



Effect of Vegetable Oil Mill Effluents on Physiological Properties of *Brassica compestris* L. Seeds

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Industrialization has made our day-to-day life easier and is also a key determinant of the economic growth of any nation. But on the dark side, industries are also known to cause almost all types of pollution including soil, air, and water pollution. Untreated industrial effluents when discharged in agricultural fields affect overall crop production. OME is believed to induce a phytotoxic effect on seed germination and overall plant growth. This paper aims to test the toxicity of soybean and mustard oil mill effluents and their impact on the Vitality Index, Seed Vigour Index, and Tolerance Index of seeds of *Brassica compestris* L. The study was conducted during July 2020 - June 2022. Two sets of experiments were settled, one for mustard OME and another for soybean OME. Various concentrations of OME were prepared; 0% oil mill effluent (tap water), 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, and 100% OME (not diluted). Treatment with tap water was

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treated as a control. A germination test with 10 seeds mounted on moist filter paper was conducted in an incubator at a temperature of 23°C. Seed vitality index, Seed vigour index, Tolerance index, and Phyto-toxicity were calculated with standard methods. Vitality index (VI) decreases to almost 1/5 with increasing OME concentration as compared to control (without OME) with both the OME. Up to 10% OME concentration in both cases there is almost no negative impact, but after that soybean OME concentration shows more negative effect on the seed vigour index. The tolerance index (TI) was reduced to almost 50% with increasing OME concentration from 0% (water) to 100% in both treatments. Toxicity is more pronounced with soybean OME during the early germination period but later it shows that toxicity increases with increasing OME concentration with both the OME. It can be concluded that higher concentrations of vegetable oil mill effluent negatively affect seed vitality, vigour and this may be due to reduced tolerance and increased toxicity with higher concentrations of OME. All these parameters under study have a significantly high correlation with the concentration of OME.

Keywords: Effluent; Vitality; Vigour; Toxicity; Tolerance; Seedling.

1. INTRODUCTION

Industrialization brings economic prosperity to a nation, it results in an increase in wealth, production of goods, standard of living, better transportation, better job opportunities, etc., but what else it brings with this is a threat of pollution. Various industries are known to cause almost all possible types of pollution. In the wake of recent industrialization and fast urbanization, the quality of groundwater in industrial areas has become an increasing concern due to contamination by various toxic chemicals [1,2].

Industries not only consume a lot of fresh water but also generate a huge amount of wastewater or industrial effluent that contains many inorganic-organic wastes as well as toxic elements. This production of wastewater is an inevitable outcome of industrial processes and is hazardous to the environment as in most cases, industries discharge their wastes into nearby fields, water bodies, and streams without proper treatment; which causes the introduction of many contaminants or potential pollutants such as heavy metals into soil and groundwater which affect these precious resources [3, 4]. In the race for rapid industrialization and urbanization, groundwater quality in industrial areas is highly compromised due to contamination by industrial effluents, wastewater, and emissions, which are discharged without any treatment in these resources [2,5]. It can result in deterioration of water quality and change in physico-chemical properties; by altering pH [6], total suspended solids [7], chemical oxygen demand (COD), biological oxygen demand (BOD), heavy metal, and toxic elements [8].

India is an agriculture-based country and thus is a major consumer of water resources for

irrigation [9]. The agricultural sector accounts for about 70% of total water usage, which makes it the largest consumer of freshwater worldwide [10]. The agricultural sector alone demands more water than utilizable renewable water resources [11]. In water-scarce arid and semi-arid regions of the country, where the natural endowment of water is poor [12], there is very high per capita water use in irrigation [11]. In India poor irrigation systems and fast-track groundwater depletion result in 90% of water use in irrigation [13]. Conventional water resources are more prone to contamination by industrial wastewater [14]. The major risk associated with polluted water sources is that when this water is used for irrigation, it can potentially cause infectious diseases in grazing animals that feed on pastures irrigated with this water, and to people who feed on such crops [15].

Increasing population and agriculture cause great demand for water for irrigation while gallons of industrial effluents are disposed of untreated in nearby water bodies or fields. The rising demand for water in the agriculture sector and diminishing supply have made treated industrial effluents and domestic wastewater an attractive alternative to conventional irrigation. Application of industrial and domestic wastewater in irrigation may prove beneficial to plant growth as it provides nitrogen, phosphorus, and other nutrients to the soil, besides helping in water conservation [16]. Thus, wastewater irrigation helps in nutrient recycling, reduces direct fertilizer inputs in agricultural fields, and minimizes pollution loads of the water bodies receiving wastewater [17,18]. It was reported that most wastewater irrigation leads to an increase in the yield of most of the crops along with decreasing the demand for chemical fertilizers thus saving the total costs of farmers [19].

Effluent irrigated soil also shows change in physico-chemical properties. Untreated effluents affect soil parameters and ultimately soil fertility [20]. Improper management of industrial waste products ultimately results in low productivity of soil by disposing of it in soil and agriculture fields which leads to lower biological and chemical soil quality index and affects the overall soil quality index [21]. Thus, industrial effluents eventually affect agriculture production and food security [22]. This paper aims to test the toxicity of soybean and mustard oil mill effluents and their impact on the Vitality Index, Seed Vigour Index, and Tolerance Index of seeds of *Brassica campestris* L.

2. MATERIALS AND METHODS

The study was conducted in Janki Devi Bajaj Govt. Girls College, Kota, India during July 20-June 2022. Certified seeds of *Brassica campestris* L variety Pusa Bold were obtained from the Department of Agriculture, Bundi district, Rajasthan, India. Healthy seeds with uniform size and shape were selected and sterilized with 0.1% HgCl₂ for 2 minutes, then washed thoroughly twice with tap water to remove traces of HgCl₂. Sterilization of seeds is followed by soaking in distilled water for 2-3 days.

Both the treated effluent samples were collected in plastic containers from outlets of mustard and soyabean oil mills of Bundi, Rajasthan, and stored at 5°C to preserve the physicochemical properties of the effluent. Effluent is then diluted from 0% oil mill effluent (water, control) to 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, and 100% OME (not diluted). Two sets of experiments were settled, one for mustard OME and another for soybean OME. 10 seeds of *Brassica* were then placed at equal distances in pre-labeled Petri dishes containing filter paper soaked in water for each set of experiments. Later on, 5ml of each prepared concentration of both mustard and soybean OME was used to irrigate both separate sets of treatment and then left in the dark for 48 hours for germination. Three replicates of all the experiments were prepared and maintained.

Seed vitality index is calculated using the standard formula given by Association of Official Seed Analysis [23].

$$\text{Vitality index (VI)} = S \times GI$$

Where

GI is the Germination index and S is the length of seedlings.

Seed Vigour Index (SVI) is calculated by multiplying germination percentage (GP) and seedling length (mm).

Seed vigour index= Germination percentage × length of seedling

The tolerance index of the seedling was calculated using the formula given by Turner and Marshal [24].

$$\text{Tolerance index} = \frac{\text{Mean length of longest root in treatment}}{\text{Mean length of longest root in control}} \times 100$$

The percentage of phytotoxicity of effluent was calculated using the formula proposed by Chou et al. [25].

$$\text{Phytotoxicity} = \frac{\text{Radicle length in control} - \text{Radicle length in test}}{\text{Radicle length in control}} \times 100$$

The correlation of the response of various OME concentrations to the physiological parameters (toxicity, vitality index, seed vigour index, and tolerance index) was statistically analyzed using Pearson's Correlation Coefficient.

3. RESULTS

Vitality index (VI) decreases to almost 1/5 with increasing OME concentration (Table 1, Fig. 1) as compared to control (without OME) with both the OME. After 20% OME vitality index shows a more decreasing trend with soybean OME but after 50% OME concentration vitality index shows the same decreasing trend in both OME.

The seed vigour index (SVI) also shows a marked decrease with increasing OME concentration (Table 1, Fig. 1). Up to 10% OME concentration in both cases there is almost no negative impact, but after that soybean OME concentration shows more negative effect. SVI with both the OME follows the same pattern after 60% of both OME have almost the same negative effect on SVI.

The tolerance index (TI) reduced to almost 50% with increasing OME concentration from 0% (water) to 100% (Effluent) with both the OME (Table 1, Fig. 1).

Toxicity (Table 1, Fig. 1) is more pronounced with soybean OME during the early germination period but later it shows that toxicity increases

with increasing OME concentration with both the OME.

It was observed that Pearson's correlation coefficient between variable concentrations of soybean and mustard OME and toxicity was positive and between OME concentrations and

vitality index, seed vigour index and tolerance index was negative. In all the cases the values of *r* were higher than the table value. This represents the highly significant correlation of OME concentration and physiological parameters of mustard seed with both the OME (Table 2).

Table 1. Parameters related to the growth of *Brassica campestris* L. seedling with soybean OME (S OME) and mustard OME (M OME). (VI=Viability index, SVI=Seed vigour index)

OME Conc. (%)	Toxicity (%)		VI on 15 th day		SVI on 15 th day		Tolerance index on 15 th day	
	S OME	M OME	S OME	M OME	S OME	M OME	S OME	M OME
0	0	0	56.88±2.63	56.88±2.54	650±4.36	650±2.55	100	100
10	0	7.69±1.02	55.98±1.69	55.12±1.06	650±3.69	640±5.61	100	98.46 ±2.52
20	23±0.23	7.69±0.64	32.02±2.33	35.52±1.08	440±5.21	488±5.22	85±2.61	93.85±3.61
30	23±0.65	8.45±0.68	29.53±2.01	35.21±2.30	416±3.25	496±3.05	80±5.11	95.38±3.21
40	31±0.54	15±0.96	29.40±1.23	34.80±1.05	441±3.55	522±3.14	75.38±4.12	89.23±3.33
50	30±1.69	30.77±0.58	21.51±0.63	22.83±0.69	343±2.31	312±2.15	75.38±3.21	80±4.12
60	38±0.96	30.77±1.06	25.93±0.96	24.62±0.94	336±3.20	300±3.60	74±2.54	76.92±2.14
70	62±2.33	46.15±1.90	14.85±1.05	15.59±0.33	200±1.96	210±1.98	62±3.25	64.62±6.01
80	62±1.23	46±2.33	13.94±1.62	15.07±2.01	185±4.21	200±2.11	57±2.55	61.54±2.58
90	62±1.89	54±0.63	10.04±0.68	11.96±1.05	144±2.14	156±3.05	55±2.11	60±2.91
100	62±0.98	53.85±2.16	10.01±0.56	11.9±0.68	180±3.25	180±1.58	55.38±2.68	55.38±2.55

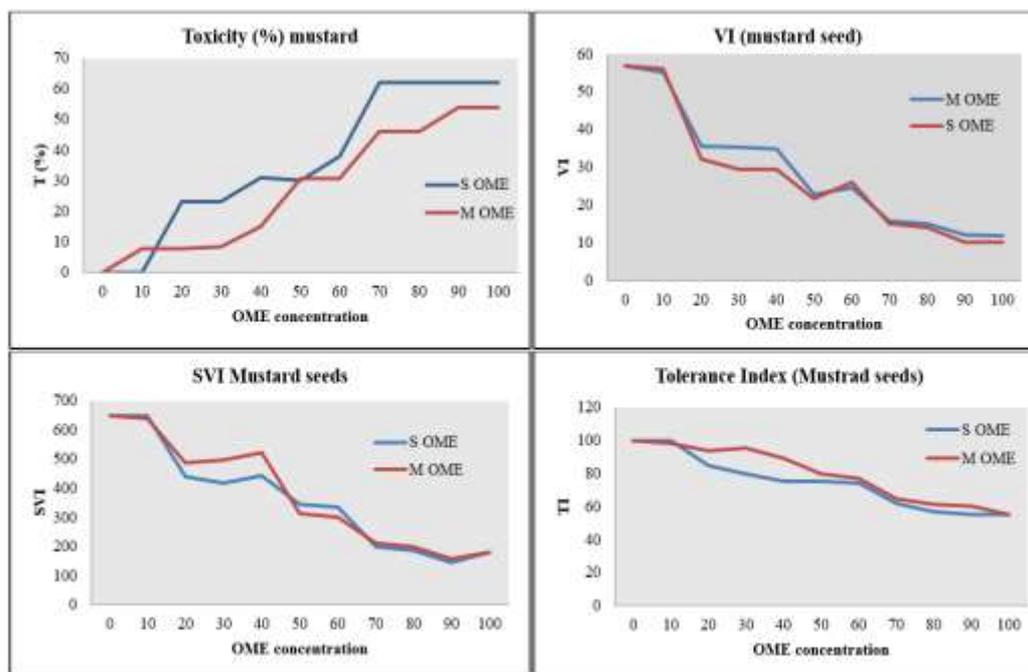


Fig. 1. Effect of Soybean and Mustard OME on Toxicity (%), Vitality Index (VI), Seed Vigour Index (SVI), Tolerance Index (TI) of *Brassica campestris* L. seeds

Table 2. Pearson's correlation coefficient with variable concentrations of soybean and mustard oil mill effluent and physiological parameters of *Brassica campestris* L seeds

	Soybean OME	Mustard OME
Toxicity	0.96	0.976
Vitality Index	-0.928	-0.953
Seed Vigour Index	-0.956	-0.96
Tolerance Index	-0.971	-0.979

4. DISCUSSION

Industrial effluents are usually considered as a serious problem but they can also be a solution for issues like chemical fertilizers and water scarcity when used in irrigation after proper treatment. Discharged effluent differs in quality based on the type of industrial raw material used. Therefore, physico-chemical examination of effluent is a must before its application in irrigation. Recent studies have shown the effect on the physico-chemical properties of soil after the application of effluent as an irrigant [7] and also proved that the effluent's pH, total suspended solids, and Chemical Oxygen demand can be brought to the permissible limit only after treatment [7]. Usually, raw or untreated effluents are more toxic than treated effluents and are efficient in causing plant growth inhibition, thus the degree of treatment is a key determinant of wastewater quality [26].

Many effluents may have high concentrations of heavy metals, some of which may have a major role in the growth and development of plants but are toxic beyond a certain level [27]. These metals along with many non-metal pollutants can easily enter the food chain and can disrupt mineral nutrient uptake by plants, inhibit photosynthesis, reduce enzyme activity, interfere with physiological processes, damage cell membranes, cease metabolite biosynthesis, and consequently reduce plant growth and yield [28,29].

Results of the present study prove that at lower concentration of soybean and mustard oil mill effluents, all the parameters studied viz. vitality index, seed vigour index, and tolerance index gives favourable results for *Brassica campestris* seeds. At the higher concentration of both influents, toxicity increases which causes retardation of seed germination; the increase in toxicity is comparatively higher for soybean effluent than mustard effluent. Toxicity increases with increasing OME concentration. It may be due to the higher concentration of heavy metals, non-metals, and other contaminants of the OME, which not only affect seed germination but the overall growth and development of seedlings. Effluent composition possesses recalcitrant chemicals with carcinogenic and mutagenic effects [30] and heavy metals present in effluent accumulate in the living cells causing a reduction of cell activities thus inhibiting of growth in plants [31]. The impact of toxicity of these pollutants or toxins can also be seen in later stages of plant

life as visible symptoms like chlorosis, yellowing and immature fall of leaves, poor growth, and retarded flowers, fruits, and seed yields. Ion toxicity also reduces the viability of the seeds [32].

Seed vitality is one of the primary determinants of high yield that directly determines the performance of seedling growth and plant growth. It is usually defined as the ability of any seed to germinate and produce natural seedlings during provided growth conditions. The present study witnessed a reduction in seed vitality with increasing OME concentration. Seed vitality is directly dependent on seed germination and seedling growth. There are many possible reasons for delay in the germination of seeds and lower seedling growth. Some of these include salt concentration outside the seeds during germination [33]; excess ammonia [34]; higher solid concentration in effluent [35]; and low oxygen supply [36].

The seed vigour test is a key quality parameter, to test the performance of the seed lot in the field or storage, thus its assessment is important to supplement germination and viability tests. The seeds with a higher vigour index are considered to be more vigorous and are highly active during seedling emergence [37]. Results of the present study state that seeds sown with a higher concentration of OME have low SVI and thus will be less active during seed germination and seedling emergence. This is probably due to the stressful seedbed environment of the field, as seeds are extremely sensitive to the physical stresses imposed by soil during germination and seedling expansion [38].

The tolerance index of any seed is its ability to tolerate stress during seed germination and early development i.e., seedling development. Results of the present study state a reduction in tolerance index with increasing OME concentration i.e., seeds become less tolerant to higher OME concentration, which is probably due to an increase in stressful conditions with concentration. Seeds can withstand only a certain level of toxicity in a seedbed environment and their performance reduces after that. Changes in the physiological mechanisms of plants during growth stages may be the reason for reduced tolerance [39].

Therefore, it is necessary to screen crops for their sensitivity or tolerance variance to different effluents before using them in agriculture [40].

Prabakaran et al. [41] also focused on the need to assess the quality of treated wastewater and its effects on plant species before using it for irrigation. Risk assessment and management are also necessary before irrigation with wastewater [42]. Results of the work done by Regar et. al. [43] and Regar & Jaiswal [44] also show that at higher concentration of OME, seed germination, and early seedling growth is negatively affected.

5. CONCLUSION

With the current study, it is clear that irrigation with suitable effluent or wastewater may be beneficial up to a certain level of concentration, and the effect of different effluents can differ for the same seed. In the present investigation, it is also clear that higher concentrations of vegetable oil mill effluent negatively affect seed vitality, and vigour and this may be due to reduced tolerance and increased toxicity with higher concentrations of OME. The tolerance index, vitality index, seed vigour index of mustard seeds and toxicity of OME to the mustard seeds were significantly correlated with variable concentrations of OME.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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