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ORIGINAL ARTICLE

Growth and Yield Quality Parameter of *Phyllanthus Amarus* as Affected by Moisture and Temperature Stress factors

N. Chandrawanshi, S.D. Upadhyaya, Preeti Sagar Nayak*, S. Paroha and A.B. Tiwari

Department of Plant Physiology, College of Agriculture, Jawaharlal Nehru Krishi Vishwavidalaya (JNKVV), Jabalpur 482 004 (M.P.) India

*E-Mail: **preetisagarnayak@rediffmail.com**

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This work investigates the influence of moisture and temperature stress on plant phenological parameters of *Phyllanthus amarus* plant. A pot experiment was conducted under controlled water stress environment in greenhouse during the Rabi season of 2007 and 2008 at the Experimental Farm area, JNKVV, Jabalpur. Plants were treated with different levels of water and temperature stresses. The experimental design was Completely Randomized Design (CRD) with six treatments and five replications. Moisture stress has been given on selected dates for which the sets of pot were first brought to field capacity and the water withhold till wilting. It was found that the Plant height, Root length, Number of leaves, Number of branches per plants, No. of fruits per plant, Leaf area, Dry weight gm per plant were found maximum in control condition.

Key words: Phyllanthus amarus, phenological, moisture stress, temperature stress

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Stresses associated with temperature, salinity and drought singly or in combination are likely to enhance the severity of problems to which plants will be exposed in the coming decades (Duncan 2000). Water stress is one of the most important factors reducing the yield of cultivated plants (Grzesiuk *et al*., 1999, Starck *et al*., 1995). The reason for water deficit in plants is a lack of available water in the ground, atmospheric drought, frequently occurring with high temperatures, as well as the transpiration process overbalancing water absorption (Boczek and Szlendak 1992). Water deficit is regarded as "the most important limiting factor in crop productivity in semi-arid agricultural areas" (Wilhite 1993). Water deficit conditions bring about the changes in hormone content and activity; first of all, the content of abscisinic acid (ABA) increases (Ristic *et al*., 1992), while the content of cytokines and gibberellins decreases.

Phyllanthus amarus (Family: Euphorbiaceae), although considered from the farm point of view a weed, is a valuable medicinal plant used by herbalists (Oudhia and Tripathi 2002). The roots, leaves, fruits, milky juice and whole plants are used in medicinal preparations. The bark yields a bitter principle called phyllanthin (Shakila and Rajeswari 2006). *Phyllanthus* species (family Euphorbiaceae) are generally well known for the biologically active compounds they possess (Rizk, 1987). Though the plants of the genus Phyllanthus (Euphorbiaceae) are widely distributed in most tropical and subtropical countries, and have long been used in folk medicine to treat kidney and urinary bladder disturbances, intestinal infections, diabetes, and hepatitis B. In recent years, the interest in the plants has increased considerably. Substantial progresses on their chemistal and pharmacological properties, as well as a few clinical studies of some Phyllanthus species have been made (Calixto *et al.,* 1998).

Phyllanthus amarus, a small herb locally known as "Bhumyamalaki", is the most widely distributed *Phyllanthus* species in tropical areas. Other *Phyllanthus* species viz. *P. fraternus*, *P. urinaria*, *P. virgatus*, *P. maderaspatensis,* and *P. deblis* also grow in southeast Asian countries and Africa. *P. amarus* has been used as expectorant, anti-febrile, anti-diarrhoea, a diuretic and folklore medicine for colic and kidney problems (Perry and Metzger, 1980; Unander *et al*., 1995). In India, traditionally leaves of *P. amarus* are used for the treatment of liver disorders (Nadkarni, 1976). The species is a rich source of lignans along with other phytomolecules (Calixto *et al*., 1998; Naik and Juvekar, 2003; Huang *et al*., 2003; Ishimaru *et al*., 1992). Pharmacological investigations have shown that *Phyllanthus* genus has several biological activities such as inhibition of hepatitis virus (Shin *et al*., 2005), hepatoprotective (Pramyothin *et al*., 2007), antinociceptive (Santos *et al*., 1995), antitumor (Somanabandhu *et al*., 1993), antioxidative (Bandyopadhyay, *et al*., 2000), anti-inflammatory (Kassuya, *et al*., 2005), endothelin-1 antagonistic (Hussain *et al*., 1995), human immunodeficiency virus (HIV) replication and reverse transcriptase inhibitory (Notka *et al*., 2004), immune stimulation (Ignacio *et al*., 2001), hypoglycemic, hypotensive, and diuretic (Dias *et al*., 1995). Hepatoprotective activity of phyllanthin, niranthin and hypophyllanthin against hepatitis B virus (Santos *et al*., 1995; Dias *et al*., 1995; Huang *et al*., 2003), cytotoxicity and multi-drug resistance (MDR) reversing activity of nirtetralin and niranthin (Leite *et al*., 2006) have been established. The major challenge facing water planners and managers is the availability of water. Its amount is fixed, but demand for it will continue to increase steadily into the fore-seeable future. Hence an attempt was made to study the influence of different moisture stress and temperature on phenological parameters of *Phyllanthus amarus*.

MATERIALS AND METHODS

Location and Duration: The present investigation was conducted in pots under Greenhouse at Medicinal Garden, Department of Crop and Herbal Physiology, JNKVV, Jabalpur during the Rabi season of 2007-08. Seeds of Phyllanthus plant were collected from Medicinal Garden, cleaned and stored at room temperature (approx 20-35^{0}C) in the laboratory. The seeds collected were then used for raising nursery on 1st Oct 2007. Seed were sown in the tray filled with soil and sand at 2:1 ratio.

Pot Experiment: The experiment was completely randomized design with six treatments of water regimes and five replications.

Treatment for moisture stress

MS1 – Moisture stress at 45 days after transplanting

MS2 – Moisture stress at 60 days after transplanting

MS3 – Moisture stress at 75 days after transplanting

MS4 – Restress at (45MS1) + (60MS2) + (75MS3) days after transplanting

MS5 – Restress at (60MS2) + (75 MS3) days after transplanting

MS6 – Control conditions (no moisture stress)

Treatment details for temperature.

MS1 – Temperature of 25^0C at 45 days after transplanting

MS2 – Temperature of 35^0C at 60 days after transplanting

MS3 – Temperature of 40^0C at 75 days after transplanting

MS4 – Control conditions (maintained at 30^0C throughout growth period)

After 30 days of sowing seedlings of approximately 4-5 cm height of uniform size was transplanted. The pots of size (15x15cm) and of 2.5 kg capacity where filled with soil and sand at 2:1 ratio. Sand and soil was fumigated before sowing the seeds in the respective pots. Four seedlings per pot were transplanted. Watering was done according to the requirements and weeding was done when ever required. Moisture stress has been given on selected dates for which the sets of pot were first brought to field capacity and the water withhold till wilting. Soil moisture status was recorded by using Trase Soil moisture meter daily from field capacity to till wilting. Temperature stress was given on selected dates for the period of 15 days which was maintained in controlled polyhouse.

Phenological observations recorded at 45, 60, 75 days and at harvesting: Day to emergence, Plant height (cm), number of leaf, No. of branches/plants, No. of fruits/plant, Root length, total leaf area, Dry wt g/plant by Gardner *et al*, (1985).

RESULTS

Phenological data at the time of harvest (moisture stress given at 45, 60, 75 DAP and control).

To study the relationships among growth rate to moisture stress, a set of pot experiment (5 replicate for each treatments) was laid. Moisture stress was created by soil drying and rewatering till wilting was done on selected dates i.e. 45 DAP, 60 DAP, 75 DAP and lastly harvested at 90 DAP and compared along with the control (without stress). Initially stress was given at 45 DAP, pots were brought to the field capacity and water was withhold. Soil moisture status was recorded daily till wilting by Mini Trase moisture meter. Initially at field capacity 28% moisture and at the time of wilting 7 % moisture was recorded. Then again rewatering was done and pots were maintained till harvest along with control. For second and third date of observations i.e. at 60DAP and 75 DAP, pots were kept for soil drying and rewatering same procedure was followed as 45DAP and finally data was observed at time of harvest to know the growth status compared with control (Table 1).

Phenological characters like days to germination, days to leaf initiation, days to flowering, and days to fruiting were affected by moisture stress imposed treatments. Initially the days to germinate and leaf initiation was more or less same in all the plants but days of fruiting was seen early in case of all the moisture stress. Days to

germination, days to leaf initiation started at 4 days after sowing.

Data observed at time of harvest reveals that stress had significantly reduced the plant height from that of non stress. Root length was more in case of stressed once, thus root was also significantly reduced from that of non stress. Number of leaves significantly affected the growth. Control leaves were 10.4 % more than 45 DAP stress, 14.9% more than 60 DAP and 9.49% more than 75 Days stress. Leaf area significantly reduced in stress plants. There was 5 fold decreases in leaf area at 60 Days stage followed by 3 fold at 75 DAP and finally 2 fold at 45 DAP. No. of fruits where significantly more in control 2084 reduced to 653.7 in 60 DAP stress followed by 1178.2 at 75 DAP stress and 1579 number of fruits at 45 DAP stress. Number of branches also significantly reduced from control i.e. 29.75 to 16 in 60 DAP stress and 20.2 in 45 DAP stress and 25.7 at 75 DAP stress. Dry weight was significantly affected it was reduced to 2 fold at 45 DAP stress and 60 DAP stress and 1.5 fold at time of 75 DAP stress.

Phenological data at the time of harvest (temperature stress 25^{0}C, 35^{0}C, 40^{0}C) given at 45, 60, 75 DAP and control.

To find out the thermo sensitive stage of Bhuianola the pots were exposed to different temperature on phenological attributes under controlled poly carbonated greenhouse.

A set of pot experiment was laid for giving temperature stress at 25^{0}C, 35^{0}C and 40^{0}C for the period of 15 days at different stages of growth. Initially at 45 DAP the temperature stress of 25^{0}C was given for the period of 15 days and then grown at 30^{0}C till harvest. The next set of pots at 60 DAP were given temperature of 35^{0}C for 15 days and then grown at 30^{0}C till harvest. Finally a set of pots at 75 DAP where kept at 40^{0}C for period of 15 days and then grown at 30^{0}C till harvest. The control was maintained at 30^{0}C throughout the period of growth. The phenological data reveals that day to germination, days to leaf initiation, days to flowering and days to fruiting were affected by temperature stress imposed treatments. Initially the days to germinate and leaf initiation was more or less same in all the plants but days of fruiting was seen early in case of all the temperature stress. The growth data (Table-2) reveals that the plant height was significant increased in control (30^{0}C), as compared to the other temperature T_1-25^{0}C, T_2-35^{0}C and T_3-40^{0}C which were at par to each other. There is low increase in plant height under extreme deficit possibly due to reduced cell turgor which affects cell division and expansion (Luvaha *et al*., 2008).

The maximum number of leaves was found in control (2613.2) and minimum in (1733.7). Leaf area was also significantly more in control followed by T_3, T_2 and T_1. Number of fruits was significantly more at 30^{o}C followed by 40^{o}C than 35^{o}C and lastly 25^{o}C. The results of the study indicate that water deficit decreased leaf number, leaf area, leaf water content, shoot height, shoot dry weight. Numbers of branches were more in number at 30^{o}C followed by 35^{o}C, 40^{o}C, 25^{o}C respectively. Significantly maximum biomass was noted at 30^{o}C (8.5g) followed by 35^{o}C (7.3g) than 40^{o}C (6.05g) and finally (5.7g) at 25^{o}C. Decreases in leaf number and leaf area are common occurrences in water deficit stressed plants (Luvaha *et al.,* 2008). Reduction in leaf number under extreme water deficit may have been due to reduction in leaf formation. Reduction in number of leaves can be a phenomenon by the plants to reduce transpiration surface hence water loss. Similar results have been observed in mango

rootstock seedlings, which show a decline in number of leaves due to drying or senescence of lower mature leaves (Luvaha *et al.,* 2008). Reduced leaf area decreases interception of solar radiation and consequently decreases biomass production for most crops (Masinde *et al*., 2005). In root length there was significant increase in control 30°C than other temperatures. Root length was 30 cm in control followed by 15 cm T_3 (40°C), 14cm in T_2 and 13.5cm in T_1.

The growth data reveals that stress significantly reduced the plant height. The leaf area also significantly reduced during all type of stress. The leaf area is a measure of vegetative growth of plant and the assimilatory surface area on which the production of photosynthates and assimilatory rates depend and it also serves as the primary value for the computation of other physiological determinates of productivity (Watson, 1947). The specific leaf area is more sensitive to environmental changes. Leaf area is most important physiological indices to estimate the final productivity of crop and is also known as photosynthetic potential (Kvet and Husak, 1978). Over all stress results into growth reduction which is due to decreased photosynthetic efficiency, decrease uptake of moisture and nutrient, and reduction in the activity of different enzyme. An Important aspect of severe desiccation is that water content in the cells become so scarce that enzyme activities are inhibited. (Vertucci and Leapold, 1987).

Table 1: Comparison of phenological data at the time of harvest (moisture stress given at 45, 60, 75 DAP and control).

Treatments	Plant height (cm)	Root length (cm)	No. of leaves	Leaf area	No. of fruit per plant	No. of Branches per plant	Dry wt gm per plant
Control	22.0	15.2	2251.8	1755.2	2083	28.98	8.50
M_1 -45 DAP	17.6	13.3	2016.1	583.9	1575	20.15	4.70
M_2 -60 DAP	17.4	13.3	757.45	302.2	652	15.62	4.30
M_3 -75 DAP	20.0	14.1	1302.7	559.9	1175	25.40	6.30
CD @ 5%	1.55	1.73	1.94	8.44	5.23	1.57	0.27
SE m±	0.67	0.75	0.84	3.66	2.27	0.68	0.11

Table 2: Comparison of phenological data at the time of harvest (temperature stress 25°C, 35°C, 40°C) given at 45, 60, 75 DAP and control (maintained at 30°C).

Treatments	Plant height (cm)	Root length (cm)	No. of leaves	Leaf area	No. of fruit/ plant	No. of Branches per plant	Dry wt gm/ plant
Control	32.3	29.9	2613.2	1329.2	2083	47.6	8.50
T_1 -45DAP	22.6	13.5	1733.7	181.5	655	33.9	5.60
T_2 -60DAP	21.8	13.6	1807.7	578.4	1177	34.6	7.30
T_3 -75DAP	22.2	14.6	1923	1286.4	1577	25.2	6.00
CD @ 5%	2.18	1.55	6.9	22.2	2.03	1.78	0.05
SE m±	0.94	0.67	3.2	9.6	0.88	0.77	0.02

CONCLUSION

The various treatment combinations of moisture and temperature stress exhibited a significantly variability in phenological parameters. The growth data reveals that stress significantly reduced the plant height. The leaf area also significantly reduced during all type of stress thereby significant reduction in the dry weight was also seen in stress.

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