Effect of glass industry effluent on seed germination and biochemical parameters of *Glycine max* (Soyabean)

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**ABSTRACT**

Effluent generated by various processes in factories is directly discharged without any end point treatment into drains and used for irrigation purposes. The present study was carried out to assess the effects of glass industry effluent on seed germination and seedling growth of *Glycine max*. Different physico-chemical parameters of effluent were analysed. The pot culture laboratory experiment was conducted with the different effluent concentrations viz., 25%, 50%, 75% and 100% along with control (ground water) used as irrigation for *Glycine max* crop on different time interval. The growth parameters and biochemical parameters of *Glycine max* were measured as root length, shoot length, vigour index, chlorophyll content, ascorbic acid, fresh biomass and dry biomass on 21st day of sowing period. Maximum results of plant parameters were obtained in 25% effluent concentration diluted with 75% ground due to optimum level of nutrients present in it.

**Key words:** Biochemical parameters, Chlorophyll, *Glycine max*, Industrial effluent, Seed germination.

**INTRODUCTION**

Water is one of the most important and abundant component of the ecosystem. It covers about 71 percent of the Earth’s surface, and the oceans hold about 96.5 percent of all Earth’s water. All living organisms on the earth need water for their survival and growth (Patil *et al.*, 2012). Pollution may be defined as undesirable changes in physical chemical and biological characteristics of soil, water and air which are harmful for all living organisms. Water pollution is one of the greatest and raising concerns now a days. In recent years, considerable attention has been paid to industrial wastes discharged to surface water. Industrial Effluents are one of the major sources of pollution it contains various toxic metals, harmful dissolved gases, and several inorganic and organic compounds (Perfus *et al.*, 2002, Purushotham *et al.*, 2011). Untreated effluents are very rich in organic carbon, potassium, sulphate, phosphate, chloride, magnesium, heavy metals and many more toxic elements. The harmful nature of industrial effluent in relation to plant growth and development is well recognized owing to the presence of toxic chemicals present in it. Some macro and micro nutrients, which are important for plant growth and yield, become toxic, beyond tolerance limit and cause adverse effect on plant growth and yield.

The glass industry is highly concentrated it includes a variety of manufacturing facilities and produces various glass objects from a wide range of raw materials among which the most important ones are silica sand, glass cullet, and intermediate/modifying materials such as soda ash, limestone, dolomite and feldspar in addition there are psychosocial and physical environment impacts with high levels of work intensity and relatively low level of autonomy which can result in a range of stress related illness such as cardiovascular disease and mental health problems. Glass product have the potential to support the transition to a low carbon energy effluent and also a significant emitter of greenhouse gases particularly (CO₂ and CH₄).

Discharges from glass industry may be affected by glass Solids, some soluble glass-making materials (*e.g.* sodium sulphate), some organic compounds caused by lubricant oil used in the cutting process, and treatment chemicals (*e.g.* dissolved salts and water treatment chemicals) for the cooling-water system (Werner *et al.*, 1994). Thus, keeping the above points in view the present study was undertaken to assess the “Impact of Glass Industry Effluent on Seed Germination and Biochemical Parameter of *Glycine max* (Soyabean)”.

**MATERIALS AND METHODS**

**Description of sampling site:** The Sampling site selected for the present study was Hindustan glass Ltd located in IDPL, Virbhadra (Rishikesh) which is 29 km away from Kanya Gurukul campus Haridwar.

**Collection and storage of sample:** Effluent Samples were collected carefully with every possible precaution so as to reduce the possibility or any error during analysis and to prevent the result from being biased. The effluent samples were collected in the plastic jerry cans were thoroughly rinsed 5-6 times with distil water before taking samples.

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Preparation of pots/petri-plates for experimental analysis

Petri-plates: Seeds of Glycine max were surface sterilized with 0.1% of HgCl₂ solution for 2 minutes and washed with water. The known volume of 25, 50, 75 and 100% concentration of glass industry effluent was poured on the petri-plates containing 10 seeds of Glycine max placed equidistantly. Tap water served as control.

Pots: In the present study, seeds of Glycine max were surface sterilized with 0.1% of HgCl₂. Ten seeds of Glycine max were sown in each pot and irrigated with different concentration of effluent. The effect of different concentration of effluent was observed after 21 days of sowing i.e. percentage of seed germination, root length, shoot length, vigour index, and root: shoot ratio, chlorophyll a, chlorophyll b, total chlorophyll, ascorbic acid, fresh biomass and dry biomass.

Parameter analysed

Physico-chemical parameter of glass industry effluent and garden soil: Physico-chemical parameters of glass industry effluent and garden soil were analysed by using standard method described by APHA et al., (2012) and Trivedy and Goel, (1998). Parameters includes under my study are temperature, turbidity, TS, TDS, TSS, pH, conductivity, alkalinity, total hardness, calcium hardness, magnesium hardness, DO, BOD. The garden soil samples parameters were measured temperature, moisture content, WHC (water holding capacity), Porosity, pH, conductivity, alkalinity, organic matter and phosphate.

Plant parameters

Germination percentage: The number of seeds germinated in each Petri plates of different concentration was observed on 7th day. The total germination percentage was calculated by using the following formula:

\[ \text{Germination} \% = \frac{\text{Total no of seed germinated}}{\text{Total no. of seed sown}} \times 100 \]

Vigour index: For the estimation of vigour index the shoot length was measured from the cotyledonary region to the tip of apex. The root length was measured from the cotyledonary region to the tip of the primary root. Vigour index was computed using the following formula:

\[ \text{Vigour index} = \frac{\text{germination} \% \times \text{mean root length (cm)}}{\text{mean shoot length (cm)}} \]

Chlorophyll content: Numbers of studies have shown that the chlorophyll content can be taken as index of photosynthetic activity. The grassland plants are found to have chlorophyll 'a' and chlorophyll 'b' as principle photosynthetic pigments.

Biomass estimation by harvest method: In this method plants were harvested after 21 days. There fresh and dry weights were taken and calculated the biomass per unit area by using given formula.

\[ \text{Fresh weight} = X \text{ gm}; \text{Dry weight} = Y \text{ gm}; \text{Area of pot} A = \pi r^2; \text{Where, } r^2 \text{ is the radius of pot.} \]

Calculation

\[ \text{Fresh biomass} = \frac{x \times 100 \times \pi r^2}{\pi r^2} = X \text{ gm}^2 \]

\[ \text{Dry biomass} = Y \times 100 \times \frac{1}{\pi r^2} = Y \text{ gm}^2 \]

RESULTS AND DISCUSSION

The present study was conducted to assess the physico-chemical parameters of glass industry effluent and its effect on the seed germination and biochemical parameter of Glycine max Soyabean. Physico-chemical parameters of glass industry effluent are given in Table 1. Physico-chemical characteristics of garden soil are given in Table 2. Effect of glass industry effluent on biochemical parameter of Glycine max (soyabean) is presented in Table 3.

Temperature is basically important for its effect on the chemistry and biological reactions in the organisms in water. In the present study temperature in glass industry effluent recorded was 26.04±0.73 (Table 1). Our finding coincides with the finding of (Singh and John, 2010) in glass industries waste water in Firozabad. A rise in temperature of water leads to the speeding up of the chemical reactions in water, reduces the solubility of gases and amplifies the taste and odours. Turbidity in water is caused by the clay, silt, organic matter, phytoplankton and other microscopic materials.

Table 1: Physico-chemical characterisation of glass industry effluent. (Values are means±S.E of 7 observations each).

| Parameter | Mean ± S.E | Permissible Limits(IS) |
|-----------|------------|------------------------|
| Temperature °C | 26.04 ± 0.73 | - |
| Turbidity (NTU) | 25.3 ± 2.67 | - |
| Total solids (mg/l) | 542.85 ± 36.88 | 1200 |
| Total dissolved solids (mg/l) | 285.71 ± 40.40 | 1000 |
| Total suspended solids (mg/l) | 228.57 ± 52.16 | 200 |
| pH | 6.85 ± 0.18 | - |
| Conductivity (S-1/cm) | 0.275 ± 0.02 | 300 |
| Alkalinity (mg/l) | 290 ± 6.72 | 360 |
| Total hardness (mg/l) | 81.14 ± 3.43 | 600 |
| Calcium hardness (mg/l) | 25.63 ± 1.27 | 200 |
| Magnesium hardness (mg/l) | 3.91 ± 0.41 | 100 |
| Dissolved oxygen (mg/l) | 1.18 ± 0.14 | >6.0 |
| BOD (mg/l) | 119.18 ± 2.24 | 100 |

Table 2: Physico-chemical characterisation of garden soil. (values are mean ± S.E of 5 observations each).

| Parameter | Mean ± S.E |
|-----------|------------|
| Temperature °C | 21 ± 0.316 |
| Moisture content% | 9% ± 0.14 |
| Water holding capacity % | 43.3 ± 2.72 |
| Porosity (sec⁻¹) | 0.65 ± 0.290 |
| pH | 6.64 ± 0.021 |
| Conductivity (S⁻¹/cm) | 0.217 ± 0.002 |
| Alkalinity (meq/100gm) | 1.97 ± 0.019 |
| Organic matter % | 2.145 ± 0.312 |
| Phosphate (ppm) | 0.244 ± 0.008 |
organism. In the present study the turbidity in glass industry effluent recorded was 25.3±2.67NTU (Table 1). Turbidity increases due to heavy load of dissolved and suspended solid and organic matter released by effluent. Total Solids are determined as the residue left after evaporation of the unfiltered sample. In the present study total Solids in glass industry effluent recorded were 542.85±36.88 mg/l (Table 1). The obtained value was within the permissible limit prescribed by Indian Standard, 1982. Dissolved Solids refer to any minerals, salts, metals cations and anions in water. Total Dissolve solids comprise inorganic salts and some small amounts of organic matter that are dissolved in water. In the present study, Total Dissolved Solids in glass industry effluent recorded were 285.71±40.40 mg/l (Table 1). The obtained value was within the permissible limit prescribed by Indian Standard, 1982. The main constituents of TDS were calcium, sodium, magnesium, bicarbonates, chlorides, sulphates. The potability of water with a TDS less than 600 mg/L is generally considered to be good whereas at TDS levels greater than 1200 mg/L drinking water becomes increasingly unpalatable (WHO, 1984). TSS of a water or wastewater sample is determined by pouring a carefully measured volume of water through a pre-weighed filter of a specified pour size then weighing the filter again after drying to remove all water. In the present study, Total suspended solid in glass industry effluent recorded were 228.57±52.16 mg/l (Table 1), which was found to be slightly higher as compared to permissible limit prescribed by Indian Standard, 1982. Kumar et al. (2016) also recorded the TSS value in glass industry effluent with in a range 364.25 ± 6.95 while working on impact of glass industry effluent disposal on soil characteristics in Haridwar region, India. pH is the measure of intensity of acidity and alkalinity and measure the concentration of Hydrogen ions in water. pH is the most important key factor to determine the water quality. In the present study pH in glass industry effluent recorded was 6.85±0.18 (Table 1) which was slightly acidic in nature. The result of present study coincides with the result of Singh and John (2010) in glass industry effluent of Firozabad (U.P) they recorded the pH of waste water was ranged between 4.2-7.4. Conductivity is the measure of capacity of a substance or solution to conduct electric current. As most of the salts in the water are present in the ionic form, capable of conducting electric current therefore conductivity is good and rapid measure of the total dissolved solids. In the present study, conductivity in glass industry effluent recorded was 0.275±0.02 seimens/cm (Table 1), which was found to be within the limit as compared to permissible Limit prescribed by IS, 1982. Kumar et al. (2016) also recorded the conductivity value in glass industry effluent with in a range 1.34 ± 0.19. The Total alkalinity of water is measure of the capacity to neutralize a strong acid. The alkalinity in water is generally imparted by the salts of carbonates, bicarbonates, phosphate and nitrate and other with hydroxyl ion in Free State. However, most of the water are rich in carbonates and bicarbonates. In the present study total alkalinity in glass industry effluent recorded was 290±6.72 mg/l (Table 1), which was found to be within the limit as compared to permissible limit prescribed by IS, 1982. Hardness in water is due to natural accumulation of salts from content with soil and geological formation or it may enter from direct pollution by industrial effluents. Hardness is temporary if it is associated with mainly carbonates and bicarbonates and permanent if associated with sulphate and chlorides. In the present study, total hardness in glass industry effluent recorded was 81.14±3.43 mg/l (Table 1) which was found to be within the permissible limit prescribed by Indian standard, 1982. Water with hardness above 200mg/L may cause scale deposition in the water distribution system and more soap consumption. In the present study, calcium hardness in glass industry effluent recorded was 25.63±1.27 mg/l (Table 1) which was found to be within the permissible limit prescribed by Indian standard, 1982. Calcium and Magnesium form a complex of wine red colour with Eriochrome Black T at pH 10.0. The EDTA has got a stronger affinity for Ca²⁺ and M⁺⁺; the former complex is

| Plant parameters | Control | Glass industry effluent |
|------------------|---------|-------------------------|
| % Germination    | 96      | 97                      |
| Root length(cm)  | 21.70±0.69 | 22.24±0.54              |
| Shoot length(cm) | 18.31±0.47 | 18.40±0.35              |
| Vigour index(cm) | 3840.9  | 3942                    |
| Root shoot ratio | 1.185   | 1.208                   |
| Chlorophyll 'a' (mg/g) | 0.365±0.140 | 0.3169±0.001          |
| Chlorophyll 'b' (mg/g) | 0.211±0.005 | 0.273±0.05             |
| Total chlorophyll (mg/g) | 0.576±0.06 | 0.642±0.05             |
| Ascorbic acid    | 0.128±0.03 | 0.168±0.17              |
| Fresh biomass (gm/m²) | 383.1  | 453.5                   |
| Dry biomass (gm/m²) | 76.43 | 77.38                   |

Table 3: Effect of glass industry effluent on seed germination and biochemical parameters of Glycine max. (Values are mean ± S.E of 3 observations each).
Organic matter is a very complex part of soil. It is the soluble salts which are present to some extent in all soil. The value of M+ can be obtained by subtracting the value of Calcium from the total of Ca++ + Mg++. In the present study, magnesium hardness recorded was 3.91±0.41 mg/l (Table 1) which was found to be within the permissible limit prescribed by Indian standard, 1982. Dissolved oxygen is one of the most factor in water quality assessment and reflects physical and biological process prevailing in to water, its presence is essential to maintain the higher form of biological life in water. Generally the oxygen concentration is associated with heavy contamination of organic matter. The solubility of dissolved oxygen increased with decreased level of the water temperature. In the present study DO recorded was 1.18±0.14 mg/l (Table1), which was found to be below as to compared the permissible limit (>6.0 mg/l) prescribed by Indian Standard, 1982. Kumar et al. (2016) reported the nil DO in the glass industry effluent of Jhabrera (Uttarakhand). It was due to the high pollution load, high organic matter, microbial activity and oxygen demanding waste. A measure of the oxygen required to oxidize the desirable organic matter in a water sample to stable the inorganic compounds and gives an approximate index of organic pollution. Biochemical oxidation is brought about by microorganisms that utilize organic matter as a source of carbon and in doing to consume dissolved oxygen. In the present study, the observed mean value of BOD in glass industry effluent was 119.18±2.24 mg/l (Table 2). The data was found to be above as compared to the permissible limit (100 mg/l) prescribed by Indian Standard, 1982. It was due to presence of high amount of organic matter.

The soil temperature is an important because it affects the physical, chemical and biological process it affects the rate of absorption of water, solutes and activities of microorganism. The thermometer gives the reading directly in °C. During the present study, the recorded value of soil temperature was 21±0.316°C (Table 2). Moisture content is important for maintaining habitat of plants and animals. In the present study, the recorded value of soil moisture content was 9%±0.14 (Table 2). Water holding capacity may be defined as the total amount of water stored or absorbed by soil. It is significantly as the tendency of soil to absorb water depends upon retention of water by given sample of soil. In the present study, the recorded value of water holding capacity was 43.3±2.73% (Table 2). Soil porosity depends upon texture of soil and particles size. Different kinds of soil have different composition and their porosity also differs. Porosity increases with increase in organic matter content. In the present study, the recorded value of soil porosity was 0.65±0.290 sec⁻¹ (Table 2), pH of soil is the measure of the hydrogen ion activity which depend largely on relative amount of absorbed hydrogen and metallic ions. In the present study, the recorded value of soil pH was 6.64±0.021 (Table 2). Electrical conductivity of soil is related to the soluble salts which are present to some extent in all soil.

The main sources of salts are mineral. Conductivity of soil is also related to the temperature of soil. In the present study, the recorded value of soil conductivity was 0.217±0.0021seimems⁻³/cm (Table 2). Alkalinity shows the alkaline nature of soil. In the present study, the recorded value of alkalinity in garden soil was 1.97±0.019 meq/100gm (Table 2). Organic matter is very complex part of soil. It is made by the root and decomposition of plants, animal’s manure and dead animals. Organic matter provides much of needed nitrogen, phosphorus and sulphur to chemical action in the soil. In the present study, the recorded value of organic matter in garden soil was 2.145±0.312% (Table 2). Phosphorus is an essential component of organic compound. Both organic and inorganic form of phosphorus are essential for plant growth but their availabilities are controlled by soil characteristics and environmental condition. In the present study, the recorded value of phosphorous as 0.244±0.008 ppm (Table 2). Effect of glass industry effluent on the growth of Glycine max after 21 days was shown in Table 3. Maximum percentage of seed germination i.e. 97% was recorded in 25% concentration of effluent (Fig 1). This promotion was possibly due to the presence of optimum level of plant nutrient in the lower concentration of effluent. The soil temperature is an important because it affects the physical, chemical and biological process it affects the rate of absorption of water, solutes and activities of microorganism. The thermometer gives the reading directly in °C. During the present study, the recorded value of soil temperature was 21±0.316°C (Table 2). Moisture content is important for maintaining habitat of plants and animals. In the present study, the recorded value of soil moisture content was 9%±0.14 (Table 2). Water holding capacity may be defined as the total amount of water stored or absorbed by soil. It is significantly as the tendency of soil to absorb water depends upon retention of water by given sample of soil. In the present study, the recorded value of water holding capacity was 43.3±2.73% (Table 2). Soil porosity depends upon texture of soil and particles size. Different kinds of soil have different composition and their porosity also differs. Porosity increases with increase in organic matter content. In the present study, the recorded value of soil porosity was 0.65±0.290 sec⁻¹ (Table 2), pH of soil is the measure of the hydrogen ion activity which depend largely on relative amount of absorbed hydrogen and metallic ions. In the present study, the recorded value of soil pH was 6.64±0.021 (Table 2). Electrical conductivity of soil is related to the soluble salts which are present to some extent in all soil.

Maximum root length and shoot length of the Glycine max was observed at 25% concentration of effluent i.e. 22.24±0.54 cm and 18.40 ±0.35 cm (Table 3) it was may be due to presence of essential nutrient in diluted lower concentration of effluent which promote the growth of plant. As increase in the effluent concentration (>25%) there was gradual decline in root length and shoot length. While the minimum root length and shoot length of the Glycine max was observed at 100% concentration of glass industry effluent was 12.44±0.97 cm and 15.66±0.69 cm (Table 2). It was due to presence of high amount of solid and toxic elements. Vigour index of Glycine max was found maximum at 25% concentration of effluent i.e. 3942 (Table 3) which was due to presence of optimum level of nutrients. While the minimum vigour index of Glycine max was observed at 100% concentration of glass industry effluent was 1395 (Table 3). This was due to the toxicity of effluent, Chlorophyll ‘a’ of Glycine max was observed maximum at 25% concentration of glass industry effluent i.e. 0.369±0.001 mg/g (Table 3) while the minimum chlorophyll ‘a’ of glycine max was observed at 100% concentration of effluent was 0.118±0.004 (Table 3). Chlorophyll ‘b’ of Glycine max was observed maximum at 25% concentration of glass industry effluent i.e. 0.273±0.05 mg/g (Table 3). While the minimum chlorophyll ‘b’ of Glycine max was observed at 100% concentration of effluent was 0.101±0.001 mg/g (Table 3). Chlorophyll is the pigment which is essential for photosynthesis. More is the chlorophyll content more of the foliage and thus more of the productivity. During the present study chlorophyll content was increase from 25% to 75% concentration of effluent. The maximum chlorophyll content
i.e. 0.642±0.16 mg/gm (Table 3) was reported in 25% effluent concentration due to presence of optimum level of micro-nutrients, phosphorus, nitrogen, calcium and magnesium level in the effluent responsible for the better chlorophyll content. While the minimum total chlorophyll content of Glycine max was observed in 100% concentration of effluent was 0.219±0.002 (Table 3). Ascobic acid is regarded as an antiphytoxican, it influences resistance against adverse environment condition. The high ascobic acid levels signify greater plant tolerance towards stress. In the present study, the maximum ascorbic acid content i.e. 0.168±0.017 mg/g (Table 3) recorded was in 25% effluent concentration due to the micro-nutrients present in optimum level in effluent. While the minimum ascorbic acid of Glycine max was observed at 100% concentration of effluent was 0.088±0.010 mg/g (Table 3). Fresh biomass of Glycine max was observed maximum at 25% concentration of glass industry effluent i.e. 453.5 gm/m² (Table 3) due to optimum nutrient level present in diluted higher concentration. While the minimum fresh biomass of Glycine max was observed at 100% concentration of effluent was 169.4gm/m²(Table 3). Dry biomass of Glycine max was observed maximum at 25% concentration of effluent i.e. 77.38 gm/m² (Table 3) due to optimum nutrient level present in diluted lower concentration. While the minimum dry biomass of Glycine max was observed at 100% concentration of effluent was 39.80gm/m² (Table 3).

CONCLUSION

The present study was conducted to analyse the physico-chemical parameters of glass industry effluent and its effect on the biochemical parameters of Glycine max (soyabean). The study was conducted from the month of January to March. The result of present study revealed that pH value of glass industry was acidic due to decomposition of organic matter, presence of high amount of carbonates and bicarbonates. Temperature, turbidity, total solid, total dissolved solid, alkalinity and total hardness of glass industry effluent were recorded with in the permissible limit prescribed by Indian Standard, 1982.

Total suspended solid, DO and BOD was recorded above the permissible limit prescribed by Indian standard, 1982. DO of glass industry effluent was very low because of presence of high amount of organic material. BOD was recorded above the permissible limits prescribed by Indian Standard, 1982. This was due to high pollution load, huge amount of organic material and many more toxic chemicals. Such high levels of BOD indicative of relatively high proportion matter, that causes depletion of dissolved oxygen in water, are known to be a threat of aquatic life. In the present study the garden soil pH was acidic in nature. Moisture content, water holding capacity, porosity, organic matter, phosphorous, conductivity, and alkalinity was low in garden soil. In the present study seed germination, root length, shoot length, vigour index, root shoot ratio, chlorophyll content and ascobic acid were recorded maximum in 25% effluent concentration due to optimum level of nutrient present in the glass industry effluent. Thus, the physico-chemical study of glass industry effluent suggests that effluent contains higher amounts of nutrients which increases crop yield substantially and reduce the need for fertilizer, and ultimately decreases overall cost of production.

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