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Phytotoxic Effect of Paper Mill Effluent Treatment on Seed Germination and Seedling Growth of Wheat (*Triticum aestivum*) and Radish (*Raphanus sativus*)

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Abstract

The present study has been carried out to see the impact of paper mill effluent treatment on seed germination and seedling growth of wheat (*Triticum aestivum*) and radish (*Raphanus sativus*). The pot culture experiment was conducted with the different concentrations viz., 20%, 40%, 60%, 80%, and 100% along with control. The growth parameters such as germination percentage, seedling vigour, shoot length, root length, fresh and dry weight, tolerance index, phytotoxicity were studied. The results indicate that at lower concentration there is a significant increase in the percentage of seed germination and other growth parameters but decreased with increase in effluent concentration.

Keywords: *Paper mill effluent, Wheat, Radish, Seed germination, Tolerance, Phytotoxicity*

1. Introduction

Diverse nature of agro-based industrial effluents from various industries are disposed off in to soil and water bodies, which has been causing major pollution problem. These effluents contain toxic materials with varying characteristics from simple nutrients to highly toxic substances. To economize the irrigation water industrial effluents are now a days commonly used for irrigation. Some effluent contain considerable amount of nutrients, which may prove beneficial for plant growth and it serves as an additional potential source of fertilizer for agricultural use **(Day, 1973, Bauwer and Chaney, 1974)**. The paper industry is one of the largest industries in India, consuming large amount of water **(Trivedy and Raj, 1992)**. At present, there are 666 pulp and paper mills in India, of which 632 units are agro-residue and recycled fiber based units with manufacturing capacity of 7.6 million tons **(CPPRI, 2005)**. The effluents generated from pulp and paper mill is dark brown in colour and having high BOD, COD, TS and organic carbon **(Kirk *et al.*, 1983; Singh *et al.*, 1994)**. It is estimated that 273-450 M^3 of water is required per ton of paper produced **(Subrahmanyam and Hannmanulu, 1976)** that consequently, generate 300 M^3 of waste water **(Khanna *et al.*, 1990)**. Effluents released from pulp and paper mill contains large amount of heavy metals which get accumulated in plant and soil. They cause huge damage to plants and biological systems **(Chandra *et al.*, 2010, Kathirvel, 2012** and **Mehta and Bhardwaj, 2012)** and even to ground water quality and soil **(Balakrishnan *et al.*, 2005, Senthilkumar *et al.*, 2011, Chopra *et al.*, 2011, Tripathi *et al.*, 2014)**. Efforts have been made by different workers to determine the toxic effect of paper mill effluent on seed germination of various crops such as mustard, pea and rice **(Medhi *et al.*, 2008)**, mustard and pea **(Medhi *et al.*, 2011)**, black gram **(Gupta *et al.*, 2016)**, *Vigna radiata* L. **(Sharmila *et al.*, 2009)**, wheat, cabbage, green gram and groundnut **(Patel *et al.*, 2013)**, *Vigna radiata* L. **(Kumar and Chopra, 2012)** and fenugreek **(Kumar *et al.*, 2010)**.

The present study is dealt with characterization of treated paper mill effluent and its effect on seed germination and seedling growth behavior of wheat and radish in different concentrations.

2. Materials and methods

An effluent sample of the paper mill (Shree Jagdambe paper mills Ltd.) situated at Sirsa, Haryana was used for the present study. Different concentrations of effluent viz., 20%, 40%, 60%, 80%, and 100% were prepared by using distilled water along with control. The seeds of wheat (*Triticum aestivum*) and radish (*Raphanus sativus*) were procured from certified local supplier. Healthy seeds of uniform size were selected and surface sterilized with 0.1%

solution of mercuric chloride for 5 min to avoid any fungal growth, followed by washing for 4-5 times with distilled water prior to germination studies. Ten seeds were sowed in each pot of different concentrations to study the response of test plants in duplicate. Germination parameters were recorded on the 14th days. Growth of the root and shoot length were measured with the help of meter scale. Fresh and dry weights of test plants were determined on a digital balance.

2.1 Germination percentage

Germination percentage was calculated by using the following formula:

$$\text{Germination percentage} = \frac{\text{Total number of seeds germinated}}{\text{Total number of seeds sown}} \times 100$$

2.2 Seedling vigour index

Vigour index of the seedling were calculated by using the formula proposed by **Aery (2010)**.

Seedling vigour index = Germination percentage × seedling length

2.3 Tolerance index

Tolerance index of the seedlings were calculated by using the formula proposed by **Turner and Marshal (1972)**.

$$\text{Tolerance index} = \frac{\text{Mean length of longest root in effluent treatment}}{\text{Mean length of longest root in control}}$$

2.4 Phytotoxicity

The percentage of phytotoxicity of effluent was calculated by using the formula proposed by **(Chou *et al.* 1978)**.

$$\text{Phytotoxicity (\%)} = \frac{\text{Radical length of control - Radicle length of treatment}}{\text{Radical length of control}} \times 100$$

3. Statistical analysis

Statistical analysis was done by using SPSS software ver. 20. Data were analyzed for mean, standard deviation and one way analysis of variance. ANOVA is used to compare these data between treated seedlings and control seedlings. P values less than 0.05 was considered to be significant.

Pic-1 Wheat (*Triticum aestivum*)

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Pic-2 Radish (*Raphanus sativus*)

4. Result and discussion

The results of physico-chemical characteristics of paper mill effluent are presented in **Table-1**.

The characterization of the effluent revealed that it is dark brown in colour having temperature of 22.4 ^{o}C and turbidity of 13.1 NTU. The pH of the effluent was found alkaline in nature having pH of 8.4. The value of total solids, total dissolved solids and suspended solids were 1900 mg/l, 1540 mg/l and 360 mg/l respectively. The electrical conductivity (EC) value was 2.97 µS. The effluent had higher BOD, 332 mg/l and COD, 1060 mg/l. The higher concentration of BOD and COD indicated the higher concentration of organic and inorganic substances in the effluent. The recommended BIS values for BOD and COD are 100 mg/l and 250 mg/l respectively. Further the values of total hardness (TH), total alkalinity (TA) and chloride were 870 mg/l, 490 mg/l and 530 mg/l respectively. The presence of high amount of COD, BOD, suspended solids, total hardness were also recorded by **Baruah *et al*., 1996**; **Medhi *et al*., 2008.**

Table-1 The physico-chemical analysis of paper mill effluent

S.No.	Parameters	Effluent	BIS for irrigation water
1	Colour	Dark brown	
2	pH	8.4	5.5-9
3	Temperature (^{o}C)	22.4	-
4	Turbidity(NTU)	13.1	10
5	TS (mg/l)	1900	1900
6	TDS (mg/l)	1540	2100
7	SS(mg/l)	360	200
8	EC (µS)	2.97	-
9	BOD (mg/l)	332	100
10	COD (mg/l)	1060	250
11	TH (mg/l)	870	600
12	TA (mg/l)	490	600
13	Chloride (mg/l)	530	500

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The observations made on the effect of paper mill effluent on growth parameters of wheat (*Triticum aestivum*) and radish (*Raphanus sativus*) are presented in **Table 2 & 3**. In plant responses tests, the treatments on wheat (Table 2), the maximum value of germination percentage (90%) was recorded at control condition while 43.33% germination was recorded at 100% dilution.

Table-2 Growth parameters of wheat under different concentration of paper mill effluent

Treatment	Germination (%)	Shoot Length (cm)	Root Length (cm)	Vigour index	Fresh Weight (gm)	Dry Weight (gm)
Control	90.00 ±10.0	12.63 ±1.40	14.20 ±2.26	2396.67 ±145.94	0.32 ±0.02	0.13 ±0.03
20%	80.00 ±10.0	15.67 ±1.45	16.23 ±1.85	2560.00 ±434.61	0.31 ±0.02	0.12 ±0.02
40%	73.33 ±5.77	12.23 ±2.32	8.73 ±1.21**	1543.00 ±232.70**	0.29 ±0.04	0.08 ±0.01**
60%	60.00 ±10.0**	12.87 ±1.72	11.13 ±2.58	1464.00 ±452.48**	0.22 ±0.06**	0.08 ±0.01**
80%	53.33 ±11.55**	11.07 ±0.61	7.00 ±1.44**	970.67 ±251.72**	0.22 ±0.04**	0.05 ±0.01**
100%	43.33 ±5.77**	10.40 ±0.72	4.83 ±0.47**	660.00 ±87.18**	0.17 ±0.03**	0.04 ±0.01**
F-value	11.040*	4.515*	17.906*	19.05*	8.471*	18.528*

** = Significant at (P<0.05), ** =significantly different to control, (n=3, Mean ± SE)*

Table-3 Growth parameters of radish under different concentration of paper mill effluent

Treatment	Germination (%)	Shoot Length (cm)	Root Length (cm)	Vigour index	Fresh Weight (gm)	Dry Weight (gm)
Control	86.67 ±5.77	5.40 ±0.79	12.57 ±1.29	1557.33 ±117.5	0.26 ±0.02	0.09 ±0.01
20%	96.67 ±5.77	5.77 ±0.25	12.27 ±1.34	1746.33 ±221.68	0.23 ±0.02	0.06 ±0.02
40%	93.33 ±5.77	5.47 ±1.10	10.60 ±4.17	1503.67 ±494.96	0.22 ±0.02	0.04 ±0.01**
60%	80.00 ±10.0	4.77 ±0.67	8.77 ±3.02	1083.00 ±287.18	0.18 ±0.01**	0.05 ±0.01**
80%	70.00 ±26.46	3.83 ±0.31	6.00 ±0.30**	686.67 ±255.8**	0.19 ±0.03**	0.04 ±0.02**
100%	46.67 ±15.28**	3.47 ±0.64**	5.63 ±0.60**	415.33 ±111.79**	0.13 ±0.01**	0.02 ±0.02**
F-value	5.412*	5.609*	5.408*	10.793*	18.285*	8.373*

** = Significant at (P<0.05), ** =significantly different to control, (n=3, Mean ± SE)*

Germination % in wheat crop was decreased in germinating seedlings treated with paper mill effluent as compared to100% dilution. These results were in accordance with the work of **Khan *et al* (2011)** who reported that seed germination was decreased when treated with waste water which is contaminated by textile industrial effluents.

In case of treatments on radish (Table-3), the maximum value of germination percentage (96.6%) was recorded at 20% dilution while 46.67% germination was recorded at 100% dilution. From 20% onwards the germination percentage was found to be gradually declined. In present study ANOVA analysis on data showed that effluent concentrations 60%, 80% and 100% significantly (P<0.05) affected germination as compared to control in wheat crop

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while 100% concentration significantly (P<0.05) affected germination as compared to control in radish crop.

Table-4 Effluent tolerance index and phytotoxity (%) under different concentration of paper mill effluent

Treatment	Effluent Tolerance Index		Phytotoxicity (%)	
	Wheat	Radish	Wheat	Radish
Control	–	–	–	–
20%	1.17±0.31	0.97±0.01	17.49±29.92	2.42±1.46
40%	0.79±0.20	0.84±0.33	36.95±17.04	15.98±32.73
60%	0.63±0.17	0.71±0.27	20.99±19.1	29.16±27.1
80%	0.49±0.08	0.48±0.06	50.60±8.31	51.82±6.46
100%	0.34±0.04	0.45±0.08	65.64±3.79	54.80±7.54
F-value	9.060*	3.944*	9.344*	4.024*

** = Significant at (P<0.05)*

In both plant response cases (wheat and radish), seedlings vigour achieved maxima at 20% concentration effluent and gradually decreases on further elevation on concentration. The effluent concentration 40%, 60%, 80% and 100% significantly (P<0.05) affected seedling vigour as compared to control in wheat crop while 80% and 100% significantly (P<0.05) affected vigour index as compared to control in radish crop.

The maximum value of shoot length was achieved at 20% dilution for both the test crops while maximum value of root length achieved at 20% and control for wheat and radish respectively. The maximum value of fresh and dry weight was recorded at control condition for both the test crops. Tolerance index value decreased with increase in effluent concentration while percentage of phytotoxicity values showed a reverse trend in both the test crops (wheat and radish). The observation was conformity with **Mycin, 2014**. The degree of inhibitory effect of paper mill effluent on the growth parameters of wheat (*Triticum aestivum*) and radish (*Raphanus sativus*) increased with the increase in the concentration of effluent when compared to the control. The inhibitory effect may be due to the excess of total nitrogen, phosphate, potassium, sulphate, chloride present in the effluent which happens to be injurious to plant growth by reducing water absorption and affecting other metabolic processes in the plants **(Dutta and Boishya, 2000, Subramani *et al*., 1995)**.

A progressive decrease of germination and seedling growth has recorded with the increasing concentration of paper mill effluent. Similar observations were also studied by many workers (**Reddy and Borse (2001); Medhi *et al*., (2008); Medhi *et al*., (2011)** and **Kamlesh and Kidwai, (2016).**

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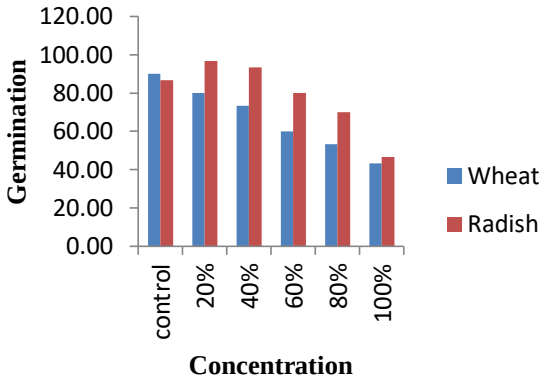

Fig.1 Germination percentage on different concentration of effluent

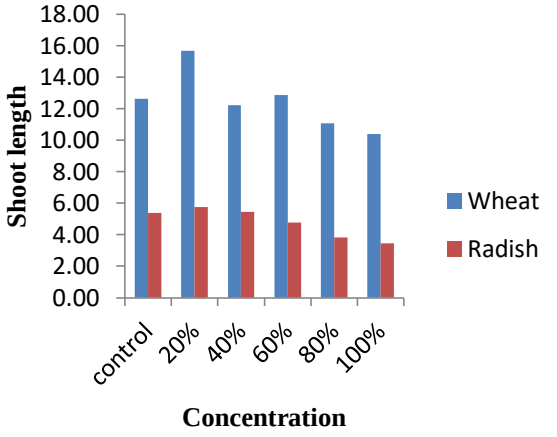

Fig.2 Shoot length (cm) on different concentration of effluent

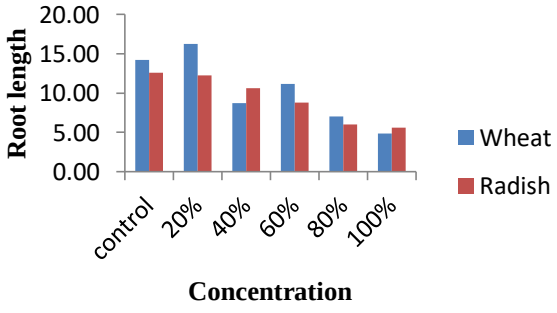

Fig.3 Root length (cm) on different concentration of effluent

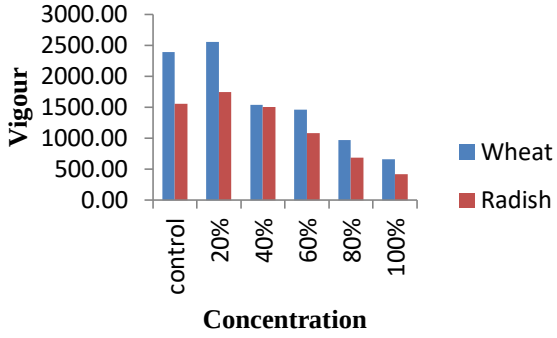

Fig.4 Seedling vigour on different concentration of effluent

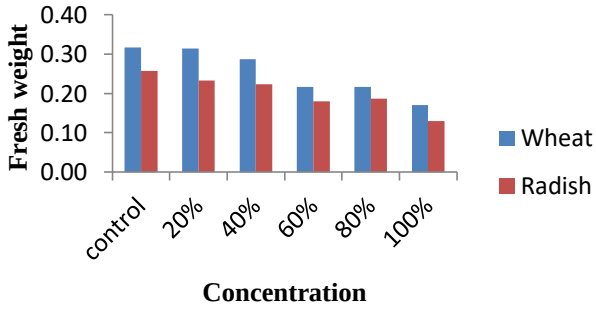

Fig.5 Fresh weight (gm) on different concentration of effluent

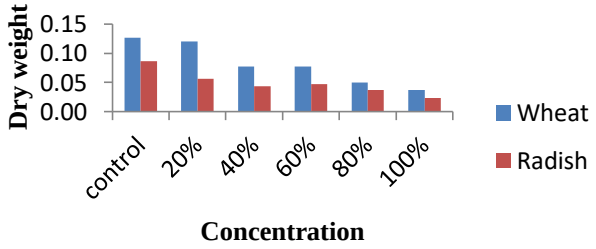

Fig.6 Dry weight (gm) on different concentration of effluent

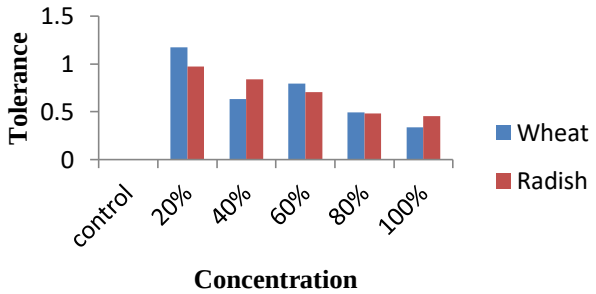

Fig.7 Tolerance index on different concentration of effluent

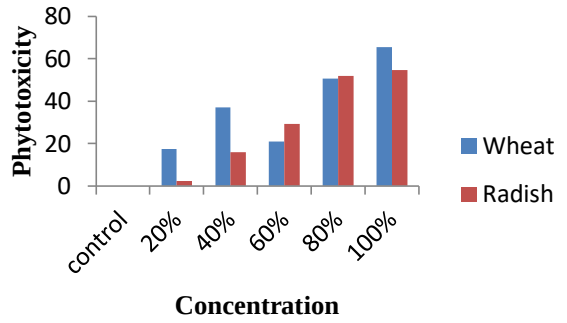

Fig.8 Phytotoxicity percentage on different concentration of effluent

5. Conclusion

From the conducted work, it is concluded that paper mill effluent had adverse effect on growth parameters at higher concentration, but lower concentrations favoured initial plant growth of both the plant crops. However, long term research work should be conducted to explore the effect of paper mill effluent on above suggested aspects before its use for irrigation.

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