



EFFECT OF PLANT EXTRACTS ON THE SHOOT AND ROOT DEVELOPMENT OF NEEM (*Azadirachta indica*) SEEDLING

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ABSTRACT

An experiment was conducted at the Agroforestry and Environment Research Field, Hajee Mohammad Danesh Science and Technology University, Dinajpur from October 2015 to February 2016 to investigate the effect of plant leaf extracts on shoot and root development of neem (*Azadirachta indica*) seedlings. Five treatments namely: T₁ (pineapple leaf extract), T₂ (napier grass extract), T₃ (ginger leaf extract), T₄ (turmeric leaf extract) and T₅ (control i.e. tap water) were used. The experiment was laid out in a complete randomized design with five replications. Blended crop residues were mixed with water in a ratio 1:2 to prepare plant extract. Results showed that effect of crop residues on the shoot height of neem seedlings did not vary significantly in different treatments after 1 and 3 months of transplantation. Same trend was recorded in case of biomass, root collar diameter, number of leaves, sturdiness and central root length except shoot-ratio ratio. Significantly the highest shoot-root ratio was recorded in napier grass extract followed by turmeric leaf extract and the lowest ratio was in control treatments after 3 months of seedling transplantation. Root architectural analysis showed that there is no significant variation of mean length of first order lateral roots (FOLR) of neem seedlings among the treatments. Same trend was recorded for mean diameter of FOLR of the seedlings. In case of number of FOLR, the highest number was recorded in the neem seedlings treated by napier grass extract and lowest was found in ginger leaf extract after 3 months of transplantation. It can be concluded that though the tested crop residues did not affect significantly on the shoot development of neem seedlings, napier grass extract stimulated the root growth of neem seedlings. However, there was no negative effect of the tested plant leaf extract on the growth of neem seedling. Therefore, pineapple, napier, ginger and turmeric can be cultivated in association of with the neem tree in an agroforestry system.

Key words: ginger, napier grass, neem seedling, pineapple, turmeric

INTRODUCTION

Crop residues are commonly found in tree-crop interface of agroforestry system. In this tree-crop interface, both tree and crop interact with each other (Somarriba 1992). This interaction may be positive or negative. Positive interaction of agroforestry practices helps to overcome the deteriorating environment of the traditional agriculture (Tang and Yu 1999; Xu and Coventry, 2003). In agroforestry system, interactions between trees and crops are the heart because sharing of the common resources by different species (Van Noordwijk *et al.* 2015). Trees interact with crops by their root and shoot system and crops interact with trees by their residues (Zhang *et al.* 2013; Cao *et al.* 2012; Lin 2010). Allelopathy includes both positive

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and negative effects of one plant on the other through environment. Interplant interactions assume significance in agroforestry programmes for selecting the types of crops complementary to the selective tree species (Kohli *et al.* 1997). Interactions may be also above ground and below ground for crops and trees (Ong *et al.* 1991). The combination of woody perennials with crop and/or animals on the same unit of land management is an age-old practice in Bangladesh. Neem is an evergreen, multipurpose tree which is a versatile medicinal plant and the unique source of a vast number of photochemical with diverse chemical skeletons. In addition, different parts of neem and their extracts have been found to be safe, cheap and ecofriendly pesticides, insecticides, parasiticides and agrochemicals. Neem-based materials are compatible with integrated pest management (IPM); neem products do not persist in the environment and are degraded by ultraviolet rays and rainfall. In recent years, there has been an increasing trend and awareness in neem research. In fact, the time has come to make good use of centuries-old knowledge of neem through modern scientific approaches and techniques, so that such a unique gift of nature can be utilised to a greater extent. From the above discussion, it is thus evident that owing to its unique properties and applications in various areas of social need, this Divine tree demands special attention and interest from the world community (Brahmachari 2004).

Neem bark and leaves contain phenolic compound those act as allelochemicals (Xuan *et al.* 2004) which may affect the crops grown in agroforestry system. Many researches were done on the allelopathic tree residues on crops (Zhang and Fu 2010; Sisodia and Siddiqui 2010). However, these interactions should take place with respect to how the component of agroforestry utilize and share the resources of the environment and how the growth and development of any of the components will influence the others (Torquaebiae 1990). Many research works have been done on the effect of tree residues on different crops (Patrick *et al.* 1964). But little is known about the effect of different crop residues on trees (Monte Júnior *et al.* 2012). Now a days, pine apple, ginger, turmeric and napier grass are cultivating widely under neem trees. But there is very little information about the residual effect of these crops on neem seedlings. Considering these facts, the present study was undertaken to find out the effect of different crop residues on the neem plant's morphology. The specific objectives are as follows: i) To determine the effect of plant leaf extracts on the shoot and root growth of transplanted neem seedling and ii) to analyze the variation of biomass allocation and quality of the seedlings due to the effect of crop residues.

MATERIALS AND METHODS

The experiment was conducted in Agroforestry and Environment Research Field of Hajee Mohammad Danesh Science and Technology University (HSTU), Dinajpur from October 2015 to February 2016. The CRD design was followed with five replications in each treatment. Four plant leaf extracts and tap water were used as treatments. These were: T₁ = Pineapple leaf extracts, T₂ = Napier grass extracts, T₃ = Ginger leaf extracts, T₄ = Turmeric leaf extracts and T₅ = Water (control). For preparing crop extracts, 1 kg fresh crop leaves with twigs were blended in blender machine mixing with two litre water. Then the mixture was boiled for about 1 hour until the extracts reduce to 1 litre. After that the extracts were cooled and filtered. After filtering the extract were preserved in containers for further use. Naturally grown seedlings were collected under the neem trees near the Central Mosque of HSTU Campus. Similar size seedlings were collected from the trees in a same season and transplanted in the polybags of the size 9"×6". The age of the seedlings was 2 months during the time of seedling collection. After collection, all poly bags were placed on the experimental field. After transplantation, the polybags were placed on the open field. After one week of transplantation, plant leaf extracts were started to apply as treatments. There

were 12 seedlings for each treatment (4 polybags in each replications). Therefore, for five treatments there were 60 poly bags (5 treatments×12 polybags). Crop leaf extract were applied 15 days interval in all the treatments. The morphology of shoot and roots and their biomass were measured in four times. The parameters that were measured are shoot height (cm), root collar diameter (mm), length of central roots, number of living branches, shoot dry weight (g), root dry weight and total dry biomass. For biomass measurement, the seedlings were divided into two parts i.e. shoot and root. The roots were separated from the soil through gentle wash of water using a bucket and sieved to collect any root fragments detached with. Branches and leaves were mixed together to get the shoot weight. Both parts were oven-dried at 80°C (Royo *et al.* 2001; Tsakalidimi *et al.* 2009) for 72 hours until they reached in a constant weight. They were weighed through an electric balance to get shoot dry weight (g), root weight (g). Then total oven dry weight (g) was calculated. The root and shoot ratio was calculated by the root and shoot dry weights (Thompson 1985). The seedling quality index (QI) was calculated using the equation (Dickson *et al.* 1960):

$$\text{Quality index} = \frac{\text{Total Biomass (g)}}{\text{Height(cm)}/\text{diameter (mm)} + \text{shoot biomass(g)}/\text{root biomass (g)}}$$

All first-order lateral roots (FOLR) were counted and measured. For this, FOLRs were separated carefully by a sharp knife. For measuring lengths and diameters, each FOLR were numbered serially. Lengths of FOLRs were measured by a measuring tape (made of steel) and diameters of FOLR were measured by a caliper (accuracy 0.1 mm). All statistics were calculated with SPSS software (v.19.0 for Windows) and MS Excel 2007. Distribution was tested for normality by Kolmogorov–Smirnov criterion and the homogeneity of variances was tested by Levene’s test. Means were separated by the Duncan Multiple Range Test (DMRT).

RESULTS AND DISCUSSION

Above ground morphology: After 1 month of transplantation, shoot height of neem (*Azadirachta indica*) seedlings did not vary significantly in different treatments. Same trend was recorded in case of root collar diameter, number of leaves, sturdiness ratio and central root length

Table 1. Effect of crop residues on the morphological characteristics of shoot and root of *Azadirachta indica* seedlings in different treatments after 1 month of transplantation

Plant leaf extract	Shoot height (cm):H	Root Collar diameter (mm): D	Number of leaves	Sturdiness (H/D)	Central Root (cm)
Pineapple	15.83a ±1.74	4.570a ±.22	14.33a ±1.86	3.46a ±.33	10.73a ±.67
Napier grass	16.67a ±0.73	4.310a ±.23	13.00a ±1.53	3.89a ±.27	11.83a ±1.09
Zinger	14.30a ±1.21	4.273a ±.35	12.67a ±1.76	3.36a ±.21	13.00a ±1.26
Turmeric	14.17a ±3.17	4.413a ±.14	15.33a ±1.20	3.26a ±.79	14.50a ±1.32
Tap water (control)	13.70a ±1.60	4.443a ±.42	12.00a ±1.00	3.11a ±.39	11.83a ±1.88

In a column having same letter(s) did not differ significantly by Duncan Multiple Range Test at P≤0.05.

After 3 months of seedling transplantation, though shoot height increased than after 1 month but they also did not vary significantly in different treatments. Similar results were found by Dhillon *et al.* (1995) and Sain *et al.* (2001) in maize. Same trend was found in case of root collar diameter, number of leaves, sturdiness and central root length (Table 2).

Table 2. Effect of crop residues on the morphological characteristics of shoot and root of *Azadirachta indica* seedlings in different treatments after 3 months of transplantation

Plant leaf extract	Shoot height (cm):H	Root collar diameter (mm):D	Number of leaves	Sturdiness (H/D)	Central root (cm)
Pineapple	16.20a±1.74	5.13a±.31	9.67a±.67	3.16a±.28	22.67a±1.76
Napier grass	16.77a±0.90	5.06a±.18	9.33a±.33	3.33a±.24	21.17a±1.83
Zinger	14.20a±0.91	4.93a±.29	10.67a±1.67	2.90a±.27	20.50a±3.50
Turmeric	15.73a±1.36	5.23a±.15	10.00a±1.53	3.03a±.33	19.67a±2.17
Tap water (control)	14.33a±1.69	5.25a±.53	8.33a±1.45	2.74a±.26	22.33a±2.19

Biomass allocation: After 1 month of seedling transplantation, shoot and root dry biomass did not vary significantly among different treatments. As a result, total dry biomass, shoot/root ratio and quality index were found statistically similar in different treatments. This result is in conformation with Gao et. al. (2012) where they worked with biomass and allocation of young *Azadirachta indica* and *Acacia auriculiformis* for different restoration patterns in dry-hot valley.

Table 3. Effect of crop residues on biomass allocation and quality index of *Azadirachta indica* seedlings at different treatments after 1 month of transplantation

Plant leaf extract	Shoot dry biomass (g)	Root dry biomass (g)	Total dry biomass (g)	Shoot/root ratio	Quality index
Pineapple	0.80a±.06	0.43a±.07	1.23a±.09	1.97a±.38	0.23a±.02
Napier grass	0.77a±.09	0.57a±.12	1.33a±.03	1.57a±.43	0.26a±.03
Zinger	0.73a±.07	0.53a±.09	1.27a±.12	1.43a±.28	0.26a±.03
Turmeric	0.67a±.12	0.57a±.12	1.23a±.12	1.30a±.36	0.33a±.12
Tap water (control)	0.70a±.12	0.53a±.09	1.23a±.19	1.37a±.23	0.28a±.02

Shoot dry biomass did not vary significantly in different treatments. Same trend was recorded in case of root dry biomass, total dry biomass and quality index among different treatments. Significantly the highest (1.10g and 0.98g) shoot/root ratio was recorded in napier grass extract following turmeric leaf extract and lowest (0.60g) in Control. Shoot/root ratio varied significantly in pineapple and ginger leaf extracts with similar quality (Table 4).

Table 4: Effect of crop residues on biomass allocation and quality index of *Azadirachta indica* seedlings at different treatments after 3 months of transplantation

Plant leaf extract	Shoot dry biomass (g)	Root dry biomass (g)	Total dry biomass (g)	Shoot/root ratio	Quality index
Pineapple	1.80a±.15	2.10a±.21	3.90a±.35	0.86ab±.05	0.97a±.04
Napier grass	1.77a±.18	1.67a±.29	3.43a±.44	1.10a±.16	0.80a±.15
Zinger	1.53a±.19	1.80a±.10	3.33a±.28	0.84ab±.06	0.89a±.05
Turmeric	1.70a±.21	1.77a±.18	3.47a±.28	0.98a±.14	0.89a±.13
Tap water (control)	1.47a±.35	2.40a±.38	3.87a±.73	0.60b±.05	1.17a±.24

Length of first order lateral roots (FOLR): Mean length of FOLR did not vary significantly in different treatments after 1 month of transplantation due to the effect of different crop residues and this result confirmed by Thompson and Schultz (1995). After 3 months of transplantation, mean length of FOLR were also statistically similar in different treatment (Figure 1).

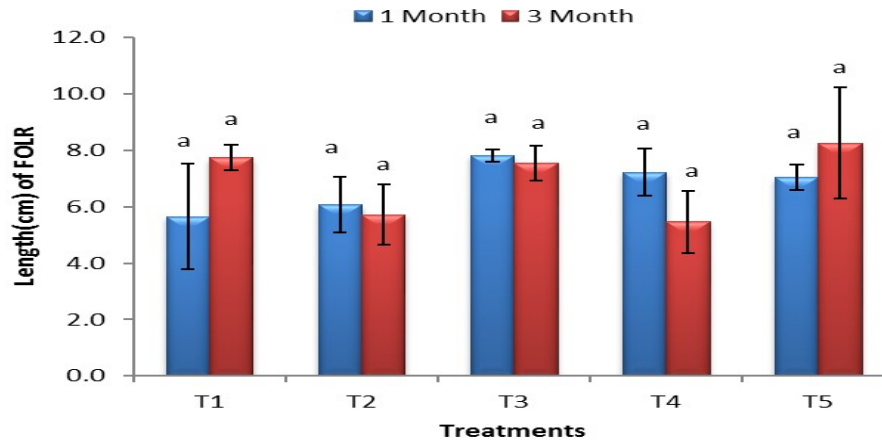


Figure 1. Mean length of FOLR of *Azadirachta indica* seedling in different time period among different treatments (In a month, bars having same letter(s) did not differ significantly by DMRT at $P \leq 0.05$). (T₁= Pine apple leaf extracts, T₂= Napier grass extracts, T₃= Zinger leaf extracts, T₄= Turmeric leaf extracts, T₅= tap water ; control).

Mean diameter of FOLR: Mean diameter of FOLR did not vary due to crop residues among the *Azadirachta indica* seedlings (Figure 2). Diameter of FOLR was recorded similar after 1-month of transplantation. Same trend was found in different treatments after 3 months of transplantation.

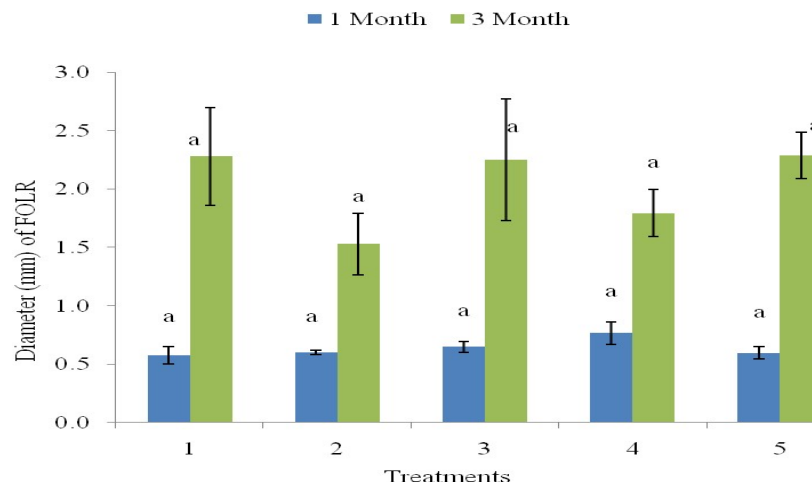


Figure 2. Mean diameter of FOLR of *Azadirachta indica* in different time period among different treatments.

Number of FOLR: Total number of FOLR per plant increased from 1 month to 3 months after transplantation but they were found similar after 1 month of transplantation. After 3 months, FOLR were recorded significantly different where the highest (5.00) average number of FOLR was found in T₂ (Napier grass extract) and lowest (2.67) was found in T₃ (Ginger leaf extract).

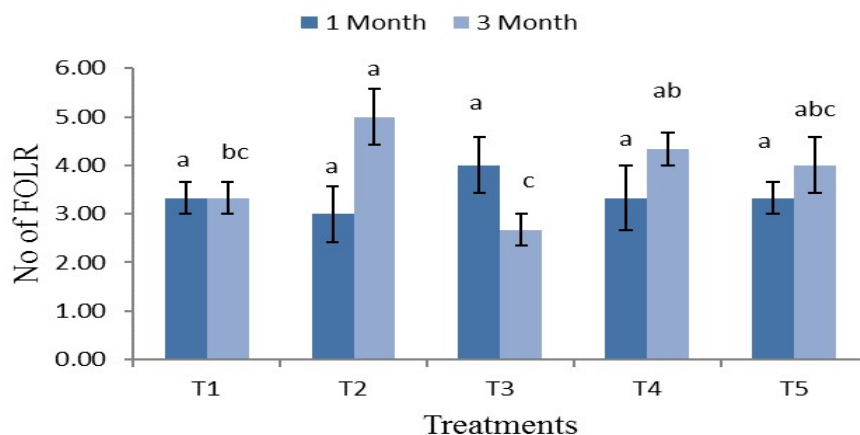


Figure 3. Average number of all FOLR of *Azadirachta indica* in different time period among different treatments.

CONCLUSIONS

It can be concluded that the tested four plant extracts such as pineapple, napier, ginger and turmeric had no negative effects on the shoot height, root growth and first order lateral roots of neem seedlings. Moreover, napier grass extracts had positively stimulating effects on the growth of root of neem seedlings.

REFERENCES

- Brahmachari G. 2004. Neem-an omnipotent plant: a retrospection. *Chembiochem*, 5(4): 408-421.
- Cao FL, Kimmins JP, and Wang JR. 2012. Competitive interactions in Ginkgo and crop species mixed agroforestry systems in Jiangsu, China. *Agroforestry systems*, 84(3): 401-415.
- Dhillon BS, Thind HS, Sazena VK, Sharma RK, Malhi NS. 1995. Tolerance to excess water stress and its association with other traits in maize. *Crop Improvement*. 22(1): 24-28
- Dickson A, Leaf AL, and Hosner JF. 1960. Quality appraisal of white spruce and white pine seedling stock in nurseries. *The Forestry Chronicle*, 36(1): 10-13.
- Gao C, Tang G, Sun Y, Zhang, C, Xie Q, and Li K. 2012. Biomass and allocation of young *Azadirachta indica* and *Acacia auriculiformis* for different restoration patterns in dry-hot valley. *Journal of Zhejiang A&F University*, 29(4), 482-490.
- Kohli RK, Batish D and Singh HP. 1997. Allelopathy and its implications in Agroecosystems, *Journal of Crop Production*, 1(1): 169-202.
- Lin BB. 2010. The role of agroforestry in reducing water loss through soil evaporation and crop transpiration in coffee agroeco systems. *Agricultural and Forest Meteorology*, 150(4): 510-518.

- Monte Júnior IP, Maia LC, Silva FS and Cavalcante UM. 2012. Use of plant residues on growth of mycorrhizal seedlings of neem (*Azadirachta indica* A. Juss.). *Journal of the Science of Food and Agriculture*, 92(3): 654-659.
- Ong CK, Corlett JE, Singh RP, and Black CR. 1991. Above and below ground interactions in agroforestry systems. *Forest Ecology and Management*, 45(1): 45-57.
- Patrick ZA, Toussoun TA and Koch LW. 1964. Effect of crop-residue decomposition products on plant roots. *Annual Review of Phytopathology*, 2(1): 267-292.
- Royo A, Gil L and Pardos JA. 2001. Effect of water stress conditioning on morphology, physiology and field performance of *Pinus halepensis* Mill. seedlings. *New Forests*, 21(2): 127-140.
- Sain D, Arora P, Kumari M and Pal D. 2001. Morphological traits determining drought tolerance in maize (*Zea mays* L.). *Indian Journal of Agricultural Research*. 35(3): 190-193.
- Sisodia S and Siddiqui MB. 2010. Allelopathic effect by aqueous extracts of different parts of *Croton bonplandianum* Baill. on some crop and weed plants. *Journal of Agricultural Extension and Rural Development*, 2(1): 22-28.
- Somarriba E. 1992. Revisiting the past: an essay on agroforestry definition. *Agroforestry systems*, 19(3), 233-240.
- Tang C and Yu Q. 1999. Impact of chemical composition of legume residues and initial soil pH on pH change of a soil after residue incorporation. *Plant Soil*, 215: 29-38.
- Thompson BE. 1985. Seedling morphological evaluation—what you can tell by looking. In: Duryea M.L., (Ed), *Proceedings of a workshop: October 16-18, 1984*. Corvallis (OR): Oregon State University, Forest Research Laboratory, pp. 59-71.
- Thompson JR and Schultz RC. 1995. Root system morphology of *Quercus rubra* L. planting stock and 3-year field performance in Iowa. *New Forests*, 9(3), 225-236.
- Torquaebiae E. 1990. *The concept of Agroforestry*. Lect. Notes. 2nd ed. ICRAF Nairobi, Kenya.
- Tsakalimi M, Tsitsoni T, Ganatsas P and Zagas T. 2009. A comparison of root architecture and shoot morphology between naturally regenerated and container-grown seedlings of *Quercus ilex*. *Plant and soil*, 324(1-2): 103-113.
- Van Noordwijk M, Lawson G, Hairiah K and Wilson J. 2015. Root distribution of trees and crops: competition and/or complementarity. *Tree–Crop Interactions: Agroforestry in a Changing Climate*. CABI, Wallingford, UK, 221-257.
- Xu RK and Coventry DR. 2003. Soil pH changes associated with lupin and wheat plant materials incorporated in a red-brown earth soil. *Plant and Soil*, 250: 113-119.
- Xuan TD, Tsuzuki E, Hiroyuki T, Mitsuhiro M, Khanh TD and Chung IM. 2004. Evaluation on phytotoxicity of neem (*Azadirachta indica*. A. Juss) to crops and weeds. *Crop protection*, 23(4): 335-345.
- Zhang C and Fu S. 2010. Allelopathic effects of leaf litter and live roots exudates of *Eucalyptus* species on crops. *Journal of Allelopathy*, 26(1): 91-100.
- Zhang W, Ahanbieke P, Wang BJ, Xu WL, Li LH, Christie P and Li L. 2013. Root distribution and interactions in jujube tree/wheat agroforestry system. *Agroforestry systems*, 87(4): 929-939.