growth and development of several crop species under field conditions, the precise mechanisms by the limitation occurs are complex and variable depending on species, developmental stage and environment. Limited N, P and K supply decreases rates of cell division, cell expansion and cell permeability (Roggatz *et al.*, 1999), photosynthesis, leaf production, and growth, plants (Chapin, 1980; Clarkson and Hanson, 1980; Evans, 1983; Radin and Boyer, 1982; Sinclair and Horie, 1989; Reddy *et al.*, 1997a; Gerik *et al.*, 1998; Zhao *et al.*, 2003, 2005a, b) and yield (Zhao *et al.*, 2007). Some reports suggest that N deficiency affects more strongly the leaf development than photosynthesis (Watson, 1952; Wong, 1979; Radin and Boyer, 1982; Reddy *et al.*, 1997a). Field and Mooney (1986) and Gerik *et al.* (1998) in their reviews described many of the responses of plants to N, P and K deficiency. They described the effects of low N, P and K nutrition on plants as causing lower photosynthetic rates and slower leaf expansion. Muchow (2009) reported that photosynthetic capacity of kenaf increased with specific leaf nitrogen from 0 to 24 g N m⁻².

For better fertilizer management the study about the effects of different levels of N, P and K on the kenaf growth and photosynthesis is very crucial. We hypothesize that critical N, P and K levels vary depending on growth and physiological processes, and growth and physiological processes differ in their responses to different levels of N, P and K. The objectives of this study were to investigate the effects of N, P and K on kenaf plant growth and physiology in nutrient solution culture to find out optimum levels of N, P and K for nutrient management practices of kenaf, and to derive functional relationship between N, P and K levels and different growth and physiological processes.

2. Materials and methods

2.1 Site description and plant materials

An experiment was conducted at the Experimental Farm No. 2, Universiti Putra Malaysia, Serdang, Selangor, Malaysia (2°59' 20.56"N, 101°42' 44.42"E) under a shade house for a period of 28 days (since 12 June to 10 July 2009). The photosynthetically active radiation (PAR) during the growing period was 500-700 $\mu\text{mol m}^{-2}\text{s}^{-1}$. Cultivar V36 of kenaf was used in this experiment as plant material.

2.2 Seedling growth

The kenaf seeds were sown directly on sand beds for germination. After 10 days, seedlings with similar size of plumule and radicle were selected for placing into the polypropylene tray containing Hoagland's nutrient solution (Hoagland and Arnon, 1950).

2.3 Treatments

The seedlings were placed into a polypropylene tray containing 20L of aerated Hoagland's nutrient solution. The following treatments were imposed: (1) five levels of N - 0, 50, 100, 200 and 400 mg/L and (2) five levels of P and K - 0, 25, 50, 100 and 200 mg/L. Each treatment was divided into three replications of 10 plants per replication. The Hoagland's nutrient solution was modified substituting CaCl₂ for Ca(NO₃)₂ to allow the different N concentration (Reddy *et al.*, 1996). It was also modified to obtain the different levels of P and K as required by the treatments. The nutrient solution was renewed every 7 days. All the plants were harvested 28 days after transplanting (DAT) in the nutrient solution.

2.4 Growth measurements

Plant height, diameter and leaf numbers were determined on five plants in each replication at 7-day intervals throughout experimental period. Stem lengths were measured as the distance between cotyledon level and the stem apex. Measurement of collar diameter was made using a digital caliper. Leaf number was counted when the main veins were first visible.

Biomass was collected from the plants under all the treatments at the end of the experiment. Roots, stems and leaves were separated and dried at 70°C in an electric oven for 48 hours to constant weight to estimate plant components dry weights. From these measurements, partitioning to above and below ground parts along with root to shoot ratios was calculated.

2.5 Physiological measurements

During treatment period (28 DAT), chlorophyll content of the third fully expanded leaf from the top was measured using a portable chlorophyll meter (Minolta SPAD-502, Japan) from 8:00 to 11:00 h at 7-day intervals. Net photosynthesis rates and stomatal conductance of the uppermost, expanded main stem leaves, which were the third from main axis terminal, from five plants in each treatment were measured between 8:00 and 11:00 h using an open gas exchange system, LI-6400 portable photosynthesis system (LiCOR Inc., Lincoln, Nebraska, USA).

2.6 Data analysis

Growth measurements such as diameter, plant height, leaf numbers, spad value and photosynthesis rates and stomatal conductance were averaged over the replications for all measuring dates and their means were used in deriving relationships between growth and physiological parameters. The differences in growth and physiology between the N, P and K treatments were calculated by using the SAS statistical procedure PROC. ANOVA and Duncan's Multiple Range Test (DMRT) at $p < 0.05$ (SAS Institute Inc., 2007). Correlation test with significance level reported ($p < 0.05$ or $p < 0.01$) was based on Pearson's correlation coefficient.

3. Results and discussion

The strategy of growing kenaf plants until 28 days of placement in Hoagland's nutrient solution and imposing various N, P and K treatments worked well for the purpose of achieving variability in plant growth and physiological parameters and to derive functional relationships between various processes and N, P and K levels. Biomass accumulated measured at 28 DAT was very sensitive to N, P and K treatments and plants grown under N, P and K deprivation showed significantly ($p < 0.05$) lower total biomass as well as all plant components dry weights (Table 1-3) similar to earlier findings (Zhao *et al.*, 2005a, b; Sinclair, 1990). Total plant biomass accumulated was less by 97%, 92% and 77%, respectively for the 0N, 0P and 0K treatments. Of all the plant dry weight components, leaf weight was lower by 98%, 94% and 87% for the plants under 0N, 0P and 0K treatments, respectively ($p < 0.05$), compared to the 200N, 100P and 100K treatments (Table 1-3). Therefore, N, P and K affected several processes in kenaf plant that ultimately led to lower the biomass production. The biomass production in root, stem and leaf increased up to 200N, 100P and 100K treatments and then declined. This might be due imbalanced nutrient concentration or toxicity caused by the higher nutrient concentration.

Diameter and plant height during the experimental period showed significant differences among the treatments with regards to the level of nutrient concentrations. Plant height extension that includes all growing internodes, expansion of all leaves and effective PAR capture and interception (Alm *et al.*, 1991; Morrison *et al.*, 1994; Reddy *et al.*, 1997b) will affect overall canopy development (Reddy *et al.*, 1997a, 2004; Gerik *et al.*, 1998). During the 28 days of treatment, there were significant differences in stem lengths due to N, P and K treatments (Table 1-3). Plants in the 200N, 100P and 100K were taller than the other treatments on all measured dates and at higher nutrient concentrations after that plant height decreased. Plants grown in 200N, 100P and 100K treatments were 76%, 64% and 58% taller than the plants grown in the 0N, 0P and 0K treatments, respectively (Table 1-3). Shorter plants under N, P and K deficiency might have been due to their effects on cell elongation as well as cell division (Roggatz *et al.*, 1999).

Leaf number was significantly different for every level of nutrient concentration. Leaf numbers increased in all treatments over time up to 200N, 100P and 100K. Fewer numbers of leaves produced under deficient N, P and K conditions might be due to their indirect effect since node addition rates are primarily controlled by temperature and modulated carbon demand/supply (Reddy *et al.*, 1997a, 2004). Decreased net photosynthesis under N, P and K deficient conditions observed in this study would result in lower carbon supply which will result in fewer leaves per plant in N, P and K stressed kenaf plants.

Leaf chlorophyll content was significantly influenced by the levels of N, P and K and declined in 0N, 0P and 0K treatments during the treatment period due to growth. The chlorophyll content of the leaf was negatively correlated with the age of the plants during the treatment period at 0 levels of N, P and K. (Fig 1-3).

Photosynthesis was significantly increased with the increase of nutrient concentration up to 200N, 100P and 100K and afterwards, decreased (Fig. 4-6). Photosynthesis of 200N, 100P and 100K were $14.45 \mu\text{mol m}^{-2}\text{s}^{-1}$, $12.56 \mu\text{mol m}^{-2}\text{s}^{-1}$ and $12.83 \mu\text{mol m}^{-2}\text{s}^{-1}$, respectively, when averaged across the measuring times. The lowest photosynthesis was observed in 0N, 0P and 0K treatments. This finding is in agreement with Reddy and Matcha (2009). They found much steeper declining photosynthesis in 0N compared to the 100N in other crop experiment. Similar to leaf photosynthesis, stomatal conductance declined with lower N, P and K levels (Fig. 7-9); the decline however, was slightly less responsive with N, P and K levels. Since internal carbon dioxide

concentration did not show any significant differences among N, P and K treatments over time except at very low N, P and K levels in the 0N, 0P and 0K treatments (data not shown; Schulze, 1986), the decline photosynthesis at low N, P and K levels might be due to both greater stomatal resistance and the less biochemical efficiency of chloroplasts (Chapin, 1980; Reddy *et al.*, 1996).

4. Conclusion

Our results show that kenaf plant growth such as diameter, stem elongation and leaf number are positively correlated with N, P and K levels until a certain level. In addition, N, P and K deficiency decreased plant height and photosynthesis of kenaf plants leading to lower biomass accumulation. Maximum growth and photosynthesis rates were achieved at 200N, 100P and 100K. Among the plant components, leaf dry weight had the greatest decrease under N, P and K deficiency while root/shoot ratio increased under N deficiency. Furthermore, the functional relationships between N, P and K level and growth and physiological processes might be useful to develop better nutrient management practices of kenaf plants that typically encounter under field conditions.

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Table 1. Influence of nitrogen nutrition levels on kenaf plant growth and dry weights of different plant parts

Parameters	Nitrogen treatment (mg/L)				
	0	50	100	200	400
Diameter, mm	2.57d	5.73c	7.58b	8.71a	7.51b
Plant height, cm	13.33c	44.67b	48.06b	55.48a	47.30b
Leaf number, plant ⁻¹	4.33d	10.33c	11.33c	16.67a	14.00b
Root dry weight, g plant ⁻¹	0.04c	0.45b	0.49b	0.64a	0.50b
Stem dry weight, g plant ⁻¹	0.04d	0.64c	1.07b	1.33a	0.97bc
Leaf dry weight, g plant ⁻¹	0.02c	0.49b	0.52b	1.07a	0.82ab
Total dry weight, g plant ⁻¹	0.10d	1.58c	2.08b	3.04a	2.29b
Root/shoot ratio	0.67a	0.40b	0.31b	0.27c	0.28c

Mean values followed by the same letter(s) do not differ statistically as per Duncan's Multiple Range Test at $p \leq 0.05$

Table 2. Influence of phosphorus nutrition levels on kenaf plant growth and dry weights of different plant parts

Parameters	Phosphorus treatment (mg/L)				
	0	25	50	100	200
Diameter, mm	1.78d	6.86c	7.57b	8.42a	7.50b
Plant height, cm	15.85c	38.17b	42.20a	43.86a	42.18a
Leaf number, plant ⁻¹	3.33d	11.33c	12.33b	14.33a	12.00b
Root dry weight, g plant ⁻¹	0.08c	0.28b	0.36b	0.63a	0.38b
Stem dry weight, g plant ⁻¹	0.10c	0.75b	0.79b	1.43a	0.95b
Leaf dry weight, g plant ⁻¹	0.05c	0.44b	0.52b	0.93a	0.62b
Total dry weight, g plant ⁻¹	0.23c	1.47b	1.67b	2.99a	1.95b
Root/shoot ratio	0.53a	0.24b	0.27b	0.27b	0.24b

Mean values followed by the same letter(s) do not differ statistically as per Duncan's Multiple Range Test at $p \leq 0.0$

Table 3. Influence of potassium nutrition levels on kenaf plant growth and dry weights of different plant parts

Parameters	Potassium treatment (mg/L)				
	0	25	50	100	200
Diameter, mm	2.86d	5.08c	6.86ab	7.48a	6.67b
Plant height, cm	18.53d	30.46c	35.71b	43.83a	40.70a
Leaf number, plant ⁻¹	6.33c	12.67b	13.67b	15.33a	13.77b
Root dry weight, g plant ⁻¹	0.15b	0.17b	0.43ab	0.78a	0.74a
Stem dry weight, g plant ⁻¹	0.20c	0.55b	0.72b	1.13a	0.93ab
Leaf dry weight, g plant ⁻¹	0.14d	0.23c	0.65b	1.09a	0.90b
Total dry weight, g plant ⁻¹	0.69c	0.95c	1.80b	3.00a	2.57b
Root/shoot ratio	0.44a	0.35b	0.31c	0.35b	0.40b

Mean values followed by the same letter(s) do not differ statistically as per Duncan's Multiple Range Test at $p \leq 0.05$

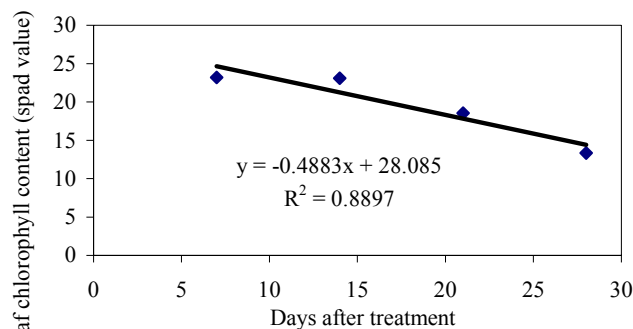


Fig. 1. Correlation between kenaf leaf chlorophyll content and 0 levels of nitrogen treatments during the treatment period, 28 days after placement.

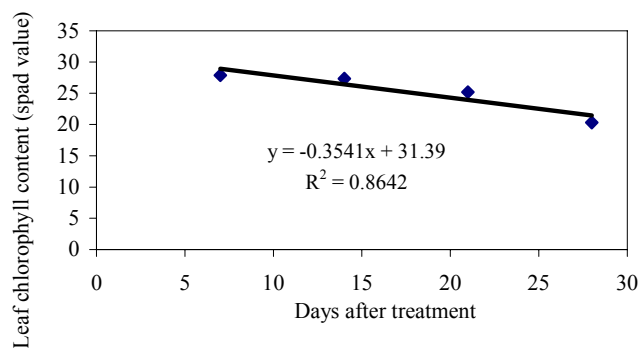


Fig. 2. Correlation between kenaf leaf chlorophyll content and 0 levels of phosphorus treatments during the treatment period, 28 days after placement.

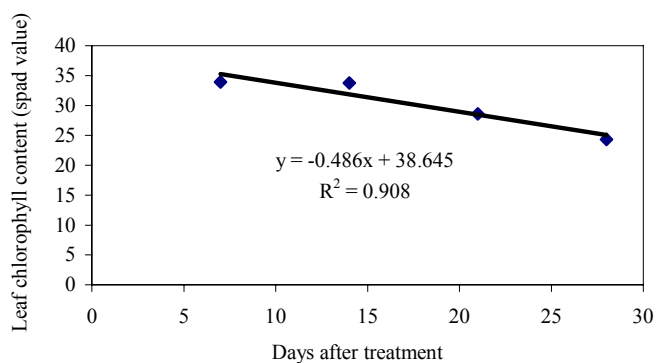


Fig. 3. Correlation between kenaf leaf chlorophyll content and 0 levels of potassium treatments during the treatment period, 28 days after placement.

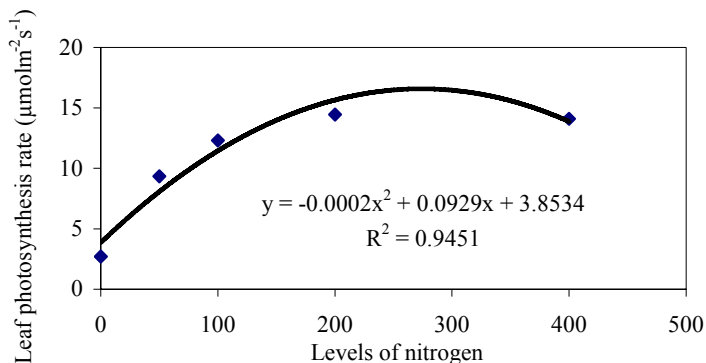


Fig. 4. Functional relationship between leaf net photosynthesis rate at 28 DAT and nitrogen levels

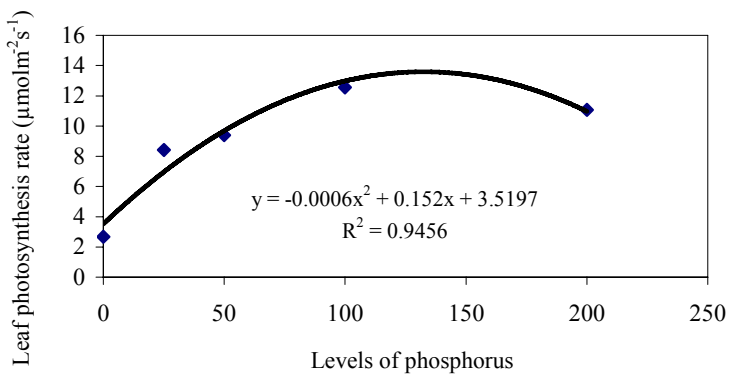


Fig. 5. Functional relationship between leaf net photosynthesis rate at 28 DAT and phosphorus levels

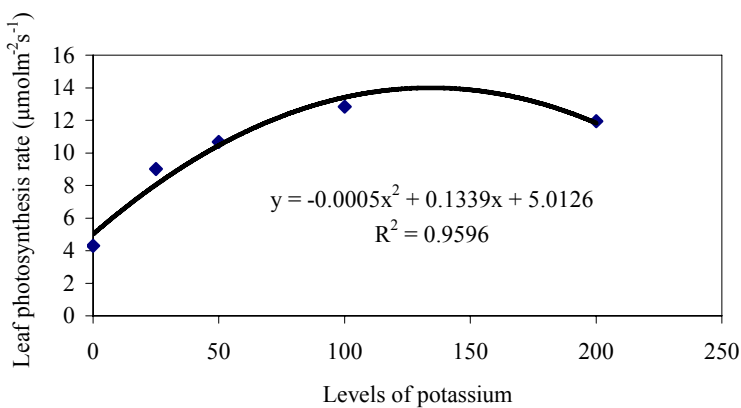


Fig. 6. Functional relationship between leaf net photosynthesis rate at 28 DAT and potassium levels

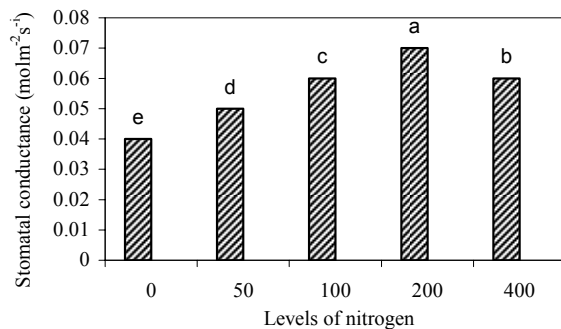


Fig. 7. Influence of levels of nitrogen on stomatal conductance of kenaf plant at 28 DAT

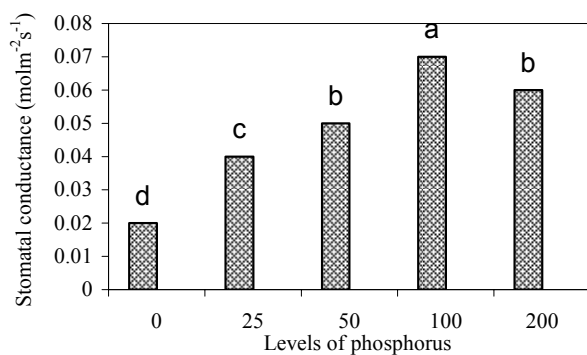


Fig.8. Influence of levels of phosphorus on stomatal conductance of kenaf plant at 28 DAT

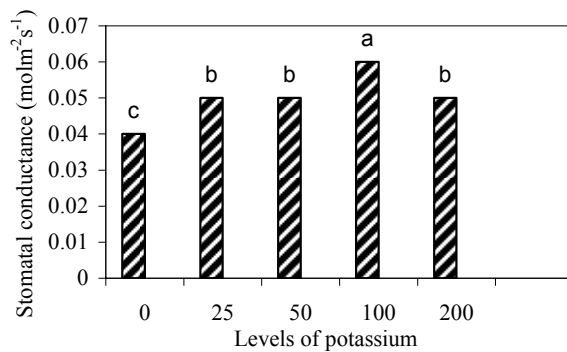


Fig. 9. Influence of levels of potassium on stomatal conductance of kenaf plant at 28 DAT