Chapter 18
An Overview of Effective Concentration of Industrial Effluent for Improving Crop Production and Its Effect on Micro-Biodiversity Zone of Soil

Sangeeta, Gita Rani, and Rani Devi

Abstract Soil and water are integral parts of the ecosystem. These are used as resources for agriculture. In recent years, most of the water and soil have become polluted by sewage, industrial waste/effluents, and a wide range of synthetic chemicals. Our planet Earth is now overburdened with the toxic substances. Industrialization has been proven as a significant milestone in the development of human civilization. But, at the same time, it has loaded the Earth with industrial wastes including toxic solids, liquids, and gaseous discharges. With the mushrooming growth pattern of industries, all important components of ecosystems particularly soil, water, air, vegetation, and all others are being affected adversely. Management of these natural resources is very important for sustainable development of living beings on Earth. Industrializations are not only land area intensive but also lead to other serious environmental humiliation (Azumi DS, Bichi MH, J Appl Sci Environ Sanit 5:23–29, 2010). Now the challenge before us in this present scenario is the careful disposal of effluent so that adverse impact on soil fertility, plant growth, and health of animals and human beings may be reduced. Thus, there is an urgent need to have innovative strategies for wiser management of effluents and land resources for its optimum and appropriate utilization. In the present study, efforts have been made for handling sugar industry along with use of its effluent for improving wheat crop and production.

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18.1 Introduction: Sugarcane Industry

Cultivation of sugarcane in India dates back to the Vedic period. Various historical and mythological evidences confirm that the original home of sugarcane production and sugar manufacturing is India. It is cheaper to get sugar from sugarcane than sugar beet. Sugarcane is cultivated in tropical climates. Globally, India is the largest producer of sugar (Solomon 2008). India stands as a major sugar-producing nation in the world, having 579 sugar mills and 319 distilleries (Patil and Gholey 2010), and is contributing around 18–20% of the world's cane sugar production and sugar potential in different states of India. Sugar industry is the backbone of all food industries, and sugar produced in it is the important ingredient for food preparation at home, functions, and parties and in marriages. It is one of the most important agro-based industries in India and is the second largest in the country, next to textiles. But a considerable amount of effluent is released during sugarcane crushing which is spilled into nearby water bodies and is causing a serious problem to water bodies and land/soil in its vicinity which is resulting in environmental degradation and affecting the crop quality as well as yield (Maliwal et al. 2004). It is also generating a huge amount of solid waste as bagasse. To handle this problem, the industry is using bagasse to produce ethanol, which can be used as an eco-friendly and renewable energy source by blending with petrol (Satheeskumar and Selvaraj 2007). The production of ethanol gives the boost to agriculture sector and reduces environmental pollution. The Government of India is trying to introduce supply of ethanol-doped petrol in the country.

18.2 Sugar Industry Effluents

All the industries consume huge quantity of water and throw back almost 70–80% of it as effluents which contain highly toxic materials in dissolved or suspended form. The demand for water is increasing due to population growth and urbanization, hence making this resource very scarce (CPCB 2009). Main wastewater pollutants are organic components (proteins, fats, and carbohydrates), inorganic components (alkalies, acids, inorganic salts, and other chemicals), and physical factors such as turbidity, color, temperature of effluent, radioactivity, etc. Presence of these organic compounds imparts a high biochemical oxygen demand (BOD) and chemical oxygen demand (COD) load to the liquid waste, and their level below a certain limit becomes a danger to aquatic life (Sharma and Habib 1997; Singh et al. 2005; Pandey et al. 2008; Chandra et al. 2009).

Application of sewage and industrial wastewater on agricultural land (irrigation) became an alternative water supply to crops as well as an alternative waste disposal method. Sugar industry wastes constitute a number of physicochemical effluents of suspended and dissolved solids with the high amount of BOD, COD, chlorides, sulfates, calcium, magnesium, nitrates, etc. The high contents of organic matter in

stillage cause surface water pollution. Organic matters cause oxygen depletion in surface waters by biodegradation. Stillage has high concentration of potassium which can cause pollution of the soil. When released into the environment without proper treatment, effluents of sugar industry produce unpleasant color and odor. These effluents also alter the physicochemical characteristics of the receiving aquatic bodies and affect aquatic flora and fauna (Baruah et al. 1993). The polluted soil becomes unsuitable for further cultivation. So, some treatments should be given to sugar mill effluent-polluted soils before they are used for crop cultivation (Roy et al. 2007).

18.3 Wheat Cultivation

Wheat is the staple food of millions of Indians, particularly in North India, and the 2nd most important food grain of India. India is the fourth largest producer of wheat in the world and accounts for 8.7% of the world's total production of wheat. It is a rabi crop sown in the beginning of winter and is harvested in the beginning of summer.

The **Meham Co-Operative Sugar Mills Ltd., Rohtak, Haryana**, chosen for this study, is located at 28°59′49.2″N 76°14′30.1″E. The study area falls in the eastern zone, which covers around 49% of the area of the state. This zone is also called wet zone.

18.4 Microbial Species in Soil

18.4.1 Identification and Characterization of Fungal Isolates

Identification and characterization of fungal isolates in the soil affected with SME (sugar mill effluent) were carried out on the basis of colony growth (diameter), presence or absence of aerial mycelium, colony color, presence of furrows in the medium, pigment production, spore morphology, etc. The fungal species (lactophenol cotton blue preparation) were identified by microscopic analysis using taxonomic guides and standard procedure (Barnett and Hunter 1972; Ellis 1976; Domsch et al. 1980; Nelson et al. 1983; Gilman 1998).

The major cultural features used for identification included color of the colony, growth pattern, mycelial structure, spore-bearing structure, and spore morphology. The identified species are the following.

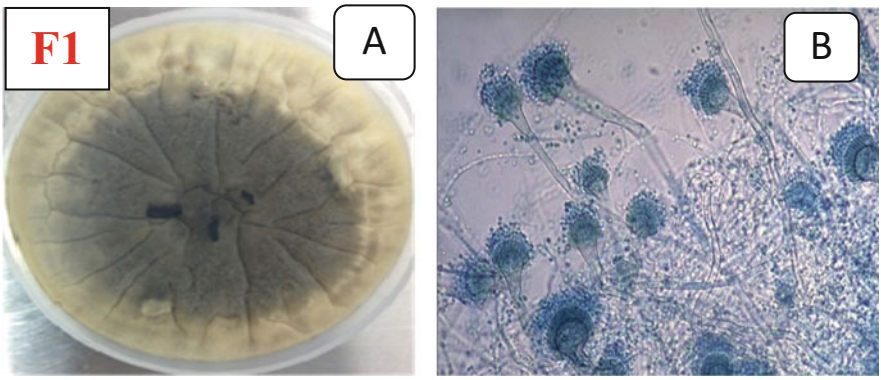

Fig. 18.1 First identified isolated fungus *Aspergillus* sp. F1 (**a**) showing growth on agar plates and (**b**) morphological observations under the microscope (100×)

18.4.1.1 First Identified Fungus F1

The first identified fungus strain was *Aspergillus* sp. F1 (Fig. 18.1a, b), which was identified on the basis of colony morphology on PDA plates and microscopic features. *Aspergillus* sp. is a filamentous fungus (mold) commonly reported from indoor environment that is used in different value added properties like food and enzyme industries. Various species of *Aspergillus* are used in the industrial preparation of citric acid, gluconic acid, and many other products of commercial importance. *Aspergillus* sp. can be isolated from many different ecological habitats such as soil, plant debris, and rotting fruits. This fungus is characterized by a round vesicle with extending conidial chains, which appear as white and fluffy strands on the substrate that the fungus inhabits. Taxonomic as well as identification points are given below.

Taxonomic Classification

- Kingdom: Fungi
- Division: *Ascomycota*
- Class: *Eurotiomycetes*
- Order: *Eurotiales*
- Family: *Trichocomaceae*
- Genus: *Aspergillus*

Identification Features

- Colonies grow very quickly.
- Colony produced is with yellow to white hyphae, turning black with the formation of conidia. Hyphae are septate.
- Conidiophores are long and globose at the tip.
- Spores are globose with conspicuous ridges or spines and not arranged in rows.

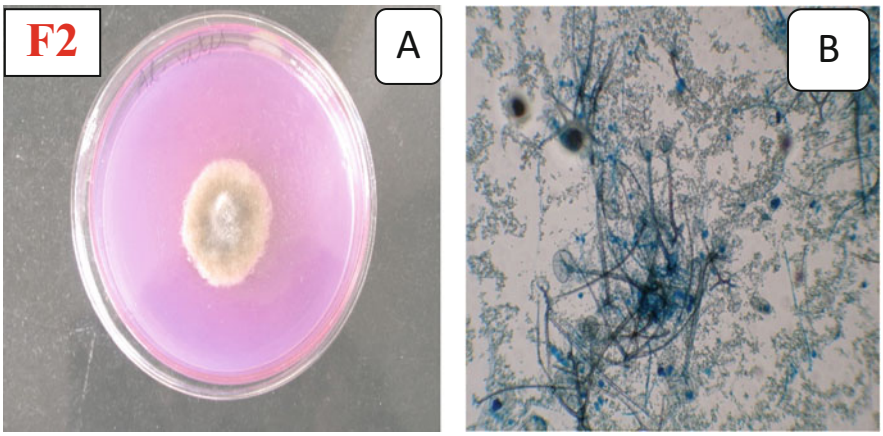

Fig. 18.2 Second and third identified isolated fungus *Rhizopus* sp. F2 (**a**) showing growth on agar plates and (**b**) morphological observations under the microscope (100×)

- Colony diameter was found to be 10–12 mm in size after 4 days of incubation on PDA plates.
- Conidia slightly roughened or finely echinulate.

18.4.1.2 Second Identified Fungus F2

The second identified fungus was *Rhizopus* sp. F2 (Fig. 18.2a, b), which is a cosmopolitan filamentous fungus frequently isolated from soil, decaying fruits and vegetables, animal feces, and old bread. Aside from being known as common contaminants, *Rhizopus* species are also occasional causes of serious, and often fatal, infections in humans. Certain species are plant pathogens as well.

Taxonomic Classification

- Kingdom: Fungi
- Phylum: *Zygomycota*
- Order: *Mucorales*
- Family: *Mucoraceae*
- Genus: *Rhizopus*

Identification Features

- Colonies spread rapidly at 25 °C, about 5–8 mm high.
- Apophysis, rhizoids, and stolons are present.
- Mycelium shows white in color at starting stage, and after that becomes dark brown or blackish brown to black in color.

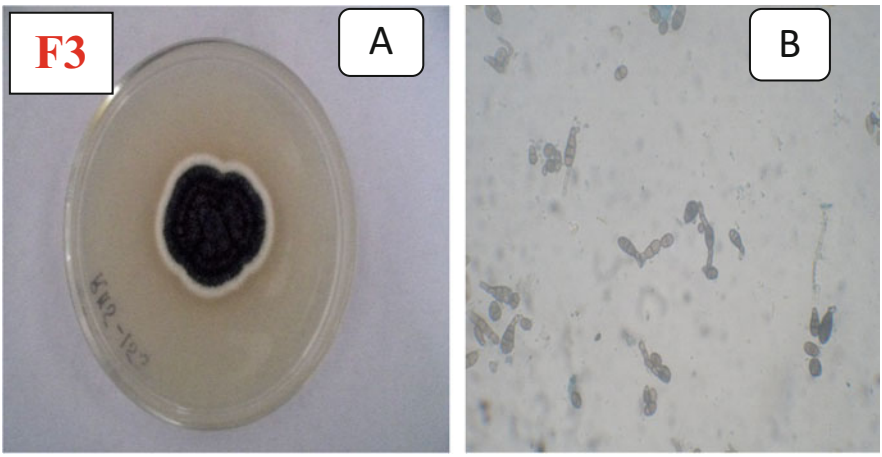

Fig. 18.3 Third identified isolated fungus *Alternaria* sp. F3 (**a**) showing growth on agar plates and (**b**) morphological observations under the microscope (100×)

18.4.1.3 Third Identified Fungus F3

Alternaria sp. F3 (Fig. 18.3a, b) is an ascomycetous fungus and is known as a major plant pathogen causing diseases in 380 host species of plant. They are also common allergens in humans, growing indoors and causing hay fever or hypersensitivity reactions. At least 20% of agricultural spoilage is caused by *Alternaria* species; the most severe losses may reach up to 80% of yield, though many human health disorders can be caused by these fungi, which grow on skin and mucous membranes, including on the eyeballs and within the respiratory tract. It is an opportunistic pathogen on numerous hosts causing leaf spots, rots, and blights on many plant parts. As a result, this pathogen propagates itself via asexual spores called conidia.
Taxonomic Classification

- Kingdom: Fungi
- Phylum: *Ascomycota*
- Class: *Dothideomycetes*
- Order: *Pleosporales*
- Family: *Pleosoporaceae*
- Genus: *Alternaria*

Identification Features

- Colonies are slow growing with whitish margin and gray to light brown in color.
- Conidiophores arise singly or in groups, usually simple, erect, straight, or curved.
- Conidiophores are occasionally geniculate, more or less cylindrical but often slightly rounded at the base.

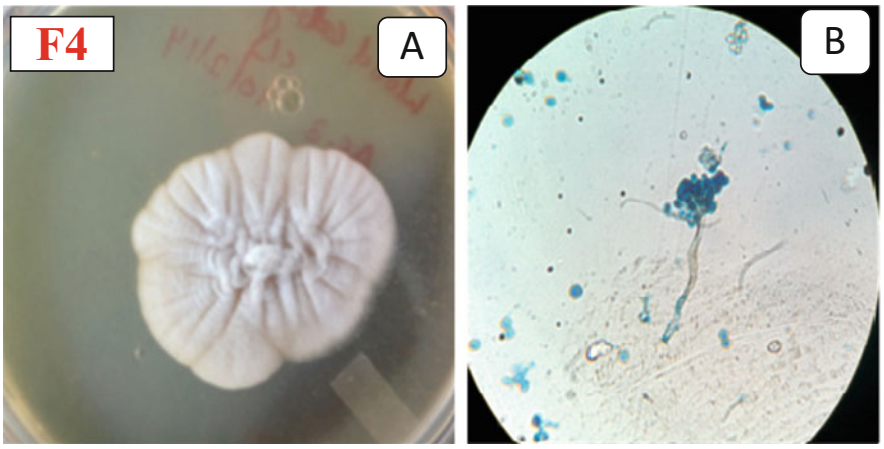

Fig. 18.4 Unidentified fungus F4 (**a**) showing growth on agar plates and (**b**) morphological observations under the microscope (100×)

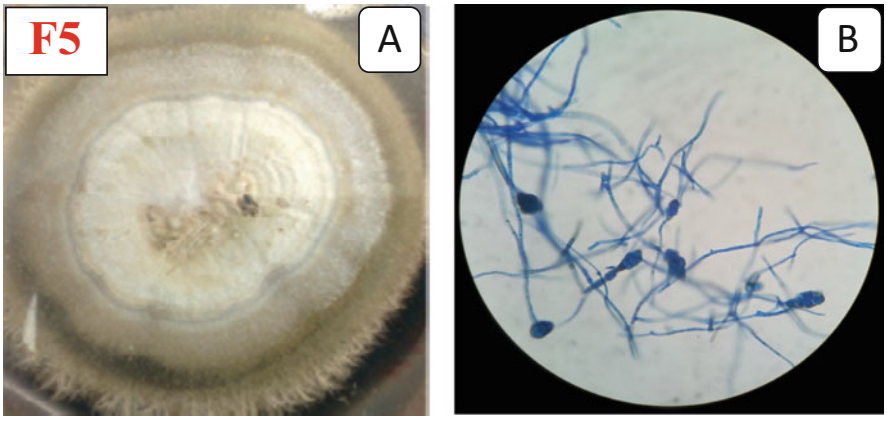

Fig. 18.5 Unidentified fungus F5 (**a**) showing growth on agar plates and (**b**) morphological observations under the microscope (100×)

- Conidia are pale brown to light brown, obclavate to obpyriform or ellipsoid, short conical beak at the tip, or beakless. Conidia are septate.

18.4.1.4 Unidentified Fungal Isolates F4 and F5

Two unidentified fungal isolates F4 and F5 (Figs. 18.4a, b and 18.5a, b) were obtained.

18.4.2 Identification and Characterization of Bacterial Isolates

The identification of bacterial isolates is based on many factors, including cell and colony morphology, chemical composition of cell walls, biochemical activities, colony growth (diameter), colony color, pigment production, nutritional requirements, etc. The bacterial species were identified by microscopic analysis using taxonomic guides and standard procedure (Barnett and Hunter 1972; Ellis 1976; Domsch et al. 1980; Nelson et al. 1983; Gilman 1998). From the samples only two bacterial isolates were identified on the basis of the following characteristics.

Gram Staining An initial step in identifying a bacterial species is determining if it is Gram-positive or Gram-negative. Gram staining is one of the most widely used tools in the identification of bacteria. Gram-negative cells appear pink after the Gram staining procedure, which enables comparison between those cells that decolorize with ethanol and those which do not.

Morphological Characteristics After the Gram staining procedure, microorganisms are also classified according to colony morphology and cell morphology. Bacterial colonies have different characteristics like size, form or shape, edge, texture, degree of opacity, and color. These characteristics describe the morphology of a single colony and may be useful in the preliminary identification of a bacterial species.

Identification of Samples On the basis of Gram staining, two isolates were found including one which was Gram-positive and the other which was Gram-negative.

18.4.2.1 First Identified Bacterial Isolate B1

The first isolated bacterial strain was identified as *Bacillus* sp. B1 (Fig. 18.6a, b).

Taxonomic Classification

- Domain: Bacteria
- Division: *Firmicutes*
- Class: *Bacilli*
- Order: *Bacillales*
- Family: *Bacillaceae*
- Genus: *Bacillus*

Identification Features

- Gram staining: Positive bacteria showing purple color cells after staining
- Shape: Rod-shaped cells

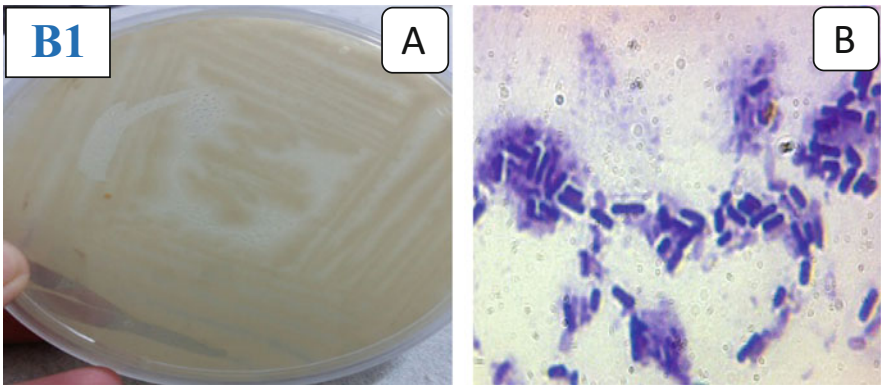

Fig. 18.6 First identified bacterial isolate *Bacillus* sp. B1, (**a**) showing growth on agar plates and (**b**) morphological features under the microscope (100×)

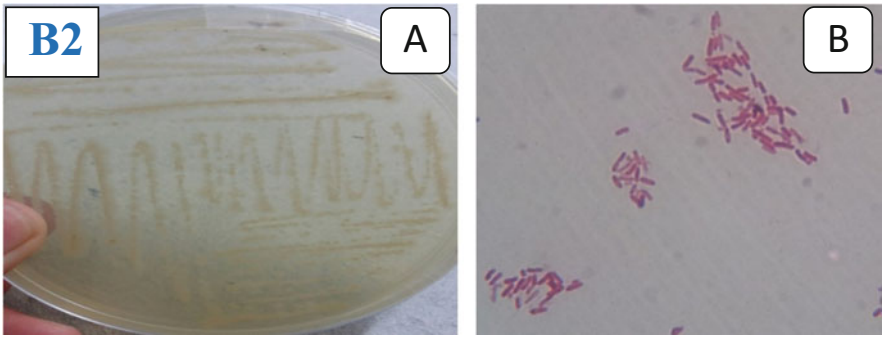

Fig. 18.7 Second identified isolated bacterial strain *Escherichia coli B*2 (**a**) showing growth on agar plates and (**b**) morphological features under the microscope (100×)

18.4.2.2 Second Identified Bacterial Isolate B2

The second identified bacterial strain was *Escherichia coli* (Fig. 18.7a, b). These are Gram-negative and rod-shaped bacteria.

Taxonomic Classification

- Domain: Bacteria
- Phylum: *Proteobacteria*
- Class: *Gammaproteobacteria*
- Order: *Enterobacteriales*
- Family: *Enterobacteriaceae*
- Genus: *Escherichia*

Box 18.1 Dilution-concentration pattern of SME with water

Water	Blank water (1: 0)	Wastewater with dilution (1:3)	Wastewater with dilution (1:1)	Wastewater wastewater (0:1)
Soil				
Soil irrigated with blank water	S1 W1 (100%)	S1 W2	S1 W3	S1 W4
Soil irrigated with wastewater with dilution	S2 W1	S2 W2	S2 W3	S2 W4
Soil irrigated with wastewater with dilution	S3 W1	S3 W2	S3 W3	S3 W4
Soil irrigated with undiluted wastewater	S4 W1	S4 W2	S4 W3	S4 W4 (100%)

Factor water was in the ratio given below:
W1: Irrigation with fresh water or BWW containing 100% blank water (1: 0)
W2: Irrigation with mixture containing 75% BWW and 25% wastewater (dilution ratio 1:3)
W3: Irrigation with mixture containing 50% BWW and 50% wastewater (dilution ratio 1:1)
W4: Irrigation with undiluted wastewater containing 0% blank water (0:1)
Factor soil was in the ratio given below:
S1: Soil of fields containing 100% blank water (1: 0)
S2: Soil watered with BWW and sugar mill effluents (3: 1)
S3: Soil watered with BWW and sugar mill effluents (1: 1)
S4: Soil watered with sugar mill effluents containing 0% blank water (0:1)

Identification Features

- Gram-negative bacteria.
- Slow-growing colonies.
- Conidiophores arising singly or in groups.
- Colonies are growing in cottony appearance.
- Colonies are light brown in color.

Isolated Microorganisms

Three fungal (*Aspergillus* sp., *Alternaria* sp., *Rhizopus* sp.) and two bacterial (*Bacillus* sp. and *Escherichia coli*) cultures have been identified on the basis of colony morphology observed on agar plates followed by microscopic features in both effluent and affected soils. However, two fungal isolates were not identified on the basis of morphological features. Moreover, all these microorganisms can be further identified on the basis of biochemical and molecular features.

18.5 Sugar Mill Effluent and Wheat Growth

Effect of effluent on wheat germination and its growth were carried out according to the following dilution-concentration pattern of SME (Box 18.1).

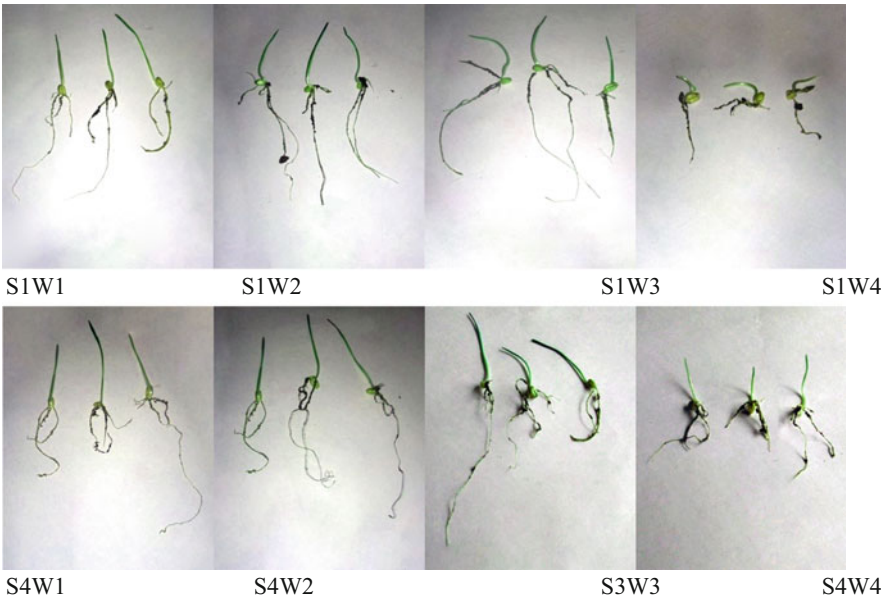

Fig. 18.8 Germination pattern of wheat plant irrigated with different concentrations (0%, 25%, 50%, and 100%) of sugar mill effluents

18.5.1 Germination of Seeds

Wheat seeds were shown in experimental pots. The pattern of germination was noted after 10 days by carefully pulling out the seedlings from each pot, and the number of roots was also counted in the seedlings. Seedling pattern of root growth and shoot growth is shown for various samples at different concentrations of sugar mill effluents (Fig. 18.8).

Wheat growth had been assessed before harvesting and after harvesting. Parameters analyzed before harvesting included germination percentage, speed of germination, peak value, vigor index, number of roots in seedling, root and shoot lengths, and fresh and total dry masses of wheat seedlings and after harvesting included plant height, length of the spike, number of spikes per plant and spikelet per spike, grain yield, straw yield, and biological yield.

18.5.2 Germination Percentage

With increase in effluent concentration, there is reduction in germination percentage of the wheat seeds but good germination rate at 50% concentration of effluents even better than with control (Table 18.1).

Table 18.1 Germination parameters of various wheat samples with different concentrations (0%, 25%, 50%, and 100%) of sugar mill effluents (mean ± SD of three values)

Samples	Germination (%)	Speed of germination
S1 W1	84.7 ± 3.85	7.21 ± 0.85
S1 W2	85.43 ± 3.85	7.58 ± 0.75
S1 W3	93.66 ± 6.67	8.21 ± 0.57
S1 W4	21.10 ± 3.85	2.16 ± 0.35
S2 W1	84.86 ± 0.00	7.98 ± 0.45
S2 W2	89.91 ± 6.67	8.54 ± 0.52
S2 W3	94.44 ± 3.85	9.21 ± 0.63
S2 W4	24.576 ± 3.85	2.11 ± 0.28
S3 W1	87.77 ± 3.85	7.59 ± 0.42
S3 W2	89.2 ± 3.85	7.23 ± 0.42
S3 W3	95.44 ± 3.85	9.34 ± 0.27
S3 W4	23.66 ± 0.00	1.89 ± 0.72
S4 W1	21.66 ± 3.85	1.21 ± 0.46
S4 W2	20.98 ± 3.85	1.29 ± 0.28
S4 W3	11.1 ± 3.85	1.01 ± 0.52
S4 W4	10.99 ± 3.85	1.11 ± 0.41

18.5.3 Speed of Germination

A comparative account of speed of germination of various wheat samples containing different ratios of sugar mill effluents (Fig. 18.8) shows clearly that speed of germination was maximum for samples S3 W3 and S2 W3, i.e., effluent concentration is beneficial up to value of 50%, and after this concentration, there is reduction in germination speed of the wheat crop.

The comparative seed germination (Fig. 18.9) represents maximum speed in green lines and minimum indicated by red lines showing inhibitory effect of effluents. Samples with 100% concentration of SME showed almost negligible germination marked by red line, and green line represents highest value of these parameters indicating usefulness of diluted SME in agriculture for better yield of crops. The seeds germinated were noted on each and every day after sowing, and then the percentage of germination was calculated. Although the seeds germinated in the samples containing S4 W4 showed least germination even at a later stage as compared to the pots irrigated with diluted effluents and containing soil prepared by irrigating with diluted effluents, still the number of seeds germinated was counted on each and every day and represented in tabular form.

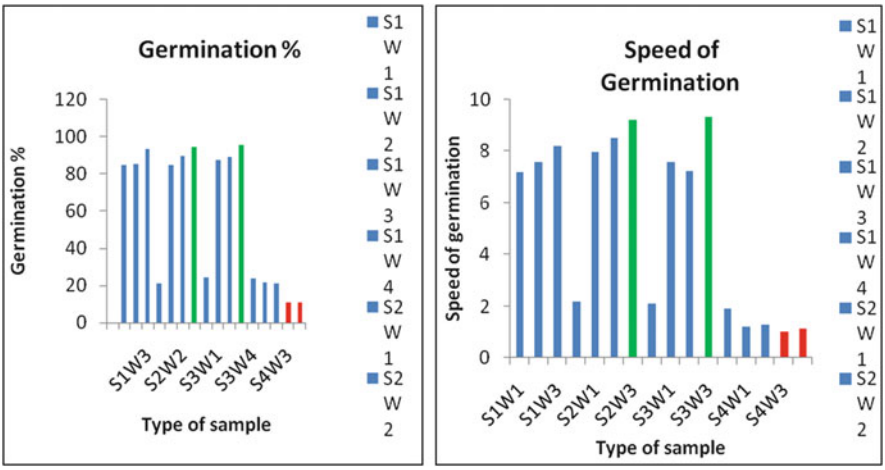

Fig. 18.9 Graph showing percentage of seed germination and speed/rate (%) of germination of various samples of wheat under different treatment conditions

18.5.4 Morphological Parameters of Wheat

18.5.4.1 Root Length (RL) and Shoot Length (SL)

The root and shoot lengths of seedlings were determined by using a scale of 10 days after sowing (DAS). The sum total of both (root and shoot) gives us the length of seedlings. The values of RL and SL of all the samples of different effluent concentrations (Tables 18.2, 18.3, and 18.4; Figs. 18.10, 18.11, and 18.12) showed clearly that the values are lowest for S4 W3 and S4 W4 and highest for S2 W3, S3 W2, and S3 W3. This is also in conformity with earlier findings that low concentration of effluents has positive effect on crops.

18.5.4.2 Number of Roots

A mature wheat plant has two distinct root types. The seminal roots are the first root types and nodal roots appear thereafter. The seminal roots are formed from the seed and nodal roots from the nodes and are related with the growth of tillers. The number of roots in various samples of different effluent concentrations is compared (Table 18.2, Fig. 18.10). It had been found that the number of roots of seedlings was maximum for S3 W3 and S2 W3 and minimum for S4 W4 and S4 W3, respectively, when compared with different treatment conditions.

Table 18.2 Shoot length, root length, number of roots, and vigor index of various samples of different effluent concentrations (mean ± SD of six values)

Samples	Shoot length (cm)	Root length (cm)	Number of roots	Vigor index
S1 W1	7.92 ± 0.3	6.65 ± 0.086	3.33 ± 0.57	12,340.7 ± 487.14
S1 W2	7.81 ± 0.15	6.51 ± 0.04	4.33 ± 0.57	12,233.5 ± 475.33
S1 W3	8.11 ± 0.19	7.11 ± 0.05	4.33 ± 0.57	14,255.0 ± 890.66
S1 W4	3.52 ± 0.079	3.08 ± 0.038	1.66 ± 0.5	1392.6 ± 44.9
S2 W1	8.41 ± 0.1	7.12 ± 0.029	4 ± 0.5	13,178.7 ± 580
S2 W2	8.45 ± 0.12	7.1 ± 0.033	5.33 ± 0.57	13,981.0 ± 914.77
S2 W3	8.92 ± 0.06	7.25 ± 0.034	5.66 ± 0.57	15,270.9 ± 534.41
S2 W4	2.79 ± 0.11	1.43 ± 0.025	2.33 ± 0.5	1037.1 ± 47.48
S3 W1	8.5 ± 0.074	7.28 ± 0.038	4.66 ± 0.57	13,850.1 ± 529.24
S3 W2	8.69 ± 0.07	8.47 ± 0.043	5.33 ± 0.57	15,306.7 ± 537.68
S3 W3	8.99 ± 0.041	8.92 ± 0.033	5.66 ± 0.57	17,093.3 ± 553.95
S3 W4	3.82 ± 0.065	2.08 ± 0.042	1.33 ± 0.57	1395.9 ± 123.12
S4 W1	2.21 ± 0.047	1.86 ± 0.058	1.33 ± 0.57	881.5 ± 78.47
S4 W2	1.39 ± 0.05	1.91 ± 0.033	1.66 ± 0.57	692.3 ± 84.76
S4 W3	1.28 ± 0.02	1.42 ± 0.035	1.33 ± 0.5	299.7 ± 66.85
S4 W4	1.18 ± 0.039	1.26 ± 0.02	0.66 ± 0.57	268.1 ± 22.88

Table 18.3 Fresh weight, dry weight, and moisture content of wheat samples under various treatment conditions (mean ± SD of six values)

Samples	Fresh weight (g)	Dry weight (g)	Moisture content (%)
S1 W1	0.82 ± 0.024	0.19 ± 0.02	0.63 ± 1.77
S1 W2	0.8 ± 0.021	0.15 ± 0.12	0.65 ± 1.23
S1 W3	0.93 ± 0.024	0.2 ± 0.012	0.73 ± 1.26
S1 W4	0.47 ± 0.033	0.09 ± 0.011	0.38 ± 1.19
S2 W1	0.72 ± 0.02	0.21 ± 0.014	0.51 ± 1.77
S2 W2	0.79 ± 0.014	0.15 ± 0.0136	0.64 ± 1.74
S2 W3	0.81 ± 0.015	0.17 ± 0.0123	0.64 ± 1.29
S2 W4	0.46 ± 0.014	0.09 ± 0.008	0.37 ± 1.42
S3 W1	0.95 ± 0.02	0.25 ± 0.011	0.7 ± 1.57
S3 W2	0.99 ± 0.017	0.3 ± 0.01	0.69 ± 1.098
S3 W3	1.05 ± 0.035	0.32 ± 0.011	0.73 ± 1.59
S3 W4	0.45 ± 0.014	0.15 ± 0.012	0.3 ± 2.66
S4 W1	0.4 ± 0.028	0.14 ± 0.007	0.26 ± 0.86
S4 W2	0.36 ± 0.035	0.11 ± 0.012	0.25 ± 1.83
S4 W3	0.21 ± 0.021	0.05 ± 0.01	0.16 ± 1.25
S4 W4	0.19 ± 0.024	0.02 ± 0.02	0.17 ± 1.86

18.5.4.3 Vigor Index

The seedling length and vigor index values of various samples of wheat are determined by the product of seeding length in mm (sum of root length and shoot

Table 18.4 Leaf length, leaf width, and leaf area of leaves of various samples under different treatment conditions after 30, 60, and 90 DAS (mean ± SD of 10 values)

	30 DAS			60 DAS			90 DAS		
Samples	Leaf length	Leaf width	Leaf area	Leaf length	Leaf width	Leaf area	Leaf length	Leaf width	Leaf area
S1 W1	9.5 ± 0.67	0.5 ± 0.019	3.56 ± 0.23	14.3 ± 0.37	1.2 ± 0.022	12.87 ± 0.45	18.5 ± 0.67	1.7 ± 0.02	23.58 ± 1.67
S1 W2	9.32 ± 0.19	0.5 ± 0.02	3.49 ± 0.41	13.12 ± 0.13	0.98 ± 0.009	9.64 ± 0.43	16 ± 0.22	1.5 ± 0.023	18 ± 1.11
S1 W3	8.66 ± 0.15	0.53 ± 0.01	3.44 ± 0.34	13.06 ± 0.12	1.15 ± 0.012	11.26 ± 0.43	15.66 ± 0.17	1.73 ± 0.019	20.33 ± 1.17
S1 W4	7.63 ± 0.13	0.32 ± 0.007	1.83 ± 0.26	11.33 ± 0.18	0.91 ± 0.01	7.73 ± 0.34	13.83 ± 0.12	1.22 ± 0.032	12.66 ± 1.34
S2 W1	8.5 ± 0.21	0.35 ± 0.02	2.23 ± 0.33	11.95 ± 0.22	0.92 ± 0.008	8.24 ± 0.23	15.5 ± 0.3	1.34 ± 0.12	15.58 ± 1.11
S2 W2	8.66 ± 0.16	0.42 ± 0.001	2.72 ± 0.28	14.66 ± 0.26	1.05 ± 0.02	11.54 ± 0.54	19.66 ± 0.3	1.42 ± 0.021	20.94 ± 1.34
S2 W3	9.33 ± 0.11	0.61 ± 0.02	4.26 ± 0.36	13.53 ± 0.21	1.42 ± 0.01	14.41 ± 0.52	16.33 ± 0.31	1.67 ± 0.16	20.45 ± 1.42
S2 W4	6.93 ± 0.04	0.38 ± 0.001	1.97 ± 0.12	10.63 ± 0.03	0.81 ± 0.01	6.46 ± 0.34	13.83 ± 0.06	1.11 ± 0.012	15.35 ± 1.65
S3 W1	8.66 ± 0.13	0.38 ± 0.005	2.46 ± 0.17	11.76 ± 0.14	0.88 ± 0.002	7.76 ± 0.26	15.66 ± 0.23	1.38 ± 0.025	21.62 ± 1.26
S3 W2	9.5 ± 0.11	0.56 ± 0.001	3.99 ± 0.46	14.5 ± 0.25	1.18 ± 0.04	12.83 ± 0.48	18.5 ± 0.16	1.76 ± 0.031	32.56 ± 1.34
S3 W3	9.83 ± 0.12	0.59 ± 0.001	4.34 ± 0.51	13.53 ± 0.2	1.16 ± 0.001	11.77 ± 0.45	16.83 ± 0.32	1.79 ± 0.01	30.13 ± 1.31
S3 W4	6.66 ± 0.13	0.46 ± 0.015	2.3 ± 0.14	11.06 ± 0.12	0.89 ± 0.003	7.38 ± 0.33	15.66 ± 0.23	1.16 ± 0.15	18.17 ± 1.42
S4 W1	8.83 ± 0.17	0.43 ± 0.013	2.84 ± 0.16	11.88 ± 0.16	0.76 ± 0.015	6.77 ± 0.22	16.83 ± 27	1.23 ± 0.03	20.70 ± 1.26
S4 W2	8.83 ± 0.14	0.48 ± 0.002	3.17 ± 0.31	15.33 ± 0.24	0.91 ± 0.013	10.46 ± 0.31	18.83 ± 0.27	1.52 ± 0.02	28.62 ± 1.37
S4 W3	9.16 ± 0.12	0.57 ± 0.008	3.19 ± 0.17	13.86 ± 0.11`	1.03 ± 0.002	10.7 ± 0.22	17.16 ± 0.12	1.67 ± 0.018	28.66 ± 1.15
S4 W4	6.33 ± 0.14	0.31 ± 0.001	1.47 ± 0.11	9.96 ± 0.15	0.69 ± 0.008	5.15 ± 0.34	12.33 ± 0.34	1.1 ± 0.011	13.56 ± 1.82

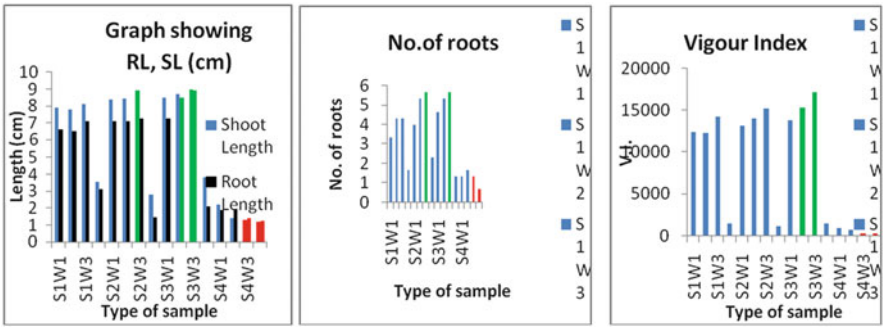

Fig. 18.10 Comparative analysis of SL, RL, no. of roots, and VI of various wheat samples with different treatments after 10 DAS

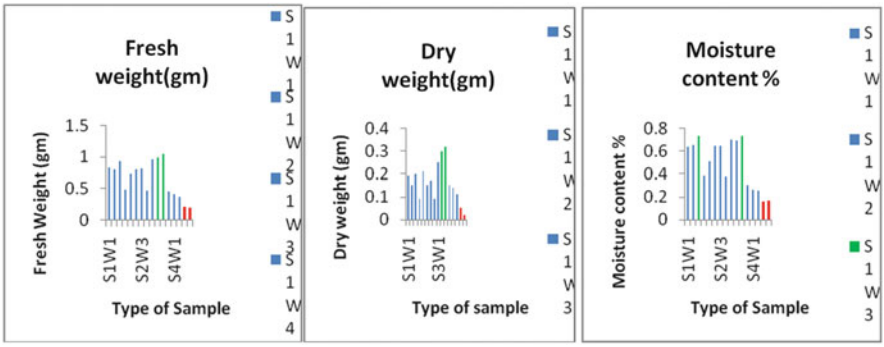

Fig. 18.11 Comparative analysis of fresh weight, dry weight, and moisture content of wheat samples under various treatment conditions after 10 DAS

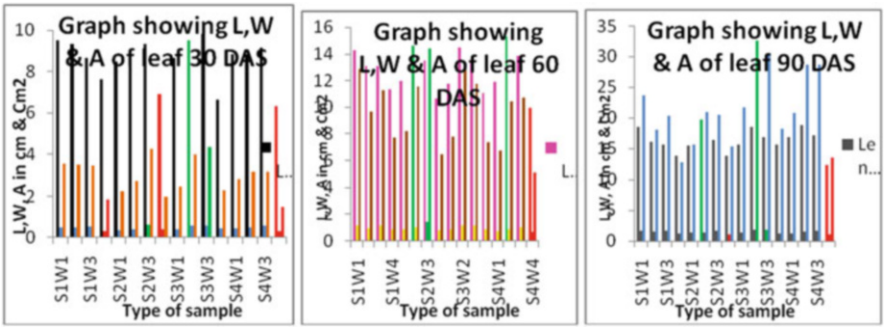

Fig. 18.12 Comparative analysis of length, width, and area of leaf of various wheat samples with different treatments after 30, 60, and 90 DAS

length) with germination percentage (Fig. 18.10). It is clearly shown that this value is lowest for S4 W3 and S4 W4 and highest for S3 W2 and S3 W3 which shows that effluent with low concentration up to 50% is suitable for good vigor crops.

18.5.4.4 Fresh Weight, Dry Weight, and Moisture Content

The fresh and total dry weights of wheat seedlings were determined after 10 days of the experiment (Table 18.3, Fig. 18.11). The plants were washed thoroughly with distilled water and were dried for 2 h under natural conditions in an open roof. The fresh weights were taken, and then the plants were packed in paper envelopes and oven-dried for 36 h at 70 °C, and the dry weight of each plant was also recorded. It showed clearly that fresh and dry weights are lowest for S4 W3 and S4 W4 and highest for S3 W2 and S3 W3. The difference of both of these, i.e., fresh weights and dry weights of the seedlings, gives the moisture content of the seedlings. Thus, it is also lowest for S4 W3 and S4 W4 but highest for S1 W3 and S3 W3 when compared with different treatment conditions which clearly shows that effluent with low concentration contributes significantly in the weight of wheat plant but high concentration of SME has negative effect on weight and moisture content of wheat.

18.5.4.5 Total Leaf Area

The leaf area was determined and was calculated on 30, 60, and 90 DAS of the experiment (Fig. 18.12). It is clearly observed (Fig. 18.12) that higher concentration of sugar mill effluents causes a decrease in leaf length, width, and area after 30 DAS, 60 DAS, and 90 DAS, whereas low concentration has stimulating effect on these factors. So it is quite clear that using low concentration of effluents in crops for irrigation purpose is beneficial for good growth of plant leaf and also increases photosynthesis rate and hence crop production. Positive effect on crops is indicated by green line and negative effect by red line (Fig. 18.12). S4 W4 and S2 W4 generally are having the least values, and S2 W2/S3 W2 are having the highest growth of leaf length, width, and area after 30 DAS, 60 DAS, and 90 DAS.

18.6 Wheat Seed and Straw Yield After Harvesting

When all the spikes became straw color, then harvesting was carried out, collected properly in separate bundles, and labeled correctly. Before drying in sunlight, plant height and number of tillers per plant for each pot were recorded. Height of the plant was recorded from ground level to the tip of the longest spike. Length of the spike was also noted in the same manner (Table 18.5).

Table 18.5 Harvested plant height and length of the spikes (cm) of various wheat samples with different treatments (mean ± SD of six values)

Samples	Plant height (cm)	Length of spike (cm)
S1 W1	73.84 ± 0.85	10.23 ± 0.24
S1 W2	70.12 ± 1.04	11.15 ± 0.38
S1 W3	68.32 ± 2.61	11.25 ± 0.58
S1 W4	34.42 ± 1.58	4.58 ± 0.38
S2 W1	70.35 ± 1.23	8.78 ± 0.36
S2 W2	77.36 ± 1.36	9.73 ± 0.27
S2 W3	85.36 ± 1.39	13.59 ± 0.4
S2 W4	29.59 ± 1.24	5.57 ± 0.36
S3 W1	70.75 ± 1.55	8.36 ± 0.36
S3 W2	66.15 ± 1.49	9.37 ± 0.15
S3 W3	88.62 ± 1.13	12.55 ± 0.39
S3 W4	26.28 ± 1.39	8.15 ± 0.47
S4 W1	30.79 ± 1.78	3.67 ± 0.2
S4 W2	32.52 ± 1.26	2.89 ± 0.26
S4 W3	32.12 ± 1.35	2.32 ± 0.31
S4 W4	23.12 ± 0.99	1.56 ± 0.43

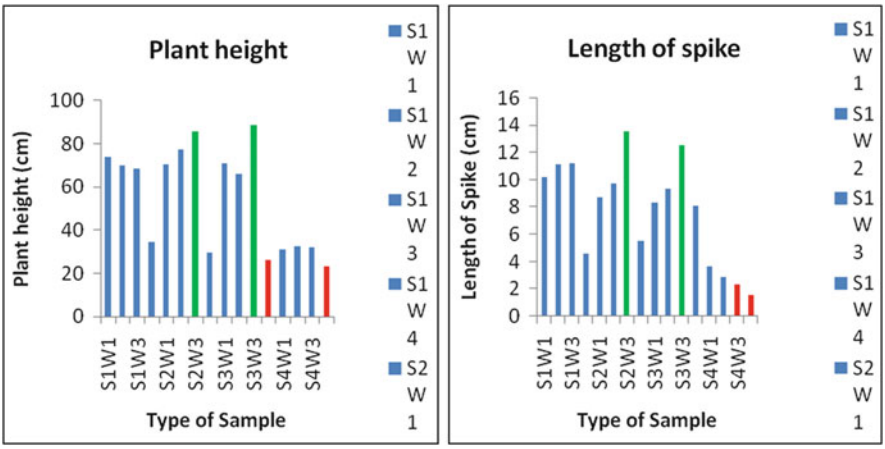

Fig. 18.13 Comparative analysis of plant height and length of spikes of various wheat samples with different treatments after harvesting

18.6.1 Plant Height and Length of the Spike

A comparative analysis (Fig. 18.13) showed that the plant height and length of spikes are lowest for S4 W3 and S4 W4 and highest for S3 W2 and S3 W3 which clearly shows that effluent with low concentration (up to 50%) is suitable for better growth over that of plant grown under control condition. The same kinds of observations were also made by Suresh et al. (2014) and Srivastava et al. (2015).

Table 18.6 Number of tillers or spikes/plant, spikelets/spike, and grains/spike wheat samples under various treatment conditions (mean ± SD of six values)

Samples	No. of tillers/plant	Spikelets/spike	Grains/spike
S1 W1	4.66 ± 0.82	15.5 ± 1.05	35.16 ± 2.79
S1 W2	4.83 ± 0.75	15 ± 0.89	34.5 ± 3.27
S1 W3	4.93 ± 0.52	17.66 ± 0.52	36.83 ± 2.63
S1 W4	2.5 ± 0.55	7.83 ± 0.75	15.66 ± 2.73
S2 W1	3.833 ± 0.75	15.5 ± 1.37	34.66 ± 3.5
S2 W2	4.66 ± 0.82	15.66 ± 1.03	32.5 ± 3.62
S2 W3	4.98 ± 0.98	17.33 ± 0.82	35.66 ± 3.26
S2 W4	2.16 ± 0.75	7.83 ± 1.17	13.33 ± 2.34
S3 W1	4.5 ± 0.54	15.66 ± 1.63	34.83 ± 2.93
S3 W2	4.66 ± 0.52	16.5 ± 1.05	36.66 ± 2.58
S3 W3	5.16 ± 0.51	16.83 ± 1.17	38 ± 2.6
S3 W4	2.3 ± 0.84	8.66 ± 0.82	15 ± 2.36
S4 W1	1.83 ± 0.75	6.83 ± 1.17	11.66 ± 1.75
S4 W2	1.23 ± 0.75	5.83 ± 1.17	8.83 ± 2.86
S4 W3	1.23 ± 0.98	5.76 ± 0.75	8.59 ± 1.67
S4 W4	1.166 ± 0.75	5.33 ± 0.82	8.34 ± 2.1

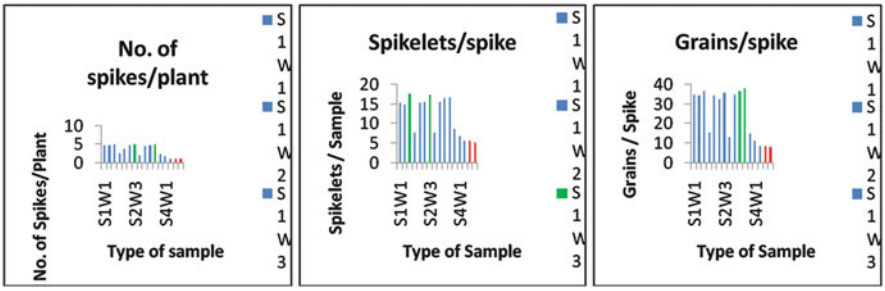

Fig. 18.14 Comparative analysis of number of spikes/plant, spikelets/spike, and grains/spike of various wheat samples with different treatments

18.6.2 Number of Tillers or Spikes/Plant, Spikelets/Spike, and Grains/Plant

The number of tillers/spikes per plant, spikelets/spike, and grains/spike had been analyzed (Table 18.6) of various wheat samples with different treatments of effluent. The comparative analysis of number of tillers/spikes per plant, spikelets/spike, and grains/spike (Fig. 18.14) shows that a number of tillers/spikes per plant, spikelets/spike, and grains/spike were maximum for samples S3 W3 and S2 W3, a number of spikelets/spike were maximum for samples S1 W3 and S2 W3, and a number grains/spike were maximum for samples S3 W3 and S3 W2 indicating that some quantity of effluent concentration (up to value of 50%) is beneficial and concentration higher

Table 18.7 Grain yield/pot, straw yield/pot, and biological yield/pot of wheat samples under various treatment conditions (mean ± SD of six values)

Samples	Grain yield/pot	Straw yield/pot	Biological yield/pot
S1 W1	95.88 ± 2.42	185.68 ± 12.31	281.56 ± 14.53
S1 W2	94.26 ± 2.97	168.22 ± 4.235	262.48 ± 7.21
S1 W3	99.67 ± 2.93	177.08 ± 8.84	276.76 ± 6.03
S1 W4	38.313 ± 3.86	90.29 ± 5.00	128.60 ± 4.09
S2 W1	99.09 ± 2.23	182.81 ± 7.37	281.90 ± 6.41
S2 W2	101.66 ± 3.01	213.44 ± 8.01	311.71 ± 10.73
S2 W3	105.303 ± 4.62	98.366 ± 4.01	318.74 ± 12.42
S2 W4	33.503 ± 3.95	93.43 ± 5.28	126.93 ± 6.48
S3 W1	95.96 ± 2.11	189.97 ± 6.91	285.94 ± 5.68
S3 W2	94.81 ± 1.78	190.28 ± 9.75	285.1 ± 10.05
S3 W3	102.006 ± 1.72	210.0433333 ± 7.74	295.76 ± 5.65
S3 W4	52.53 ± 1.11	193.76 ± 4.54	150.89 ± 3.39
S4 W1	65.716 ± 2.92	99.073 ± 1.55	164.79 ± 3.04
S4 W2	65.99 ± 1.14	91.9 ± 2.63	157.89 ± 2.88
S4 W3	68.13 ± 2.55	97.42 ± 4.82	165.5 ± 8.19
S4 W4	22.71 ± 2.41	86.61 ± 4.15	109.3 ± 5.94

than this leads to a reduction in the number of tillers/spikes per plant, spikelets/spike, and grains/spike.

18.6.3 Yield and Yield Components (Grain Yield, Straw Yield, and Biological Yield)

Grain yield, straw yield and total biological yield in grams per pot (Table 18.7) along with values of standard deviations of various wheat samples with different treatments. The grain yield per 10 plants was measured and converted into final grain yield per replicate. Similar to the grain yield, the straw yield was also determined after drying of grains in the sun and converted into yield per replicate. An average value of the pot of each replicate was noted along with values of their standard deviations. Both grain yield and straw yield collectively make biological yield, and it is also calculated per replicate, and an average value of the pot of each replicate was noted along with values of their standard deviations. It was observed that S3 W3 and S2 W2 are best conditions for grain yield and S3 W3 and S2 W2 for straw yield. The comparative analysis of grain yield, straw yield, and biological yield (Fig. 18.15) shows that some quantity of effluent concentration (up to value of 50%) is beneficial and concentration higher than this leads to reduction of wheat yield.

As mentioned in other parameters, also diluted effluents result in increase in crop yield, while increase in concentration of effluents causes reduction in yield of crops.

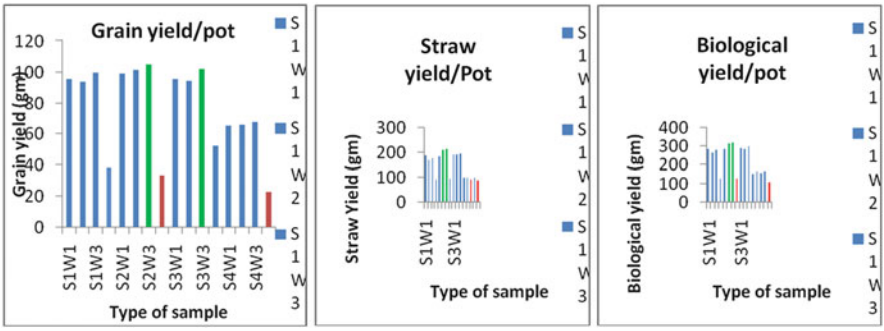

Fig. 18.15 Comparative analysis of grain yield/pot, straw yield/pot, and biological yield/pot of various wheat samples with different treatments

Grain yield is highest for S2 W3 and S3 W3, even higher than the normal field soil, whereas S2 W4 and S4 W4 have lowest yield due to contamination of soil. Similarly, S4 W2 and S4 W4 are on the lowest side, and S2 W2 and S2 W3 have the highest straw yield. These results indicate that high concentration of nutrients in the effluents cause increase in the yield of grain and straw as well. S4 W4 and S2 W4 samples have the lowest values of biological yield due to contamination of soil resulting in reduced growth, whereas S2 W2 and S2 W3 have shown the highest yield.

Variation in germination, shoot length, root length, and yield with variation in concentration of SME that was utilized for treatment of seeds clearly revealed that effluents exhibit profound effect on the abovementioned physiological parameters. Observations made from the experiments conducted indicate clearly that with gradual increase in concentration of the effluents (50–100%), a gradual decrease in germination rate, RL, SL, leaf area, and yield of crop was observed. Among various concentrations of effluents which were utilized during the study, 25 and 50% concentration of effluents was found to be most effective in increasing the germination rate and other parameters in wheat plant.

Highest germination speed was observed in SME concentrations of 25 and 50%. However, germination percentage decreases to 80–65% in wheat when seed was treated with higher concentration of effluents (more than 50%) (Pandey et al. 2008). The growth and germination percentage of seed inhibited at higher concentration of effluents may be due to osmotic pressure of high dose, which make imbibitions more difficult and reduce oxygen uptake by seedling (Khatoon et al. 2010), while diluting the effluent enhances the plant activities by providing required amount of nutrients present.

18.7 Conclusion

It is concluded that almost all the anions and cations are much higher than the permissible limits in SME. Considerable high values of BOD and COD were also observed. A number of tillers/plant and grains/plant were maximum for samples S3 W3 and S2 W3, a number of spikelets/spike were maximum for samples S1 W3 and S2 W3, S3 W3 and S2 W2 are the best conditions for grain yield and straw yield, and biological yield was best under S2 W2 and S2 W3 conditions. It has been observed that germination, harvesting, and postharvesting parameters were best for S2 W2, S2 W3, or S1 W3. Hence, some quantity of effluent concentration (up to value of 50%) is beneficial for crop growth as tests for wheat growth and concentration higher than this lead to reduction in vegetative growth as well as crop yield. Numbers of bacterial and fungal species are less in SME as compared to normal soil in this study, and it may be because of the negative impact of contaminants of SME on micro-biodiversity of soil.

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