# Effect of Nitric Oxide on Root and Shoot Development of Rambutan (Naphelium lappaceum) Seedlings

G.M. HATHARASINGHE and S.J.B.A. JAYASEKERA

Department of Horticulture and Landscape Gardening, Faculty of Agriculture and Plantation Management, Wayamba University of Sri Lanka, Makandura, Gonawila (NWP), 60170, Sri Lanka

## ABSTRACT

The growth performance of a budded or grafted plant is enhanced by the root system of a rootstock plant species. *Nephelium lappaceum* seedlings are widely used as rootstocks in propagation of *rambutan* budded plants; hence its root system plays a major role for obtaining a better growth performance. In the present study, the effect of sodium nitroprusside as nitric oxide donor on root architecture and growth characteristics of *Nephelium lappaceum* seedling planted in polybags under net house conditions were investigated at  $2^{nd}$ ,  $4^{th}$  and  $6^{th}$  weeks after transplanting. Three treatments with 25, 50 and 75  $\mu$ M of sodium nitroprusside concentrations and a control treatment without sodium nitroprusside were used. Observations on seedling height, shoot fresh weight, length of tap root, root dry weight, number of leaves, number of lateral roots were recorded. All treatments recorded higher values for growth parameters over control. The treatment with 25  $\mu$ M showed the best mean in shoot and root growth of seedlings than other treatments over control. Vigorous and strong seedling for a rootstock could be developed through this method.

KEYWORDS: Nitric oxide, Rambutan, Rootstock, Root system, Sodium nitroprusside

### INTRODUCTION

Rambutan (Nephelium lappaceum) is a medium sized tropical popular fruit belonging to family Spandaceae. Rambutan is native to Malay Indonesian region and other regions of tropical Southeast Asia. In Sri Lanka best suited for low country wet zone (LCWZ) and mid country wet zone (MCWZ). Major growing areas are Gampaha, Kegalle, Rathnapura, Kandy, Galle, Matara. Potential areas for future expansion are Badulla and Monaragela. There are three recommended varieties of rambutan in Sri Lanka namely; Malwana Special, Malayan red and Malayan yellow. Malwana special is round shaped, with red coloured fruits.

Rootstock is the part which donates the root system for a budded or a grafted plant. Rootstock species possess certain qualities to consider as a rootstock. The particular plant selected as rootstock should have а compatibility with the scion, be hardy and vigorous to survive in harsh environment conditions. Most importantly the selected rootstock variety should possess a healthy vigorous and strong root system with good rooting ability and adaptability to various soil types.

Root system is an important part of a plant, normally located below ground level. Its primary functions are anchorage of the plant, absorption of water and dissolved minerals and conduction of these to the stem, and storage of food reserve. Plant can optimize their root architecture by initiating lateral root primordia and influencing growth of primary or lateral roots. The root system results from the coordinated control of both genetic endogenous factors (regulating growth and organogenesis) and the action of abiotic and biotic environmental stimuli (Nibau *et al.*, 2008). As seedling, it develops; lateral roots branch out from primary roots greatly increasing the total surface area and mechanical strength of the root system.

The role of nitric oxide (NO) is important in root initiation, development, metabolism and regulatory functions in plants (Guo and Crawford, 2005). Molecular, genetic and bio chemical studies have identified key players in signaling pathways, regulating growth and development as well as defenses (Durner and Klessig, 1999). Nitric oxide plays a central role in determining lateral roots development (Correa-Aragunde *et al.*, 2004) and is frequently used (Correa-Aragunde *et al.*, 2012; Chohan *et al.*, 2012; Kong *et al.*, 2014) to improve root growth.

Nayanakantha *et al.*, (2014) have reported that improved root architecture and growth of young budding plants of rubber using SNP as a donor of NO. Samaradiwakara and Jayasekera (2015) have reported improved root and shoot architecture and growth of *Citrus medica* seedlings using SNP as a donor of NO.

Objective of this study was to investigate the effect of NO on root and shoot development of rambutan under different concentration levels and produce rootstock of *Naphelium lappaceum* with maximum root development for production of better grafted plants for distribution among farmers.

## MATERIALS AND METHODS Experimental Site

This study was carried out in a net house at the Faculty of Agriculture and Plantation Management, Wayamba University of Sri Lanka, Makandura, situated in the low country intermediate zone (IL<sub>1a</sub>), at an elevation of 30 m above mean sea level. Experiment was carried out from March to May 2016.

Average rainfall at Makandura during the experimental period was 1.02 mm while the mean relative humidity and temperature were 80%, 32.7 °C respectively.

## Layout of Experiment

Treatments were arranged in Randomized Complete Block Design (RCBD) with three replicates. There were four treatments. Each treatment had six plants planted in black polybags.

## Growing of Rambutan Seedlings

The seeds of *rambutan* variety Malwana special were collected from a market, washed and the mucigeneous layer around the seeds was removed. Seeds were sown in a germination bed filled with sand. The seeds were allowed to germinate and continuous watering was done twice a day. After one month, uniformly germinated seedlings were selected and transplanted at the rate of one seedling per polybag filled with top soil, compost and sand in 1:1:1 ratio. Transplanted seedlings were regularly watered.

## Treatments with Sodium Nitroprusside (SNP)

Three different concentrations of SNP (Dihydrate; AnalaR NORMAPUR), 25 (T<sub>2</sub>), 50 (T<sub>3</sub>) and 75 (T<sub>4</sub>)  $\mu$ M, were compared with the control (T<sub>1</sub>) devoid of SNP. These concentrations were prepared at the laboratory prior to application. The treatments were applied seven days after transplanting at the rate of 50 mL per seedling twice a day continuously for five days. The control was provided with same amount of distilled water without SNP.

### **Measurements of Growth Parameters**

Growth characteristics and the root architecture of the seedlings were assessed at three stages. They were two weeks, four weeks and six weeks after transplanting. At each growth stage, seedling from each treatment were selected randomly and measurements were taken and recorded.

## Seedling Height (cm)

Height of the seedling was measured from the base to the tip of the stem.

#### Number of Leaves

The number of leaves in the seedling was counted.

### Shoot Fresh Weight (g)

Fresh Weight of the aerial part of the plants was measured.

### Number of Lateral Roots

Number of Lateral Roots on tap root was counted.

## Length of Taproot (cm)

The length of taproot was recorded from end of the root tip up to the base of the stem.

## Root Dry Weight (g)

Root dry weight was measured after oven drying the roots at 105 °C for 24 hours.

## Statistical Analysis

The data generated from the experiment were statistically analyzed using SAS statistical package (version 9.2).

#### **RESULTS AND DISCUSSION** *Growth Parameters*

Application of SNP improved the shoot and root characteristics of *Nephelium lappaceum* seedlings as compared to the control, at two weeks, four weeks and six weeks after transplanting in polybags.

The magnitude of increase in seedling height, shoot fresh weight, number of leaves, number of lateral roots, length of tap root and root dry weight was more profound after six weeks from transplanting (Tables 1, 2 and 3). All the concentrations of SNP, resulted increase of almost all the morphological characters recorded in *Nephelium lappaceum* seedlings over the control and 25  $\mu$ M SNP was more effective over the other concentrations.

In two week old Nephelium lappaceum seedlings difference in seedling height among treatments were not significant (Table 1). Higher value was recorded in  $T_2(31.93 \text{ cm})$  and lowest value recorded in T<sub>4</sub> (19.63 cm). Neither of the treatments showed any significant difference for shoot fresh weight (Table 1). However,  $T_1$  recorded lowest value (1.51 g) and T<sub>2</sub> recorded highest value (1.91 g). Differences in length of tap root were not significant (Table 1). Higher value was recorded in  $T_4$  (45.67 cm) and lowest value recorded in T<sub>2</sub> (37.00 cm) for number of lateral roots. Neither of the treatments showed any significant difference for root dry weight (Table 1). T<sub>1</sub> recorded lowest value (0.22 g) and T<sub>3</sub> recorded highest value (0.31 g). Neither of the treatments showed

Treatment	Seedling height (cm)	Number of leaves	Shoot fresh weight (g)	Length of tap root (cm)	Numbers of lateral roots	Root dry weight (g)
T <sub>1</sub>	19.83	15.67	1.51	43.00	18.88	0.22
T₂	31.93	12.50	1.91	37.00	15.43	0.29
<b>T</b> <sub>3</sub>	26.73	9.25	1.58	40.67	22.58	0.31
T₄	19.63	10.0	1.65	45.67	10.60	0.24
SE	. 8.75	7.60	3.87	5.44	17.49	0.18
CV	35.98	34.5	144.5	41.6	43.03	60.98

Table 1. Mean growth parameters of seedlings at after two weeks after transplanting

 $T_{1}$ - Control,  $T_{2}$ - 25  $\mu M$ ,  $T_{3}$ - 50  $\mu M$ ,  $T_{4}$ - 75  $\mu M$ 

## Table 2. Mean growth parameters of seedling at four weeks after transplanting

Treatment	Seedling height (cm)	Numbers of leaves	Shoot fresh weight (g)	Length of tap root (cm)	Numbers of lateral roots	Root dry weight (g)
T <sub>1</sub>	25.07	15.33	3.15	37.50	18.45	0.32
T <sub>2</sub>	33.18	13.50	3.39	53.83	24.45	0.34
Τ3	30.03	16.25	3.20	64.50	19.67	0.39
T₄	30.00	14.75	3.29	57.17 ·	22.62	0.40
SE	2.95	7.62	1.21	4.53	21.62	0.09
CV	9.88	32.5	37.24	21.30	40.60	26.05

 $T_{1}$ - Control,  $T_{2}$ - 25  $\mu$ M,  $T_{3}$ - 50  $\mu$ M,  $T_{4}$ - 75  $\mu$ M

#### Table 3. Mean growth parameters of seedling at six weeks after transplanting

Treatment	Seedling height (cm)	Numbers of leaves	Shoot fresh weight (g)	Length of tap root (cm)	Numbers of lateral roots	Root dry weight (g)
T <sub>1</sub>	30.63	15.33	4.04	55.2	16.42	0.33
T <sub>2</sub>	37.65	17.75	4.29	93.0	23.77	0.45
T <sub>3</sub>	32.93	11.00	3.15	61.5	18.22	0.40
T.	30.98	14.75	3.68	71.0	17.88	0.41
SE	4.70	7.56	0.75	5.64	24.56	0.07
CV	14.20	19.00	19.81	29.38	34.67	17.60

 $T_{1}$ - Control,  $T_{2}$ - 25  $\mu$ M,  $T_{3}$ - 50  $\mu$ M,  $T_{4}$ - 75  $\mu$ M

any significant difference for number of leaves and number of lateral roots (Table 1).

In four week old Nephelium lappaceum seedlings, difference in seedling height among treatments was not significant (Table 2). Higher value was recorded in T<sub>2</sub> (33.18 cm) and lowest value recorded in  $T_1$  (25.07 cm). Neither of the treatments showed any significant difference for shoot fresh weight (Table 2). T<sub>1</sub> recorded lowest value (3.15 g) and  $T_2$  recorded highest value (3.39 g). Differences in length of tap root among treatments were not significant (Table 2). Higher value was recorded in  $T_3$  (64.50 cm) and lowest value recorded in  $T_1$  (37.50 cm). Neither of the treatments showed any significant difference for root dry weight (Table 2). T<sub>1</sub> recorded lowest value (0.32 g) and T<sub>4</sub> recorded highest value (0.4 g). Neither of the treatments showed any significant difference for number of leaves and number of lateral roots (Table 2).

In six week old Nephelium lappaceum seedlings, difference in all the growth parameters among treatments were not significant (Table 3). Difference in seedling height among treatments were not significant (Table 3). Higher value was recorded in  $T_2$  (37.65 cm) and lowest value recorded in  $T_1$  (30.63 cm).

In shoot fresh weight higher value was recorded in  $T_2$  (4.29 g) and lowest value

recorded in T<sub>3</sub> (3.15 g) for length of tap root, T<sub>1</sub> recorded lowest value (55.2 cm) and T<sub>2</sub> recorded highest value (93.0 cm). In root dry weight higher value was recorded in T<sub>2</sub> (0.45 g) and lowest value recorded in T<sub>1</sub> (0.33 g).

Neither of the treatments showed any significant difference for numbers of leaves and number of lateral roots (Table 3). Maximum values were recorded for all parameters by 25  $\mu$ M treatment in six week old *Nephelium lappaceum* seedlings. After six weeks there was a significant difference in root dry weight.

During the six week period after treatments, it was evident that root and shoot development had occurred faster in all treated plants than the control treatment. This indicates that NO from SNP has affected the root growth by initiating a longer tap root and increased number of lateral roots. This root growth was observed more in T<sub>2</sub> than in T<sub>3</sub> and T<sub>4</sub>. The length of tap root in control treatment increased from 43 cm to 55.2 cm during the six week period, while the rate of increase in T<sub>2</sub> was from 37 to 93 cm during same period. The number of lateral roots in control decreased from 18.88 to 16.42 cm, while in T<sub>2</sub> treatment the lateral roots showed an increase from 15.43 to 23.77 during the same six week period. Similar trend was recorded for root dry weight as well. These results have shown clear indication of improved root development of the seedlings which would

help the plant to absorb more nutrients for better plant growth in treated plants than the control.

Improved root growth and absorption of nutrients has resulted in better shoot growth in treated plants than the control. This was evident from higher values recorded for seedling height, number of leaves and shoot fresh weight in treated plants during the six week period after treatments.

The increased growth performance of shoot has affected through efficient photosynthesis that would have taken place in the treated plants with higher number of leaves. However,  $T_2$  (25  $\mu$ M) proved to be a better treatment than others. These results are in par with the results obtained by Samaradiwakara and Jayasekera (2015) in *Citrus medica*.

## CONCLUSIONS

The SNP, NO source, would be recommended for improving plant growth during early stages of seedlings of *Rambutan*, to make them better rootstocks for budding and grafting purposes.

The treatment with 25  $\mu$ M of SNP has improved shoot and root characteristics, thus making the seedlings more vigorous and strong.

## **ACKNOWLEDGEMENTS**

Authors like to offer their sincere gratitude to Mr. K.H.M.I. Karunarathne, Information and Communication Technology (ICT) Centre, Wayamba University of Sri Lanka, for his advice in statistical analysis. The help given by the non-academic staff of the Faculty of Agriculture and Plantation Management is also acknowledged.

#### REFERENCES

Chohan, A., Parmar, U. and Raina, S.K. (2012). Effect of sodium nitroprusside on morphological charactersunder chilling stress in chick pea (*Cicer arietinum* L.). Journel on Environmental Biology, 33, 695-698.

- Correa-Aragunde, N., Graziano, M. and Lamattina, L. (2004). Nitric oxide plays a central role in determining lateral root development in tomato. *Planta*, **218**, 900-905.
- Durner, J. and Klessig, D.F. (1999). Nitric oxide as signal in plants. *Plant Biology*, **2**, 369-374.
- Guo, F.Q. and Crawford, N.M. (2005). Arabidopsis nitric oxide synthase 1 is targeted to mitochondria and protects against oxidative damage and dark induced senescence. *The Plant Cell*, **17** (12), 3436-3450.
- Kong, J., Dong, Y., Xu, L., Liu, S. and Bai, X. (2014). Effect of foliar application of salicylic acid and nitric oxide in alleviating iron deficiency induced chlorosis of *Arachis hypogaea* L. *Botanical studies*, 55 (9) 1-9.
- Nayanakantha, N.M.C., Pathirana, P.D., Senevirathna, A.M.W.K. and Senevirathne, P. (2014). The poly bagged plants of Rubber (*Hevea brasiliensis*). In: A.P. Keerthipala (ed.) proceedings of the 5<sup>th</sup> Symposium effect of Nitric Oxide donor Sodium Nitroprusside on root architecture and growth of young budding on Plantation Crop Research, Colombo,Sri Lanka, 15-17 October, 2014. 77-89.
- Nibau, C., Gibbs, D.J. and Coates, J.C. (2008). Branching out in new directions: the contrl of root architecture by lateral root formation. *New Phytologist*, **179**, 595-614.
- Samaradiwakara, S.H.M.R.N.P. and Jayasekera, S.J.B.A. (2015). Study on effect of nitric oxide on root and shoot development of *citrus medica* seedling. In: proceedings of 14<sup>th</sup> Agriculture Research Symposium, Makandura. Wayamba University of Sri Lanka. 59-63.