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# Phytotoxic effect of synthetic dye effluents on seed germination and early growth of red amaranth

ABSTRACT

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#### ARTICLE INFORMATION

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Among the industrial chemical wastes, the effluents of loom-dyeing industry raise great concern in recent times because of their diverse environmental hazards. A laboratory experiment was conducted to determine the phytotoxic effect of different loom-dye effluents on seed germination and early seedling growth of red amaranth (Amaranthus tricolor L.) in the Department of Agricultural Chemistry of Bangladesh Agricultural University, Mymensingh, during May to August, 2017. Results revealed that the physico-chemical parameters of loom-dye effluent such as pH, electrical conductivity (EC), total dissolved solids (TDS), sulphate, phosphate, sodium, potassium, iron, manganese and lead concentrations were relatively higher than the suggested range. On the other hand, the concentrations of calcium, zinc, copper and cadmium were relatively lower. Germination experiment of red amaranth were done in sterilized petridishes containing 0, 5, 10, 25, 50, 75 and 100% concentrations of three different types of untreated loom-dyeing effluents following completely randomized design. Three replications were used for each treatment. The germination energy and capacity, growth parameters like root and shoot length, relative toxicity, seedling vigour and phytotoxicity on seed germination of red amaranth in response to different loom-dyeing effluents at various concentrations were also calculated. There was a gradual lessening in the percentage of seed germination and seedling growth with increased concentration of effluents. Relative toxicity and phytotoxicity was extreme at 100% effluent concentration. Different growth parameters such as root and shoot lengths of seedlings was minimum at 100% and maximum at 5% dye effluent concentration. Overall results indicate that pink dye effluent was less toxic to the germination and early growth of red amaranth than violet and black dyes. For most of the growth parameters of red amaranth, the order of phytotoxicity among the loom-dye effluent was black > violet > pink.

**Keywords:** Germination energy, phytotoxicity, dye effluent, relative toxicity, vigour index

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#### 1 Introduction

Environmental contamination has been changed over into a clarification focal point of distress for all the nations worldwide, as not only the developing countries but developed nations as well are affected by and experience the ill effects of it. Production of waste effluent is an integral part of industrial activities which is released in the environment, coupled with the development of industrialization has brought about massive and destructive changes in our biological communities. The industrial effluents are a complex mixture of numerous waste substances such as organochlorine-based pesticides, heavy metals, pigments and dyes (Rohit and Ponmurugan, 2013); harmful gases, and several organic and inorganic mixes (Balaji et al., 2012) which make water toxic (Robinson et al., 2001) affecting the photosynthetic organisms when exposed and consequently impact negatively on the food chain (Coulibaly et al., 2003). The degree of contamination has been so intense near the industrial areas. The final toxic effect of the dye effluent may be synergistic, additive or antagonistic as a function of the different effluent components that make up the effluent. The release of material and colour effluent harms the nature of soil and water bodies. It is reported that to dye 1 kg of cotton goods with reactive dyes requires an average of 70-150 L water, 0.6 kg NaCl and 40 g reactive dyes, alkalis and others pretreatment and dyeing auxiliaries (Allègre et al., 2006). Accumulation of excessive salts makes the soils saline, while presences of excessive colour in effluent pollute the water bodies and prevent the penetration of light, which in turn impedes with the photosynthetic activities of aquatic flora (Parameswari and Udayasooriyan, 2013).

Bangladesh is an agricultural country, where industrialization is occurring in a bit by bit expanding stage. These industries are the major sources of effluents and eventually results in high wastewater generation. Colouring industrial facility of 10 tons production capacity generates nearly 100 to 150 m<sup>3</sup> of wastewaters per hour (Haque, 2008). There are some reports that the farmers irrigate their gardens with water obtained from rivers and streams, which receive effluents from different dyeing industries. They regard the effluent as a resource that can be connected for beneficial utilization since it contains nutrients that have the potential for use in farming. This practice has been on for a very long time unabated. They found some phytotoxic effects in case of mixing the larger amount of dye effluent with the irrigation water. The use of industrial effluents for irrigation has emerged in the recent past as an important way of utilizing wastewater, taking the advantage of the presence of considerable quantities of N, P, K and Ca along with other essential nutrients (Niroula, 2006). Though it is considered as a resource it creates

many problems. Rodosevich et al. (1997) considered seed germination a critical step as it ensures reproduction and controls the progression of plant populace and in addition likely harvest efficiency. Seed germination and early plant development bioassays are the most widely recognized systems used to assess phytotoxicity (Kapanen and Itawara, 2001). Such kinds of works have performed scattered way in many countries of the world. While working with Cicer arietinum, Dayama (1987) reported that even highly diluted industrial effluent (5% effluent) adversely reduced the seed germination. Furthermore, it also causes phytotoxicity results from the intoxication of living tissues by substances accumulated from the growth medium (Chang et al., 1992). The adverse effects of effluents on plants depend on the type of species, types and concentrations of toxic materials in the effluent (Hassan et al., 2013). This necessitates a detailed scientific study before any specific waste can be used for irrigation for the particular crop and environmental conditions. Despite the importance of seed germination treated with different levels of loom-dying effluents the mechanism of phytotoxicity or beneficiary influence in seeds is relatively poorly understood. This research was therefore aimed at studying the phytotoxic effect of loom-dyeing effluent collected from various power loom-dyeing industries on the germination of red amaranth as a preliminary step before field trials and to advise the appropriate government agencies to monitor the activities of these farmers effectively and discourage them from using effluents from industries as irrigational means.

#### 2 Materials and Methods

#### 2.1 Dye effluent sampling

The effluent samples were collected from loomdyeing industries located at Belkuchi upazila of Sirajganj district in 2 L plastic containers in May 2017. After collection, the effluent was immediately transported to the laboratory for physico-chemical analysis.

#### 2.2 Analysis of dye effluent

The collected dye effluent sample was analyzed for their physico-chemical properties in the laboratory of Department of Agricultural Chemistry, Bangladesh Agricultural University, Mymensingh. Physico-chemical parameters such as pH, electrical conductivity, total dissolved solids, sulphate, phosphate, calcium, sodium, potassium, iron, zinc, manganese, copper, lead and cadmium were analyzed as per the standard methods (APHA, 2012). For the determination of metal concentration, the effluent sample (100 mL) was digested with di-acid mixture (HNO<sub>3</sub>: HClO<sub>4</sub> = 2:1). Then, the digest was filtered through filter paper (Whatman no. 42) and the filtrate volume was made up to 100 mL with distilled water.

#### 2.3 Seed germination assay

The germination experiment was conducted at postgraduate research laboratory, Department of Agricultural Chemistry, Bangladesh Agricultural University, Mymensingh during the period of August to October, 2017. Germination experiments were carried out in sterilized petridishes lined with double layer of filter paper. The petri dishes were incubated at  $26\pm2$  °C for germination.

#### 2.3.1 Selection of test crop

Red amaranth var. BARI Lalshak-1 (*Amaranthus tricolor* L.) was chosen for both germination and early seedling growth obtained from Bangladesh Agricultural Development Corporation (BADC), Mymensingh.

#### 2.3.2 Preparation of different effluent concentrations and germination studies

The healthy and uniform sized seeds of red amaranth were selected and surface was sterilized with 0.1% HgCl<sub>2</sub> and thoroughly washed with distilled water to avoid surface contamination. Twenty sterilized seeds of red amaranth were taken in each petridish. Seven treatment levels viz., control, 5, 10, 25, 50, 75 and 100% of untreated loom-dyeing effluent were used following Completely Randomized Design (CRD) with three replications. Experimental treatments comprised of control: 0 mL effluent + 100 mL distilled water (DW); 5%: 5 mL effluent + 95 mL DW; 10%: 10 mL effluent + 90 ml DW; 25%: 25 mL effluent + 75 mL DW; 50%: 50 mL effluent + 50 mL DW; 75%: 75 mL effluent + 25 mL DW; 100%: 100 mL effluent + 0 mL DW. Three replicates were maintained for each treatment including control. The observed germination percentage was recorded after 3 and 7 d. The growth parameters like shoot and root length were recorded after 7 d. After 7 d, seedlings were harvested; fresh weights were recorded. For dry weight, seedlings were incubated at 60 °C for 24 h.

#### 2.3.3 Data collection

**Germination percentage** The germination percentage was calculated using the following formula (Raun et al., 2002).

$$G = \frac{S_g}{S_T} \times 100 \tag{1}$$

where, G = gerimnaiton (%),  $S_g$  = number of seed germinated, and  $S_T$  = total number of seed set for the test.

**Germination energy** Percentage of germinated seed at 3 DAS (Bam et al., 2006).

**Germination capacity** Percentage of germinated seed at 7 DAS (Bam et al., 2006).

**Germination speed** Speed of germination was measured by using following formula (Krishnaswamy and Seshu, 1990).

$$G_s = \frac{S_{g72}}{S_{g168}} \times 100 \tag{2}$$

where,  $G_s$  = speed of germination,  $S_{g72}$  = number of seed germinated at 72<sup>th</sup> h, and  $S_{g168}$  = number of seed germinated at 168<sup>th</sup> h.

**Relative toxicity** Relative toxicity (%  $R_T$ ) of dye effluent on the seed germination of red amaranth over control was calculated by using the following formula of Chapagain (1991).

$$R_T = \frac{x - y}{x} \times 100 \tag{3}$$

where,  $R_T$  = relative toxicity (%), x = germination percentage in control at particular hour of incubation, and y = germination percentage in the presence of effluent at the same hour of incubation

**Phytotoxicity** The phytotoxicity was calculated using the formula of Chou and Lin (1976).

Phytotoxicity (%) = 
$$\frac{R_c - R_t}{R_c} \times 100$$
 (4)

 $R_c$  = radical length of control and  $R_t$  = radical length of test.

**Vigour index** Vigour index of the seedlings was calculated using the formula proposed by Abdul-baki and Anderson (1973).

Vigour index = 
$$G \times L$$
 (5)

where, G = germination (%) and L = seedling length.

**Fresh weight** Ten seedlings were collected from each treatment and their fresh weights were measured. Then it was calculated as weight per plant.

**Dry weight** The same seedlings used for fresh weight were kept in hot air oven at 60 °C for 24 h. Then, the seedlings were taken from the oven and kept in desiccators for some time. Then their dry weights were taken.

#### 2.4 Statistical analyses

The physico-chemical analyses of elements present in dye effluent was repeated three times and data recorded each time were assembled for statistical analysis. Collected data of seed germination assay were also analyzed to determine the significance of variance (P<0.05). For comparison of treatment means, standard errors were computed using SPSS (20.0) and Microsoft Excel programme.

#### 3 Results and Discussion

### 3.1 Physico-chemical properties of dye effluent

The physico-chemical parameters of dyeing industry effluent have been reported in Table 1. The color of the collected loom-dye effluent was pink, violet and black. All the three effluents had slight to very bad and pungent odour The pH range of the loom-dyeing effluent was from 12.74 to 12.89 which was basic in nature and was beyond the permissible limit of pH (6.0–8.5) for irrigation water prescribed by Ayers and Westcot (1985). The basic pH of the effluent might be resulted from the use of caustic soda during the dyeing process. Shuchismita and I (2015) observed the pH of textile effluent ranged between 3.9 and 14.0. Higher EC value reflected the higher amount of salt concentration which can affect the irrigation water quality related to salinity hazard.

EC found in the effluent (782–4380  $\mu$ S cm<sup>-1</sup>) was greater than that of the permissible limit (0–1200  $\mu$ S  $cm^{-1}$ ) of environment conservation rules (ECR, 1997) except the pink dye effluent. This might be due to the continuous discharge of the chemicals and salts used along with dyes in the industries. Nahar et al. (2018) found the average EC value in dye effluents as 1557.44  $\mu$ S cm<sup>-1</sup>. Akbar et al. (2014) reported average EC value of Narsingdi textile industries effluent as 25781  $\mu$ S cm<sup>-1</sup>, which was above the accepted limits. The effluent recorded total dissolved solids ranging between 1320 and 5382 mg  $L^{-1}$  beyond the standard value. According to environment conservation rules (ECR, 1997) standard presence limit of TDS in textile effluent is 2100 mg  $L^{-1}$ . FAO standard range of TDS value for irrigation practices is from 450 to 2000 mg  $L^{-1}$  (Ayers and Westcot, 1985). Chhikara et al. (2013) found TDS in the effluent of different textile industries in India as 3586.67 mg  $L^{-1}$ . In the present study, sulphate level was recorded almost same value *i.e.*, 69.16 and 69.67 mg  $L^{-1}$ . The range of calcium  $(28.06-46.10 \text{ mg L}^{-1})$  and magnesium (2.48-6.42 mg) $L^{-1}$ ) was lower than the permissible limit described by Ayers and Westcot (1985). Islam et al. (2015) found Ca and Mg concentration in industrial effluent samples ranging between 14.0–61.0 mg  $L^{-1}$  and 10.0–34.0 mg  $L^{-1}$ . But the concentration of potassium (9.52– 19.39 mg  $L^{-1}$ ) and sodium (70.95–109.22 me  $L^{-1}$ ) was beyond the limit for irrigation.

Ayers and Westcot (1985) indicated that irrigation water generally containing less than 40 me  $L^{-1}$  Na is suitable for crops and soils. Islam et al. (2015) also found K content as  $34 \text{ mg L}^{-1}$  in the industrial effluent. Among the metals, iron content in the effluents ranged from 3.78 to 13.37 mg  $L^{-1}$  (Table 1) which was higher than the permissible limit described by Ayers and Westcot (1985). Sivakumar et al. (2011) reported 37.0 mg  $L^{-1}$  Fe in the effluent of textile dyeing and bleaching industries at Karur, Tamil Nadu. Manganese and lead content in the effluents was also beyond the permissible limit. The concentration of Pb exceeded the value (0.289 mg  $L^{-1}$ ) reported by Akter et al. (2010), but lower than the value (19.0 mg  $L^{-1}$ ) reported by Sivakumar et al. (2011). Among three effluents, copper content of violet dye effluent exceed the recommended limit. The concentration of Cu in dyeing and printing industrial effluents of Belkuchi, Sirajganj was reported as 0.749 mg  $L^{-1}$  by Akter et al. (2010). Nahar et al. (2018) found the average Pb concentrations in 25 dyeing effluent samples in Narsingdi, Bangladesh as  $0.170\pm0.149$  mg L<sup>-1</sup>. But the concentrations of zinc and cadmium did not exceed the limit. The order of the concentration of different chemical parameters of three effluents for electrical conductivity, total dissolved solids, sodium, potassium and phosphate was pink < violet < black dye effluent; whereas the calcium, iron, zinc and copper follow the order pink < black < violet, But lead and cadmium, sulphate and manganese did not follow the previous trend showing the highest value in the pink dye effluent.

#### 3.2 Seed germination assay

#### 3.2.1 Germination Energy

Seed germination and early seedling growth are vital for continuation of plant life and they are extremely vulnerable to environmental stress. Since germination is the first physiological process, several growth parameters such as germination and ultimately growth and yield of the crops are taken as criteria to assess the degree of pollution (Mishra and Pandey, 2002). The maximum seed germination percentage of red amaranth at 3 DAS was recorded at control. But with dye effluent maximum germination energy (93.3%) was found with 5% black dye effluent (Fig. 1a). At the same time, the minimum germination was recorded in 100% concentration of the three dye effluents. For pink effluent, germination energy increased up to 25% effluent concentration and then slowly decreased to 48.3% at 100% effluent concentration. But another two effluents (violet and black) gave the decreasing rate of germination energy with increasing the effluent concentrations. In stress conditions, the energy forming molecules may be disturbed

Parameters	Unit	Effluent-1	Effluent-2	Effluent-3	Acceptable limit <sup>†</sup>
Colour		Pink	Violet	Black	_
Odour		Slightly pungent	Pungent	Very pungent	_
Temperature	°C	36.5	37	37	_
pH	_	12.86	12.74	12.89	6.0-8.5
ĒC	$\mu { m S}{ m cm}^{-1}$	782	2947	4380	_
TDS	$mg L^{-1}$	1320	3584	5382	450-2000
Calcium	$mg L^{-1}$	28.06	46.1	30.06	800
Magnesium	$mg L^{-1}$	6.42	5.5	2.48	60
Potassium	${ m mg}~{ m L}^{-1}$	9.52	9.58	19.39	2
Sodium	$me L^{-1}$	70.95	88.06	109.22	40
Sulphate	$ m mg~L^{-1}$	69.67	69.16	68.25	20
Phosphate	${ m mg}~{ m L}^{-1}$	17.85	21.45	38.28	2
Iron	${ m mg}~{ m L}^{-1}$	3.78	13.37	8.18	5
Zinc	${ m mg}~{ m L}^{-1}$	0.15	0.41	0.38	2
Manganese	${ m mg}~{ m L}^{-1}$	0.43	1.72	0.18	0.2
Copper	$mg L^{-1}$	0.07	0.49	0.17	0.2
Lead	$mg L^{-1}$	0.63	1.14	1.12	_
Cadmium	$mg L^{-1}$	0.013	0.009	0.007	0.01

Table 1. Physico-chemical properties of used loom-dye effluent

<sup>+</sup> Ayers and Westcot (1985); EC= electrical conductivity, TDS = totoal dissolved solids

Dye effluent	Concentration of dye effluent (%)								
	0	5	10	25	50	75	100		
Germination speed (%)									
Pink	$100.0 {\pm} 0.0$	93.2±4.4	91.2±4.6	92.6±4.9	92.4±3.8	90.4±5.2	62.3±7.8		
Violet	$100.0 {\pm} 2.9$	$94.7 \pm 3.0$	$93.0{\pm}3.5$	$91.2 \pm 3.5$	89.0±3.3	$89.7 {\pm} 4.2$	$0.0{\pm}0.0$		
Black	$98.2 {\pm} 1.7$	$94.9{\pm}2.9$	$94.8{\pm}3.0$	92.9±3.8	$70.0 \pm 15.2$	$0.0{\pm}0.0$	$0.0{\pm}0.0$		
Root length (cm)									
Pink	3.6±0.15	3.7±0.15	$3.4{\pm}0.15$	2.9±0.20	$2.4{\pm}0.09$	$1.9 {\pm} 0.06$	1.2±0.12		
Violet	$3.6 {\pm} 0.15$	$3.5 {\pm} 0.12$	$3.0 {\pm} 0.15$	$2.5 {\pm} 0.12$	$1.9 {\pm} 0.44$	$0.5 {\pm} 0.35$	$0.0{\pm}0.0$		
Black	$3.6{\pm}0.15$	$3.4{\pm}0.18$	$3.1 {\pm} 0.24$	$2.5 {\pm} 0.32$	$1.8{\pm}0.49$	$0.0{\pm}0.0$	$0.0{\pm}0.0$		
Shoot length	(cm)								
Pink	4.7±0.23	$4.9 {\pm} 0.14$	$4.6 {\pm} 0.06$	$4.2 {\pm} 0.15$	$3.9 {\pm} 0.14$	3.2±0.20	2.5±0.19		
Violet	$4.7 {\pm} 0.23$	$4.6 {\pm} 0.29$	$4.3 {\pm} 0.12$	$3.9 {\pm} 0.21$	$2.9 {\pm} 0.25$	$2.7 {\pm} 0.09$	$0.0{\pm}0.0$		
Black	$4.7 {\pm} 0.23$	$4.5{\pm}0.18$	$4.2{\pm}0.25$	$3.5 {\pm} 0.23$	$2.9{\pm}0.18$	$2.2 {\pm} 0.15$	$0.0{\pm}0.0$		
Fresh weight (mg seedling <sup>-1</sup> )									
Pink	$1.85 \pm 0.023$	$1.87 {\pm} 0.015$	$1.82 \pm 0.023$	$1.77 {\pm} 0.018$	$1.71 \pm 0.036$	$1.58 {\pm} 0.035$	$1.38 \pm 0.034$		
Violet	$1.85 {\pm} 0.023$	$1.81 {\pm} 0.021$	$1.75 {\pm} 0.012$	$1.67 {\pm} 0.021$	$1.59 {\pm} 0.025$	$1.26 {\pm} 0.045$	$0.0{\pm}0.00$		
Black	$1.85 {\pm} 0.023$	$1.83{\pm}0.015$	$1.76{\pm}0.015$	$1.74{\pm}0.034$	$1.57 {\pm} 0.020$	$0.0{\pm}0.00$	$0.0{\pm}0.00$		
Dry weight (mg seedling <sup><math>-1</math></sup> )									
Pink	$0.28 {\pm} 0.02$	$0.30 {\pm} 0.02$	$0.28 {\pm} 0.02$	$0.27 {\pm} 0.01$	$0.26 {\pm} 0.02$	$0.24{\pm}0.02$	$0.21 \pm 0.02$		
Violet	$0.28{\pm}0.02$	$0.28{\pm}0.02$	$0.27 {\pm} 0.02$	$0.26 {\pm} 0.028$	$0.24 {\pm} 0.02$	$0.19 {\pm} 0.02$	$0.0{\pm}0.00$		
Black	$0.28{\pm}0.02$	$0.28{\pm}0.02$	$0.27{\pm}0.02$	$0.27{\pm}0.02$	$0.24{\pm}0.02$	$0.0{\pm}0.00$	$0.0{\pm}0.00$		

Table 2. Effects of loom-dye effluent on seedling parameters of red amaranth seedlings

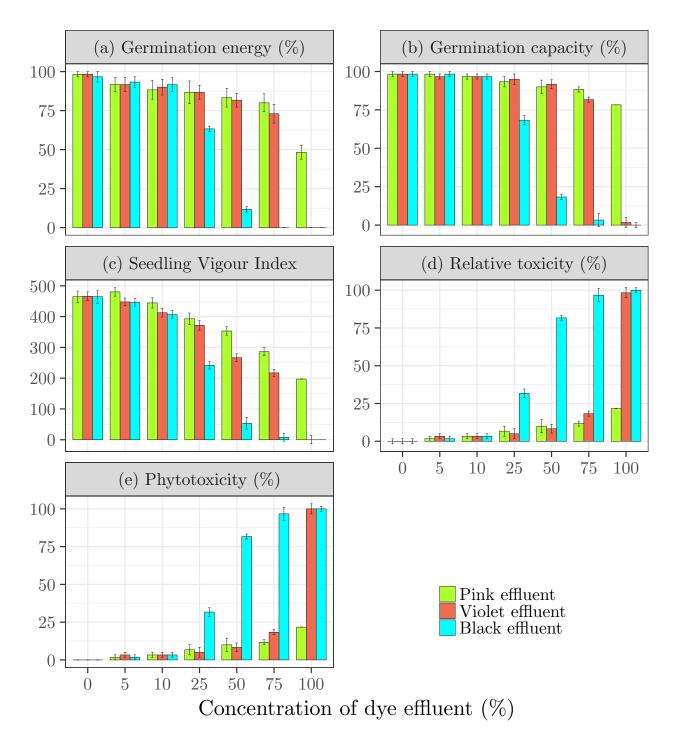


Figure 1. Effects of loom dye effluent on (a) germination energy, (b) germination capacity, (c) seedling vigour index, (d) relative toxicity, and (e) phytotoxicity of amaranthus seed

and subsequently carbohydrates and protein metabolites of the membrane are altered (Kannan and Upreti, 2008), which might lead to reduction in absorption of water by the seeds/seedlings. Suppression of germination at higher concentrations of effluent might be due to high levels of total dissolved solids which enhanced the salinity and conductivity of the solute absorbed by the seeds before germination. The promotion of seedling growth by lower concentration of effluent might be due to the presence of plant nutrient in the effluent.

#### 3.2.2 Germination capacity

The germination capacity of red amaranth significantly reduced with higher levels of effluent concentration (Fig. 1b). The highest values of seed germination (98.3%) at 7 DAS were recorded at control and 5% pink and black dye and the lowest values of germination percentage (0%) were recorded at 100% concentration of black dye effluent. In effluent treatment, maximum germination capacity was in 5 and 10% concentrations of effluent and it decreased as the effluent concentration increases. The maximum seed germination capacity (98.3%) was recorded at 5% effluent of pink and black dye. At the same time, the minimum germination capacity was recorded in 100% concentration of the three dye effluents. For all effluents, germination capacity decreased with increasing the effluent concentrations. Germination capacity was affected more in black and violet dye effluent than the value of pink colour effluent with the increased level of effluent concentrations.

Some researcher related this delayed germination directly with the presence of higher salt/metal concentrations (Baruah and Das, 1997), while others with lower amounts of auxin production which is also a result of higher metal ion concentrations (Mukherji and Das, 1972).

The increased germination percentages at lower effluent concentrations (5 and 10%) than those of higher concentrations (more than 10%) indicated the stimulation of physiologically inactive seeds of the lot by the effluent treatment (Fig. 1a,b) as reported by Vinod (2014) and Suresh et al. (2014). It might be also due to the lower concentrations of effluent, which created the favourable environmental condition for the germination and utilization of nutrients present in the effluent (Kannan and Oblisamy, 1992). The diluted effluent might have played a role in promoting plant growth in lower concentrations (Augusthy and Mani, 2001). At the same time the higher concentrations of loom-dye effluent inhibited the germination of red amaranth; it might be due to the effect of higher amount of total solids and heavy metals stress on the seed germination process during effluent treatment. The salt content outside the seed is known to act as limiting factor and causes less absorption of water by osmosis and inhibit the germination of seeds (Palanivel et al., 2004; Malla and Mohanty, 2005).

#### 3.2.3 Germination speed

The interaction effect between effluent type and their concentrations on germination speed was varied as presented in Table 2. The maximum speed of germination (100%) was observed in control and the minimum from 100% of all the effluent (Table 2). Exposition to different effluent water solutions inhibited the germination speed by 5.1 to 100% in comparison with the germination speed in the control solution. Undiluted dye effluent statistically inhibited the growth of red amaranth significantly the root length in undiluted wastewater was slightly lower than in the control sample which affected speed of germination. Speed of germination increased up to 25% concentration of pink dye effluent whereas it decreased with the increasing level of concentration of other effluents. Rashid et al. (2010a,b) reported that seeds of Bidens pilosa and Lolium perenne exhibit slower germinatation speed when they were exposed to toxic plant extracts.

#### 3.2.4 Root and shoot lengths

The effect of synthetic loom-dye effluent application on plant rooting and shooting varied with the effluent type and concentration used in the experiment. The higher concentrations of loom-dye effluent minimized the fresh and dry weight of seedlings. The growth parameters such as root and shoot length (Table 2) at 7 DAS showed reduction as the concentration of effluent increases. The maximum root length (3.7 cm) and shoot length (4.9 cm) were observed at 7 DAS in 5% concentration pink dye effluent. The minimum range of root length and shoot length was obtained from 100% of all the effluent. No radicle was formed from the 75% concentration of black and 100% effluent of both violet and black. At 100% effluent concentration, no shoot growth was found for violet and black dye effluent. Inhibitory as well stimulatory effects of various effluents on the germination of a number of plant species were observed by many researchers (Baruah and Das, 1997; Crowe et al., 2002; Nawaz et al., 2006; Yousaf et al., 2010).

The germinated seeds did not get any oxygen thus restricting their energy supply through aerobic respiration, which was necessary for growth and development of young seedlings. The net result observed the growth restriction of radicle and plumule (Hadas, 1976). Singh et al. (2006) studied the effect of fertilizer factory effluent on seed germination, seedling growth and shoot and root lengths of gram (*Cicer aeritenum*) at different concentrations of the effluent and time intervals and reported at 25% concentration of the effluent, growth promotion in terms of root and shoot

length was recorded on 21 d. The lower concentration of effluent had many nutrients such as nitrogen, phosphorous which might have promoted the plant growth as suggested by Augusthy and Mani (2001).

The interference of heavy metals decreased the root and shoot length of the plant might be due to the effect of physiological processes of plant and it also involved in inhibition of enzyme activities, affected the nutrition, water imbalance and alternation of hormonal status changed the membrane permeability (Sharma and Dubey, 2005). Vaithiyanathan and Sundaramoorthy (2017) found the highest shoot length (29.10 cm seedling<sup>-1</sup>), root length (6.80 cm seedling<sup>-1</sup>) in African marigold at lower concentration (10%) of sugar mill effulent and the lowest shoot length (9.43 cm/seedling) and root length (1.83 cm seedling<sup>-1</sup>) at higher concentration (100%) of sugar mill effluent.

#### 3.2.5 Fresh weight and dry weight

The seedling fresh weight and dry weight increased in the lower concentration and decreased in higher concentration of loom-dye effluent shown in le2Table 2. The highest fresh weight (1.87 mg seedling<sup>-1</sup>) and dry weight (0.30 mg seedling<sup>-1</sup>) were observed at 7 DAS in 5% concentration of pink dye effluent. The minimum fresh weight and dry weight were recorded in 100% of dye for all the three effluents. In all cases, fresh weight decreased with the increasing level of effluent concentration. These observations were in conformity with previous reports of various industrial effluent treatments on various crops (Sarathchandra et al., 2006).

The presence of optimum level of nutrients in the lower concentrations of loom-dye effluent might have increased the fresh weight and dry weight of seedlings because of various chemicals present in the effluent (Mishra, 1987)(Mishra et al., 1987). The higher concentrations of loom-dye effluent decreased the fresh weight and dry weight of seedlings. The reduction in seedlings weight might be due to the poor growth of seedlings under effluent stress. These observations were in conformity with (Balashouri, 1994) in *Vigna radiata*, *Cajanus cajan* and *Sorghum bicolor*. The root which continuously remained in direct with the effluent and hence the higher concentrations of the effluent could affect cell multiplication or the growth of root (Kannan and Upreti, 2008).

Vaithiyanathan and Sundaramoorthy (2017) found the highest fresh weight (3.59 g seedling<sup>-1</sup>) and dry weight (0.420 mg seedling<sup>-1</sup>) in African marigold at lower concentration (10%) of sugar mill effluent and the lowest fresh weight (0.760 mg seedling<sup>-1</sup>) and dry weight (0.074 mg seedling<sup>-1</sup>) at higher concentration (100%) of sugar mill effluent.

#### 3.2.6 Seedling vigour index

The decrease in seedling vigour index of red amaranth was observed in all tested effluents solutions with their increasing level of concentrations (Fig. 1c). The maximum vigour index (480%) was observed at 7 DAS in 5% concentration of pink dye effluent. The minimum seedling vigour index was recorded at 7 DAS in 100% concentration of all three dye effluents. The variation of seedling vigour might be due to genotypic differences and the effluent stress in different concentrations of the effluents. Seedling vigour index affected more by the black and violet effluents than the pink effluent.

#### 3.2.7 Relative toxicity of seed germination

Minimum relative toxicity percentage (1.7–3.3%) of seed germination of red amaranth was with 5% concentration and maximum (86.7–100%) was recorded at 100% (Fig. 1d). At higher concentrations of the effluent toxic effects was observed. As far as percentage of relative toxicity of red amaranth treated with dyeing industry effluent is concerned, minimum relative toxicity was observed at 5% concentration and maximum at 100% concentration. Above 10% concentration of the effluent, the relative toxicity was significantly increased. Maximum relative toxicity was observed in black dye effluent. David and Rajan (2015) found minimum relative toxicity percentage in 25% of effluent concentration and that was increased gradually as the concentration was also increased.

#### 3.2.8 Phytotoxicity

Phytotoxicity was considerably increased with the increasing levels effluent concentration. The maximum phytotoxicity (100%) was observed at 7 DAS in 100% concentration of black dye effluent. The minimum range of phytotoxicity was recorded at 7 DAS in control (Fig. 1e). High level of phytotoxicity of loom-dyeing effluent to red amaranth might be due to its high pH, EC, TDS and metallic contents. With effluent application, minimum phytotoxicity was observed at 5% concentration which was identical with 10% concentration for all three types of effluent. Maximum phytotoxicity was observed in untreated effluent red and violet colour dye effluent whereas pink dye effluent shows less phytotoxicity to red araranth. Similar toxic effects of dyeing industry effluent were observed by David and Rajan (2015) in ladies finger.

#### 4 Conclusion

Germination and early seedling growth of red amaranth were significantly affected by the different types and concentrations of dyeing effluents. Based on the experimental observations, it is concluded that

the physico-chemical parameters such as electrical conductivity, total dissolved solids were relatively high in the loom-dye effluent causing toxic to the plant, severely affected seed germination and early seedling growth. Germination energy, germination capacity, germination speed, root length, shoot length and seedling vigour index were declined gradually with increasing effluent concentrations. Effluent with lower EC and TDS (e.g. pink dye effluent) had the less adverse effect on the germination and early growth of roots and shoots. But the effluents with high EC and TDS affected the germination and growth drastically. The exposure of seeds to the low concentration of dye effluent during germination gave no or lower phytotoxicity to early seedling growth of red amaranth as compared with the higher concentrations. As a result, the untreated dyeing industry effluent could possibly lead to soil deterioration and low productivity. The results of this study suggest that the physico-chemical properties of the effluent wastewater of loom-dyeing industry might be harmful for crops and human being with food chain contamination. So, the use of loom-dye effluent wastewater should be restricted in crop fields without proper treatment.

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#### **Conflict of interst**

The authors declare that there is no conflict of interests regarding the publication of this paper.

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