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Studies on Germination behaviour and Physiological response of *Terminalia arjuna (Roxd.)* towards fly-ash during its initial stage

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ABSTRACT

An experiment was conducted at the nursery of Guru Ghasidas Vishawavidyalaya, Bilaspur (Chhattisgarh). Studied on germination behavior, physiological response and initial seedling growth performance of *Terminalia arjuna (Roxd.)* on growing media of different concentration of fly ash and soil during March to August 2022. Growing media was prepared by admixing fly ash to nursery soil (S) at five concentrations (T0, control), T1 20% (fly ash+soil), T2, 40% (fly ash+soil), T3, 60% (fly ash+soil) T4, 80% (fly ash +soil), T5, 100% (fly ash), The experimental design was CRD with six treatments and three replications. Freshly collected seeds were washed with cold water and sown at 2.0-3.0 cm depth in germination trays filled with media of different treatments. Significant (P<0.05) variation in germination period, rate, with respect to flyash concentration in media was observed (n=100). Maximum rate (60%) was found in media having 20% FA after 30 days of sowing. Significant (P<0.05) in seedling survival rate, plant height, diameter growth, leaf number, nodules per plant and seedling quality index were observed. The survival rate (91.11%), plant height (40.33 cm), collar diameter (4.23 mm), root length (40.11cm) and seedling quality index (7.92) were at maximum in growing media having 20% fly ash (T1). It is concluded form the present investigation that fly ash can be admixed 20% (w/w) in forest nurseries for improving germination and promoting seedling growth and quality improvement of *Terminalia arjuna (Roxd*)

Key word: Collar diameter, Fly ash, "Germination period, Growing Media, Plant height

Introduction

Natural resources are materials from the Earth that are used to support life and meet people's needs. Any natural substance that humans use can be considered a natural resource. Like Oil, coal, natural gas, metals, stone and sand are natural resources. Natural resources such as coal, natural gas and oil provide heat, light and power. Carbon stored in fossil fuels are used for generating needed energy (electricity) for running combustion engines in industry and coal fired thermal power plants holds a principal share of this (Canning *et al.*, 2017). Fly ash (FA) is one of the major noxious wastes generated from coal based thermal power plants (Behera *et al.*, 2018). It is ultrafine in nature and contains a number of toxic metals such as arsenic (As), barium (Ba), mercury(Hg), chromium (Cr), nickel (Ni), vanadium (V), lead (Pb), zinc (Zn), etc. depending upon the source of coal (Dwivedi and Jain 2014). Extracting,

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processing and using natural resources can cause environmental problems such as: air, land and water pollution; disruption or destruction of ecosystems; and a decrease in biodiversity. Proper disposal and management of such a huge quantity of fly ash possessing potential threats of air and water soil pollution is a great challenge (Rawat et al., 2018). Fly ash is a repository of nutrients which can benefit plant growth and increase biomass production. Fly ash has been reported to contain low amounts of C and N, medium amounts of available K and high concentration of available P (Sharma and Kalra, 2006). Fly ash is being used in manufacturing cement, concrete, bricks, wood substitute products, in road construction, wasteland reclamation; filling of underground mine spoils (Kaur and Goyal, 2015). Fly ash contains almost all the plant nutrients except nitrogen, phosphorous and humus, which can be supplemented by organic matter (Sharma and Karla, 2006). Hence there is a scope for utilization in agriculture and forestry sector. The addition of biomass to waste land @ 10-15 tonnes/ha/year helps in the improvement of soil fertility. Plant species achieve a chemical equilibrium with respect to a particular medium or pot mixture exposure (Mountouris et al., 2002). Many research findings infers to the positive growth and nutritional efficiency of fly ash. Crop plants of the families Brassicaceae, Chenopodiaceae, Fabiaceae, Leguminoceae and Poaceae are most tolerant to fly ash toxicity (Cheung et al., 2000). One of the most potential areas of utilization is in forestry sector where it can be consumed either in nursery or for tree plantation activities. This will help in locking the toxic heavy metals in the wood biomass for longer period of time. Fly ash as planting material in forest nursery is not a new concept. Goyal et al. (2002) reported its use in nursery as growing media but commercial use is scanty or absent. Present study was made to know its impact on seed germination and growth of seedlings at early stages. Terminalia Arjuna (Combertacae). It is an important species for social and agroforestry plantation. Tolerance in infertile, acid, alkaline or seasonal waterlogged soil, mine overburden soil makes it very useful species for reclamation purpose. Studies on regeneration and development of established tree species on land fill site is also necessary for understanding the ecological balance in man-made ecosystem and restoration of different biogeochemical cycles. Information on limiting dose of fly ash for seed germination will be helpful for self-sustenance of a habitat. Perusal of literature reveals that such kind of information is scanty. Keeping this in view the present study was carried to know the impact of fly ash (at different concentrations) on seed germination and initial seedling growth attributes .

Materials and Methods

The experiment was conducted in Department of Forestry, Wildlife & Environmental Sciences Guru Ghasidas Vishawavidyalaya, Bilaspur (Chhattisgarh) situated at 22.12' N latitude and 82° 13' E longitude with altitude 264 m amsl. Fly ash was collected from NTPC Sipat.

Growing media preparation and analysis for physicochemical properties

Growing media was prepared by mixing fly ash to forest soil at concentrations 20%, 40%, 60%, 80% and 100% W/W. There were five treatments (T1-20% FA+S, T2-40% FA+S, T3- 60% FA+S, T4-80% FA+S, T5-100% FA). The growing media was analyzed for physical and chemical properties. Bulk density (BD) and water holding capacity (WHC) was determined by using the protocol given by Piper (1966). pH and electrical conductivity (EC) were measured following protocol given by Jackson (1967), organic carbon (OC) was estimated as per Walkley and Black (1934). Available nitrogen, phosphorus and ammonium acetate extractable potassium were estimated as per the procedure given by Subbiah and Asija (1956), Olsen et al., (1954) and Merwin and Peech (1951) respectively. The physical and chemical properties of growing media are given in Table 1.

Seed treatment and sowing

Germination study

Freshly collected seeds were given hot water treatment prior to sowing. Seeds were soaked in boiled water at 70 °C for 20 minutes followed by cold water treatment (the volume of water was five times the volume of seed) for 24 hours (Tadros *et al.*, 2011). Eighteen germination trays having dimension 90 cm (L) X 45 cm (B) X 15 cm (H) were filled with above six mentioned substrates to the brim leaving 3.0 cm. Hundred seeds per replication (totaling 300 seeds per treatment)were sown at a depth of 0.5-1.0 cm in germination trays. Manual watering to the field saturation level once a day during morning hours was made. Observations pertaining to germination parameters were recorded daily up to 30 days after sowing. Germination period was determined by observing the day taken for first germination (DTFG) to 30th day when about 40-60% seeds have germinated. Based on the number of seeds germinated the following parameters were calculated as per the standards given by Czabatore (1962) and AOSA (1983).

Germination(%) =
$$\frac{\text{Number of seeds germination}}{\text{Number of seeds sown}} \times 100\%$$

Seedling growth study

After completion of germination study, seedlings were transplanted into poly pots containing substrates of above mentioned treatment combinations. Growth parameters such as shoot length, collar diameter and number of leaves were assessed monthly after 30 days of transplanting for 3 months. Total shoot length was measured by using ruler (taken from the apical bud of the plant to the base of the shoot) and stem diameter by using electronic digital caliper for recording the quantitative parameters pertaining to root growth, the entire seedling was dipped in a bucket of water at 90 days to remove adhering soil from it. It was then carefully washed so that no damage was made to root system. Length of roots (starting from collar region to the end point) and number of leaves were recorded. Thoroughly washed seedlings (without damage to root and shoot) were dried under sun for 30 minutes. The shoot and root portions were separated for each seedlings. The shoot was cut from the collar portion. Fresh weight of the shoot and root were recorded by a digital balance. Then the root and shoot sample were put in paper bags separately and were oven dried at 80°C for 48 hours. The dry weight were recorded and expressed in gram. Growth observation was based on 45 numbers of randomly selected plants from each treatment. The seedling quality index (SQI) was calculated by using the formula as described by Dickson *et al.* (1960).

The experiment was completely randomized design with three replications. The collected data were analysed with a general linear model using SPSS software version 20 for windows operating system. Means were analysed according to the Duncan Multiple Range Test (DMRT) at P < 0.05.

Results and Discussion

There exists a vast scope for utilizing fly ash in forestry sector particularly as a growing media for quality seedling production and rhizosphere improvement material at difficult sites prior to plantation. The matrix of application depends upon the elemental composition of fly ash to be used, tolerance limit of plant species selected and physiochemical property of plantation site soil or growing media in which fly ash need to be added.

Effect of substrate on seed germination

The biochemical process of seed germination is affected by a number of intrinsic and environmental factors. Effective pre sowing treatment of seed reduce unfavorable endogen window. The exogenous variable mainly the substrate and climatic condition can be managed successfully to achieve maximum germination rate. Substrate property especially pH and water retention capacity have a marked impact on germination. pH affects germination either by

Table 1. Physico-chemical properties of fly ash, forest soil and fly ash substratum

Property	Nursery soil	Fly ash (FA)	FA substrate (Nursery soil + FA %)			
			20% FA	40 % FA	60 % FA	80 % FA
pН	8.2	8.4	7.01	7.18	7.38	7.58
EC (dS m-1)	1.47	0.542	0.63	0.93	1.11	1.28
N (kg ha-1)	0.19	0.001	97.03	80.73	71.22	59.66
P (kg ha-1)	90.21	5.98	50.23	41.11	31.23	20.11
K (kg ha-1)	52.33	126.66	480.23	502.33	537.77	611.23
OC (%)	255.45	0.003	0.52	0.46	0.33	0.22
BD (g cm-3))	0.470	0.58	1.28	1.18	0.93	0.72
Pore space (%)	21.12	51.22	39.39	35.23	31.23	25.21
Water holding capacity (%)	30.23	55.21	41.11	46.42	47.33	53.25

FA- fly ash, EC- Electrical conductivity, NPK-Available Nitrogen, Phosphorous and Potash, OC-Organic carbon, BD-Bulk density, WHC-Water holding capacity.

increasing the osmotic pressure of the media to a plant that will retard or prevent the intake of water or by causing toxicity to the embryo (Rashid, 2004).

During this research study, it was revealed that fly ash have a significant (P < 0.05) impact on seed germination parameters like germination period, rate, however did not have any impact on the number of days taken for first germination (NDFG) (Table 2). The highest seed germination percent (60%) was observed in growing media having 20% fly ash (T1) and was significantly (P>0.05) higher than other treatments (Table 2). Germination responses in relation to seed size and pre-treatments (Agboola et al., 1993; Murali, 1997; Schiotz et al., 2006; Shivanna et al., 2007; Likoswe et al., 2008; Amri, 2010). The germination rate of 53.33% in T2, 48.66% in T3, 30.44% in (T4) and 20% T5 respectively are statistically as par (P>0.05) with each other. Minimum germination rate (20%) was observed in substrate having 100% fly ash (T5, Table 2). The increased germination rate (60%) in 20% fly ash the reduction in germination rate beyond 60% fly ash addition (w/w) in media was due to enhanced pH and elemental toxicity. Higher pH and metals like Cu²⁺, Zn²⁺ at higher EC are reported toxic to embryo and reduces biological activity during germination process (Gupta et al., 2000). There existed negative relationship between fly ash rate with germination percentage, but it was positive with DTFG, germination period and (Table 2).

Effect of substrate on seedling growth

After 90 days of transplanting significant difference (P>0.05) in survival rate, plant height, diameter growth, mean root length and seedling quality index of Terminalia arjuna was observed. But no significant difference (P>0.05) in number of leaves was recorded (Table 3). Highest seedling survival rate (91.57%) was found in substrate containing 20% fly ash (T2) which and was statistically (P>0.05) different from treatment T2 (81.16%), T3 (61.23), T4 (51.11) and T5 (40.23). The maximum survival rate (91.57%) of seedlings in substrate containing 20% fly ash (T2) was due the improved aeration and water retention capacity of substrate. The survival rate decreased linearly with increased concentration of fly ash up to minimum 40.23% in growth media having 100% (Table 2). The maximum height (40.33 cm) was found in treatment having 20% fly ash and it was statistically at par with Treatment T2 (33.4 cm), T5 (29.86 cm) and T4 (26.66). (Tripathi and

 Table 2. Effects of substrates on germination of Terminalia arjuna seeds at 30 days after sowing

Parameter Treatment	DTFG	Germination Percentage	Survival Percentage
T1	2	60%	91.57%
T2	2	40%	81.16%
T3	4	46%	61.23%
T4	3	60%	51.11%
T5	5	20%	40.23%

Treatments T1=(20% FA+S), T2=(40% FA+S), T3=(60% FA+S), T4=(80% FA+S), T5=(100% FA), FA- fly ash, S-Forest Soil, DTFG- Days taken for first germination, Mean values followed by same letter are statistically indifferent.

Khan, 1990; Ke and Werger Seiwa, 1999; 1998, 2000). The diameter growth was maximum in treatment T1 (4.23 cm) and statistically at par with T2 (4.21 cm), T3 (4.01 cm), T4 (4.07) and T5 (4.15). Longer mean root length was recorded. In treatment T1 (34.11 cm) and statistically at par with Treatment T5 (33.03 cm), T4 (32.13 cm) T2 (25.53) and T3 (21.2) numbers leaves were found in growing media having 20% FA T1 (24.66) and statistically at par with T5 (20.22), T4 (18.66), T3 (17.33) and T2 (15.12). A similar trend in growth of seedlings with respect to fly ash concentration was also reported by Gupta et al. (2000) and Pandey et al. (1996). The vigour in seedling height, diameter and root growth of this species at 20% fly ash was due to light alkali pH, improvement in availability of nutrients in ionic form at rhizosphere solum and improved nitrogen fixation rate (Table 2), and reduced or no attack of nursery insect and pest. Goyal et al. (2002) observed 10% increase in the growth of Eucalyptus tereticornis, Acacia auriculiformis and Casuarina equisetifolia during early 6 months, grown in fly ash amended soils [ESP FA@18-24% (v/v)]. Further the plants grown in 20% fly ash were observed to be very healthy. (DICKSON et al. 1960). Better seedling quality index in T1 (7.92) was obviously due to the improved availability of micronutrients that supported higher biomass production and shoot: root ratio (Gupta et al., 2000).

Conclusion

The present study of pre-sowing treatments of seeds would prove itself potential. Among the treatments applied in the experiment for Terminalia arjuna, Seeds soaking in cold water for 48 hours was found more effective in respect to faster Germination,

Parameter Treatment	Plant height (cm)	Collar diameter (mm)	No. of Leaves	Root length (cm)	SQI
T1	40.33	4.23	24.66	34.11	7.92
T2	33.41	4.21	15	25.53	7.75
Т3	27.33	4.01	17.33	21.2	6.49
T4	26.66	4.07	18.66	32	5.12
T5	29.86	4.15	20	33.03	5.02

Table 3. Effects of substrate on growth and quality of Terminalia arjuna seedlings at 180 DAT

Treatments T1= (20% FA+S), T2= (40% FA+S), T3= (60% FA+S), T4=(80% FA+S), T5=(100% FA), FA- fly ash, SQI-Seedling Quality Index, Mean values followed by same letter are statistically indifferent.

higher germination percentage, seedling growth, and comparison to control and other treatments soaking in cold-water for a Terminalia arjuna particular period of time is easily applicable and cost effective, the treatment may be recommended for large-Effects of Pre-sowing Treatments. Terminalia *arjuna* is a most important species that tolerance in fertile, acid, alkali and mine overburden soil well accomplished for improving waterlogged and reclamation of mine area. The results of present investigation recommends fly ash should be admixed at 20% (w/w) level in nursery beds for early sprouting and improving germination percentage. However, it should be admixed at 20% in potting mixture for production of healthy and quality planting material. Further study is necessary to quantify the economic benefit or net profit gain from utilizing fly ash in forest nursery.

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