DISTILLERY EFFLUENT: PROBLEMS AND PROSPECTS- A REVIEW

K. Kukreja, D.K. Gahlot and S. Suneja

Department of Microbiology, CCS Haryana Agricultural University, Hisar-125 004, India

Received: 10-08-2011

Accepted: 30-02-2012

ABSTRACT

Distillery effluent generally known as spent wash is considered one of the worst pollutants produced by industries. For every litre of alcohol 12-15 litres of spent wash is produced which has got appreciable amounts of organic load, high biological oxygen demand (BOD), chemical oxygen demand (COD) and is highly acidic in nature. To decrease the BOD and COD values, several distilleries have installed the biomethanation units but still contains high concentrations of organic pollutants and can not be discharged directly. Spent wash being plant originated, contains large quantities of soluble organic matter and plant nutrients, therefore, spent wash if utilized rationally and in appropriate concentration for crop production can prove to be a good source of nutrients and is expected to solve the problem of waste disposal. The purpose of this review is to discuss and summarize the available data on problems and prospects of distillery spent wash.

Key words: Distillery effluent, Spent wash, Pollution, Biological oxygen demand, Chemical oxygen demand.

Although industrialization is believed to be the index of modernization, it has its unavoidable effects on the pollution of air, water and soil depending upon the type of industry, nature of raw material being used and the manufacturing process involved. Since the industrialization and pollution are complementary to each other, necessary steps are to be taken for disposal of the pollutants. In a developing country like India, distilleries, the alcohol producing industries have become a major source of pollution as 88% of the raw materials are converted into waste and discharged into the water bodies causing water pollution. In the distillery, the commercial production of ethyl alcohol is carried out with diluted molasses by fermentation using yeast. After the recovery of alcohol, the fermented broth together with fermented washings and yeast sludge is discharged as spent wash, also known as sillage or vinase. For every liter of alcohol produced, about 15 liters of spent wash is released as wastewater which is one of the most recalcitrant and unutilized waste in nature. At present there are about 315 distilleries in India producing 50 to 60 billion liters of effluent annually. The distillery waste is dark brown colored, acidic in nature and is generally characterized by high level of biological oxygen demand (BOD), chemical oxygen demand (COD) and nutrient elements such as nitrogen (N) and potassium (K), that poses significant treatments and disposal problems. The presence of dark brown color in distillery effluent is mainly due to colored compounds which are formed during manufacturing of sugar from sugarcane juice (Wedzicha and Kaputo, 1992). Caramels and melanoidins constitute the main ingredients which are formed by Maillard amino-carbonyl reaction. These compounds have antioxidant properties which make them toxic to microflora generally present in waste water treatment plants (Kitts *et al.*, 1993).

Indiscrimate disposal of distillery waste water is becoming a serious problem throughout the world. The disposal of raw spent wash into natural water bodies may lead to eutrophication since the waste water contains high inorganic content, particularly N and P components that are available as nutrients for algae and aquatic plants and therefore can result in excessive growth in water bodies. The dark brown color of the spent wash is not only aesthetically objectionable but may have adverse effect on photosynthetic activity restricting

Corresponding author's e-mail: kkukreja@hau.ernet.in

the availability of light to aquatic plants. Acidity of spent wash may cause lowering of pH of streams which may also disrupt the aquatic system. Since the water has very high BOD, it putrifies under anaerobic conditions producing offensive odors. Metabolism of the organic matter by native heterotrophic microorganisms of the receiving water causes rapid oxygen depletion, which adversely affects the aquatic flora and fauna.

The spent wash is not only detrimental to aquatic life of receiving water streams but its land application is also hazardous (Mahida, 1981). If directly applied to soil, spent wash reduces soil porocity, oxygen transfer, soil alkalinity and manganese availability which results in the loss of soil fertility. The degradation of organics may also deplete the soil of its nitrogen, which may result in the inhibition of seed germination and plant growth. It also causes soil sickness due to clogging of pores by suspended matter leading to anaerobic conditions in the root zones because of which organic acids are liberated. Under these conditions H_2S is produced which is toxic to plants.

Considering the potential environment hazards due to the discharge of untreated spent wash, State and Central Pollution Control Boards have laid down the standards for treatment of effluent before disposal. According to the prescribed standards, waste waters should have a BOD less than 30 mg L⁻¹ before disposal into natural water bodies and less than 100 mg L⁻¹ for disposal on land. Therefore, in order to prevent uncontrolled discharge of waste and resulting deterioration of land and water resources, the need for the development of suitable technology for the treatment of distillery waste water is highly desirable.

Treatment of distillery effluent

Distillery spent wash which is considered the worst pollutants and is produced in large volumes is also very complex in composition. Therefore, more systematic studies are required to solve the problems of waste water treatment before disposal. The possible alternatives that have been explored for the treatment of spent wash can be divided into physico -chemical and biological methods.

Physico- chemical methods

The physico- chemical methods include dilution with fresh water, lagooning, coagulation,

filtration, adsorption, ion exchange, reverse osmosis, electro-flocculation, chemical oxidation, precipitation and incineration etc. (Rembowski, 1974). In general, physico- chemical treatment of spent wash has met with little success. Sedimentation and flocculation were found to be unsatisfactory even with the addition of coagulants (Ramteke, 1989).

Each of these methods has limitations and the choice of technology to use is left mainly with the distilleries. The first technique namely dilution and disposal is the simplest and easiest provided adequate water is available to reduce the pollution to an acceptable limit as prescribed by Indian Standard Institute (ISI). Each litre of effluent will need about 150-200 litres of fresh water. Unfortunately, at present this large amount of water is not available for dilution before disposal. Hence, dilution with fresh water is an attractive proposition but is impracticable.

The second technology available is lagooning and disposal. The effluent is stored in tanks which are 3 to 6 meters deep, for weeks or months. Spent wash stored in these lagoons undergoes decomposition in the absence of oxygen and is converted to gaseous products which are released into surrounding atmosphere. This leads to serious environmental problems mainly because of foul smell. Further these lagoons need adequate space and distilleries which do not have adequate space can not use this technology.

The third process of effluent treatment is incineration. This involves direct volatilization of effluent at high temperature. This technology is the best since it does not leave any residue behind. However, this process is highly expensive and for the industries having large volume of effluent like distilleries, this is not economical.

Chemical methods involve oxidation of dissolved organic and inorganic compounds. Chemical oxidation is relatively expensive. The oxidants that have been used to treat distillery waste include ozone, hydrogen peroxide, permanganate and chlorine etc.

Biological methods

In general biological processes take advantage of microorganisms to metabolize organic material to obtain energy for the biosynthesis and for maintenance. Organic compounds are converted to end products through oxidation/ reduction processes to serve as primary energy source and for the synthesis of cellular material. Biological treatment is considered comparatively more cost effective than physico- chemical methods.

The biological treatment comprises of anaerobic and aerobic treatment methods. Biodegradation of several organic compounds occurs under anaerobic conditions although the rates may not be as rapid as observed under aerobic conditions.

Anaerobic treatment of distillery effluent

In India, the primary spent wash is generally subjected to anaerobic digestion for the generation of methane as fuel while reducing its organic load (Das, 1990). The microbial degradation of organic material in anaerobic environment can be accomplished by microorganisms capable of using molecule other than oxygen as terminal electron acceptor. The anaerobic digestion ultimately results in the production of mixture of gases (biogas) comprising of methane (60-70%), CO_2 (25-30%) and small amounts of H_2 , N_2 and H_2S (Speece, 1983). The overall reaction is:

Organic matter \longrightarrow CH₄+CO₂+N₂+H₂S

The process is carried out by three different interdependent groups of bacteria (Sahm, 1984). The first group involves hydrolytic fermentative bacteria and acetogenic bacteria. The second group is collectively called as hydrogen producing acetogenic bacteria, which convert various intermediates formed during previous hydrolytic step into CO_2 , H_2 and acetate and the third group comprises of methanogenic bacteria, which utilize H_2 , CO_2 and acetate in the production of final product i.e. methane. In this process CO_2 is the terminal hydrogen acceptor and is analogous to oxygen in aerobic respiration. Different anaerobic treatment systems have been used for treatment of distillery effluent which include, anaerobic lagoons conventional anaerobic digesters, upflow anaerobic sludge blanket process and by phase anaerobic digesters (Sachiz et al., 1985, Rich, 1986, Pandey, 1989).

Secondary aerobic treatment

Anaerobic treatment methods are not sufficient to meet the norms of the statutory boards

for the disposal of anaerobic treated spent wash to prescribed limits of BOD. The primary anaerobic treatment is followed by secondary aerobic treatment which is very expensive and generally done by the use of trickling filters or by activated sludge system. Singh et al. (2010) have described polishing of biomethanated spent wash in constructed wetland and have recommended application of wetlands as High Rate Evaporation System, to reduce volume of spent wash after which effluents may be treated further conventionally. Bioremediation could be an alternative strategy to reduce the pollution load. Pollution treatment by microorganisms has the advantage of being simple in design and low in cost compared with other physical and chemical treatments. Accordingly, increasing attention has been directed towards utilizing microbes for decolorization of the effluent (Aoshima et al., 1985, Ohmomo et al., 1987, Ohmomo et al., 1988a, Ohmomo et al., 1988b, Murata et al., 1992, Singh et al., 1995).

Microorganisms living in polluted habitats evolve mechanisms either to avoid the toxic effect of the pollutants or to detoxify/degrade them. The polluted soils may act as reservoir of bacterial consortium capable of detoxifying pollutants. Extreme and rational screening of biodegradable microorganisms from areas contaminated or exposed to recalcitrant compounds for prolonged periods can facilitate evolution of biodegradable capability of microorganisms enriched cultures being used to search for organisms with degradable ability. A large number of bacteria and fungi have been reported to decolorize spent wash and to reduce its COD (Miyata et al., 2000, Gupta et al., 2001, Dikshit and Chakraborty, 2006). Another alternative to avoid this expensive secondary treatment, the possibilities of using distillery effluent in agriculture came into consideration.

When spent wash is disposed off on land, pollution of soil, water and air occurs. But disposal on land is still the most convenient practice. This is supposed to solve the problems of waste water disposal and source of plant nutrients for crop production. Several possibilities have been explored in order to utilize spent wash as a resource material and to reduce environmental pollution. The direct land application of spent wash in agricultural fields as irrigation water and fertilizer has shown to be advantageous for improving physical and chemical properties of soil. In case of continuous use, several negative effects have been reported. In soil, a maximum of 3.5-5.0 thousand litres of spent wash can be applied per hectare. This will require storage facilities for the remaining huge volumes and will need large areas for distilleries.

Agricultural uses of distillery effluent

The use of distillery effluent either solid or liquid has been practiced in India since the inception of this industry and hence application of distillery effluent to agricultural land is receiving an attention. This is supposed to solve the problem of waste water disposal as well as source of plant nutrients for crop production. Scientists in the field of agriculture are therefore, deeply concerned about its disposal on the agriculture land which is a limited commodity. The possible uses of spent wash for agronomical purposes are as manure, preparation of mixed fertilizer and for irrigation after suitable dilution. Digested spent wash can serve as a potential source of fertilizers if used rationally and in appropriate concentration.

For proper utilization of spent wash with minimum detrimental effect on crop ecology, it was felt necessary to understand its chemical composition, Being plant originated preliminary analysis has shown that it contains considerable amounts of plant nutrients, and organic matter. Nitrogen content in spent wash ranges from 1660-4200 mg/L and phosphorus (P) from 225-3038 mg/ L and potassium (K) from 9600-17475 mg/L. Calcium (Ca), magnesium (Mg), sulphate (SO_4) and chloride (Cl) are, also present in appreciable amounts. Thus it can effectively be used as a source of plant nutrients and as soil amendment. The high concentration of Ca (2050-7000 mg/L) in spent wash may have the potential in reclaiming the sodic soils similar to that of gypsum effect. Recently the presence of appreciable amounts of plant growth promoters i.e. gibberellic acid (GA) and indole acetic acid (IAA) have also been detected in spent wash which further enhances its nutrient value and would save the cost on fertilizer and facilitate reduction in pollution load (Murugaragavan, 2002). For proper utilization of spent wash with minimum detrimental effect on crop ecology, laboratory and field studies are required to outline the potential value and problems associated with the usage of spent wash and its environmetal

impacts. Some of the work done by different scientists in this field has been reviewed here.

Effect of digested spent wash on seed germination

Germination is a critical stage which ensures reproduction and consequently controls the dynamics of population, so it is a critical test for the probable crop production (Radosevich et al., 1997). The distillery spent wash is a mixture of organic and inorganic nutrient, but at higher concentrations, the ions present in the effluent may have detrimental effect on metabolic functions, adversely affecting seed germination and plant growth. Nevertheless the sensitivity of the plants varies from species to species to the effluent salinity. The high value of the total dissolved solids (TDS) of distillery effluent seems to be responsible for seed germination inhibition due to disturbed osmotic relation of the seeds with water or due to enrichment of salinity and conductivity of solutes (Hadas, 1976).

Sahai et al. (1983) found that higher concentrations (25% and above) of digested spent wash retarded both the speed of germination and seedling growth of paddy crop. At 5% concentration, overall growth of seedlings was better than in control. Mukherjee and Sahai (1988) studied the effect of distillery waste on seed germination, speed of germination and seedling establishment of Cajanus cajan. No germination was observed in 100% concentration of the effluent. The 5% concentration of the effluent provided optimum conditions for seed germination, speed of germination index (SGI), seed output and dry weight, beyond which these parameters decreased. They concluded that the distillery effluent is very toxic at higher concentrations to the growth of this crop but at lower concentrations (2.5% and 5%), the effect is beneficial.

Rajaram and Janardhanan (1988) investigated the effect of spent wash on seed germination of soybean, cowpea, rice and sorghum. Their study revealed that lower concentrations of the effluent promoted seed germination and early seedling growth in cowpea, rice and jowar while the higher concentrations retarded not only seed germination but also the early seedling growth in all the crop plants investigated. In soybean, percent germination and early seedling growth were markedly suppressed with the increasing concentrations of the effluent. Singh and Bahadur (1995) conducted an experiment to assess the relative tolerance of seeds of cereals, legumes and oilseeds with respect to their germination in distillery effluent of varying concentrations. They found that seeds of maize, rice, mustard, blackgram, pigeonpea, soybean and chickpea germinated normally in 20% effluent, whereas greengram seeds germinated normally even in 50% effluent. Seed germination of lentil and rice was drastically reduced in 50% effluent. In pure effluent (100% concentration), seeds of any crop did not germinate.

Sharma et al. (2002) conducted bioassay studies to assess the toxicity of raw and diluted distillery effluent on seed germination and seedling growth of sugarbeat. They reported that higher concentrations (>5%) of effluent were toxic, however, the effluent can be used for irrigation purpose after proper dilution. Ramana et al. (2002a) investigated the effect of different concentrations of distillery effluent on seed germination of some vegetable crops and found that lower concentrations of the effluent were not inhibitory for seed germination except in tomato, however complete failure of seed germination was observed at higher concentrations (75% and 100%) of effluent irrespective of crop species. These workers concluded that the effect of distillery effluent is crop specific and due care should be taken before using the effluent for irrigation purpose. Pandey et al. (2007) studied the effect of distillery effluent on seed germination of wheat (Triticum aestivum), pea (Pisum sativum) and lady's finger (Abelmoschus esculentus). They reported that germination percentage decreased with increasing concentrations of effluent in all the tested seeds, whereas the germination speed, peak value and germination value increased at 25% and 50% concentrations and decreased from 50% to 100% effluent concentrations. Deora et al. (2008) found that lower concentrations (10 and 20%) of digested distillery effluent were not inhibitory to seed germination. Germination rate was poor in 50% effluent concentration, while higher concentrations (75 and 100%) of effluent lead to complete failure of seed germination in wheat. Gahlot et al. (2011) studied that lower concentrations (2.5-20%) of digested distillery effluent were not inhibitory to seed germination under laboratory conditions while

higher concentrations (50 and 100%) lead to poor seedling growth and delayed seed germination in chickpea. Under pot house conditions, seed germination was observed up to 20% concentration (irrigation) and 250 m³ha⁻¹ (pre-sowing amendment) of digested spent wash and complete inhibition of seed germination was observed at higher concentrations (50% and 500 m³ha⁻¹). However, germination of chickpea seeds was observed at 50% dilution level of spent wash under laboratory conditions. The reason for this may be perhaps the seedlings being delicate, fail to absorb nutrients from soil saturated with high concentrations of spent wash, since emergence of plumule was taken as a criterion for germination under pot house conditions. All the seedlings were established in the control set and in those irrigated with lower concentrations of spent wash.

Effect of digested spent wash on nodulation

The soil fertility can be build up by spent wash application. If directly applied to soil, spent wash reduces soil porosity, soil alkalinity and oxygen transfer which results in loss of soil fertility (Mahida, 1981). The spent wash application may also have adverse effect on soil processes like organic matter decomposition, nutrient mineralization and nitrogen fixation. The soil microorganisms which are the major constituents of soil ecosystem and are involved in recycling of nutrients, get affected due to spent wash application. Juwarkar et al. (1990) found that raw distillery waste water reduced growth rate of Rhizobium and Azotobacter. These workers observed that when groundnut plants were irrigated with raw distillery waste water no fruits were produced and nodulation was also reduced. Joshi and Kalra (1995) observed the absence of nitrogen fixing bacteria in soil treated with distillery effluent.

Ramana *et al.* (2002b) conducted an experiment to evaluate the relative efficacy of three distillery effluents: raw spent wash (RSW), biomethanated spent wash (BSW) and Lagoon Sludge (LS) vis-à-vis recommended fertilizer (NPK + farmyard manure) on growth, yield and nitrogen fixation of groundnut and found increase in all growth parameters but decrease in nodulation and nitrogen fixation by all the distillery effluents. Application of distillery effluent significantly increased the yield of various crops (wheat, maize and sorghum etc.) but had adverse effect on legumes (Baskar et al., 2003). Bhalerao et al. (2004) observed that pre-sowing application and fertigation up to 300 m³ha⁻¹ showed beneficial effect on yield, nodulation and nutrients uptake in daincha but higher concentrations of digested spent wash showed adverse effect on nodulation in daincha. Gahlot et al. (2011) revealed that increased concentration of the digested spent wash affected the nodulation of chickpea. However, level and extent of affect was dependent upon the application of spent wash through soil amendment or application along with irrigation. Inclusion of digested spent wash with irrigation affected the nodulation even at lower concentration of 5% whereas application of digested spent wash in soil up to 100 m³ha⁻¹did not have adverse effect on nodulation. In our studies also root system of chickpea was very much affected with higher doses of spent wash application. There was decrease in length and size (biomass) of roots which is essential for nodule formation. This could also be one of the reasons for reduced nodulation. It is quite possible that the toxic components of spent wash might have affected the population of *Rhizobium* as well.

Effect of digested spent wash on photosynthetic activity

Plant productivity is limited by availability of photosynthates and its utilization. Production of photosynthates depends upon photosynthesis which is regulated by the rate of light and dark reaction, availability of CO_2 , moisture and chloroplastic pigments etc. All green colored plant parts contain pigments which absorb light and play important role in photosynthesis. These are chlorophyll *a*, chlorophyll *b*, xanthophylls and carotenoids. Studies have shown that high concentrations of digested spent wash may lead to reduced level of plant pigments affecting photosynthesis. In recent years, chlorophyll fluorescence analysis which is indicator of photosystem II has become a powerful tool to measure photosynthetic activity.

Sahai *et al.* (1983) investigated the effect of digested distillery waste on pigment contents of rice and found that chlorophyll a and chlorophyll b contents decreased with increase in spent wash concentrations. However, chlorophyll b content was higher in 5% concentration of spent wash than in control. The carotenoid content continued to

decrease up to 50% effluent concentration. They concluded that distillery effluent after proper dilution can be used for irrigation purpose. Babu *et al.* (1996) observed significant increase in total chlorophyll pigments, protein and N contents in distillery spent wash treated plants of *Sorghum vulgare* and *Cajanus cajan*. They concluded that spent wash does not contain heavy metals and can be used as fertilizer to increase crop production. Ramana *et al.* (2002c) revealed that the application of distillery effluents resulted in increased leaf area and chlorophyll content in maize.

Bioassay studies were carried out to assess the toxicity of distillery effluent (1-30%) on pigment content of sugarbeat. Higher concentrations were found to be toxic to chlorophyll pigment of sugarbeat (Sharma et al., 2002). Jain et al. (2002) studied the impact of distillery waste on growth attributes. chlorophyll content and enzyme activity of sugarcane. They found that low rate of crude spent wash (<100 ml kg⁻¹ of soil) increased these parameters while higher dose (>100 ml kg⁻¹ of soil) decreased these parameters significantly due to higher metal availability in soil and thus toxic to sugarcane. Singh et al. (2003) recorded that lagoon sludge; a distillery waste, increased the total chlorophyll content in groundnut leaves when irrigated with these effluents. Chandra et al. (2004) reported that lower concentrations (1-10%) of distillery effluent increased shoot, root length, biomass, protein content and chlorophyll values in *Phaseolus aureus* but raw distillery effluent had adverse effect on growth and chlorophyll content. Gahlot et al. (2011) measured chlorophyll a fluorescence to study the effect of digested spent wash on photosynthetic activity in chickpea. Chlorophyll a fluorescence is also an indicator of photo system II efficacy and this was maximum at 10% concentration level (irrigation) and at 100 m³ha⁻¹ (soil amendment) of digested spent wash. Slight decrease in chlorophyll a fluorescence was observed at higher application rate (20% and 250 m³ha⁻¹) indicating that application of digested spent wash either along with irrigation or as soil amendment at these concentrations, slightly enhanced the photosynthetic activity. Almost similar type of results have been reported elsewhere also.

Effect of digested spent wash on plant growth, nutrient uptake and soil properties

Studies have shown that spent wash application at higher doses is detrimental to crop growth and soil fertility, its use at lower doses remarkably improves germination, plant growth, nutrients uptake and yield of crops. Somawanshi and Yadav (1990) found that addition of diluted spent wash would not add soluble salts to the soil provided there was sufficient leaching of soil solution. However, addition of concentrated spent wash would result in increased salinity of both soil and ground water. Pawar et al. (1992) explored the possibility of improving the physico-chemical properties of typical saline calcareous soil by the application of spent wash. The addition of diluted spent wash improved the physico-chemical properties as indicated by decrease in electrical conductivity (EC) of soil. There were significant changes in the exchangeable K^+ , Ca⁺ and Mg²⁺. The nutrient status of the experimental soil improved considerably as evidenced by performance of sugarcane grown on this soil in a pot culture experiment. These workers (Pawar et al., 1993) suggested that irrigation with spent wash could prevent iron chlorosis in sugarcane plants grown on saline calcareous soil. The application of distillery waste water up to 160m³ha⁻¹ in mungbean lead to increase in dry matter production, N and P uptake, while at 640 m³ha⁻¹ application rate, drastic decline in dry matter was observed and it was even less than the control (Goyal *et al.*,1995). They also observed that the electrical conductivity of soil increased to about three folds by application of distillery waste water equivalent to 320 m³ha⁻¹ irrigation.

Babu *et al.* (1996) studied the foliar application of distillery spent wash (0-100% dilution) on *Sorghum vulgarae* and *Cajanus cajan*. The application of spent wash did not cause deformities and derangement of plant metabolism. Significant increase in all growth parameters was observed while the greatest increase was observed in *C.cajan*. Total chlorophyll, phenols, proteins, amino acid, starch and nitrogen contents in the leaves increased in treated plants. They concluded that as the spent wash does not contain heavy metals, it may be used as a fertilizer to increase crop production. Zalawadia *et al.* (1997) conducted a pot house experiment to study the effect of irrigation with tube-well water and diluted spent wash on yield and nutrient uptake by sugarcane. Studies revealed that less diluted spent wash gave higher yield than the concentrated one, though there was tendency for slight increase in soil salinity due to the use of spent wash, yet the nutrients availability of P, K and S increased considerably.

Singh and Bahadur (1997) found that regular irrigations with five times diluted effluent had no advantage over ten times diluted one and at the same time it had no harmful effect on plants. Single application with effluent at 45 days had no significant effect on maize yield, but two such irrigations of effluent at 45 and 60 days increased yield by 37%. Alternate irrigations of effluent and fresh water increased yield by 42% over continuous fresh water irrigation. Regular irrigation with ten and five times diluted effluent increased yield by 78% and 59% respectively over fresh water irrigation. Thus five times diluted effluent can be safely applied with advantage to maize crop. An increased yield of wheat and rice on irrigation with diluted post methanated distillery effluent has also been reported by Pathak et al. (1998, 1999). Joshi et al. (1996) have also shown increase in biomass of wheat and rice with application of distillery effluent. These workers reported increase in organic carbon content, available K and electrical conductivity (EC) of post harvest soil in effluent treated plants but there was no significant change in soil pH.

Patil et al. (2000) found that application of spent wash at 50 m³ha⁻¹ had the highest dry fodder yield (374 g pot¹), protein content (61.2%) and nutrient uptake (3.24 g N pot⁻¹) in 37.5 m³ spent wash pot⁻¹ of fodder maize. Addition of spent wash at 62.5, 75 and 87.5m³ha⁻¹ increased the total leaf chlorophyll content (0.94, 0.96 and 0.91mg g⁻¹ fresh weight, respectively) in maize. Gopal et al. (2001) conducted a field experiment to assess the optimum dilution of distillery effluent for sugarcane crop in sandy soil. Sugarcane yield was highest in soil irrigated with the treated effluent (1:10) dilution. There were no significant differences in pH and electrical conductivity between soils irrigated with the treated effluents (1:10, 1:20, 1:30, 1:40 and 1:50) and control. Levels of organic carbon, macro and micro nutrients and soil microbial population were significantly higher in treated soils.

Pushpavalli et al. (2002) studied the effect of treated distillery effluent application on sugarcane at Tamilnadu using different dilutions with the well water viz. 1:10, 1:20, 1:30, 1:40 and 1:50 and found higher nutrient status (163 kg N ha-1 ,67 kg P ha-1 and 143 kg K ha-1) in lower dilutions of effluent and decreased nutrient status (11.8 kg N ha⁻¹, 21 kg P ha^{-1} and 103 kg K ha^{-1}) with higher (1:50) dilution of effluent. However, effluent application did not have any influence on juice quality. The highest cane yield obtained was 170 tones per hectare in the plot where the effluent was applied at dilution of 1:30. Similar results were obtained by Devarajan et al. (1998). Ramana et al. (2002c) compared the manurial potential of raw spent wash, biomethanated spent wash and lagoon sludge in groundnut and found that all the three distillery effluents increased crop growth rate, total dry weight, nutrient uptake (N, P and K) and finally seed yield compared to control. However, distillery effluent did not influence protein and oil contents. It was concluded that these effluents because of their high manurial value could supply nutrients, particularly K, N, and S to the crops and reduce the fertilizer requirement of the crop. Nevertheless, crop performance and yield was overall less than that of recommended NPK+FYM, probably on account of the failure of the effluents to supply balanced nutrition to the plants for achieving their potential growth capacity. Some other workers have also reported the possibility of saving N, P and K fertilizers through effluent irrigation (Zalawadia and Raman, 1994, Rajannan et al., 1998).

Jain *et al.* (2002) studied the impact of distillery effluent on growth attributes of sugarcane in pot culture conditions and found that low rate of crude spent wash showed improvement in bud sprouting and growth attributes, whereas, higher doses (100 ml spent wash kg¹ soil) decreased these significantly. Jeyabaskaran *et al.* (2003) studied the effect of potassium rich cement kiln flu dust and distillery effluent as substitute for potassium fertilizer on growth, yield and quality of banana and observed that cement kiln flue dust, distillery effluent and 60% of K dose gave the highest bunch weight which was even higher than that obtained with 100% commercial K alone.

Application of distillery effluent significantly increased the yield of various crops (wheat, maize

and sorghum) but had adverse effect on legumes (Baskar et al., 2003). These worker demonstrated that application of distillery spent wash/effluent after proper dilution (1:10 to 1:50) significantly increased the EC, organic carbon, available N, P, K, Ca, Mg and micro nutrient status of the soil. Singh et al. (2003) studied the effect of distillery effluents (biomethanated spent wash, raw spent wash and lagoon sludge) on plant and soil enzymatic activities and groundnut quality. They found that distillery effluents did not affect the oil contents, crude and true protein contents in groundnut but increased the seed yield and content of methionine by 44% and cystein by 24%. The application of distillery effluents did not affect the nitrate reductase activity but biomethanated spent wash significantly increased the nitrate content in rhizospheric soil. All the three effluents significantly increased the dehydrogenase and alkaline phosphatase activity more than recommended NPK+FYM.

Bhalerao et al. (2004) investigated the effect of pre-sowing application and fertigation of secondary treated biomethanated effluent on soil properties, growth, yield and nutrient uptake by green manuring crop, dhaincha. They showed that pre sowing application and fertigation at 300 m³ secondary treated biomethanated effluent (STBME) ha-1 had beneficial effect on yield and nutrient uptake in dhaincha. The application of STBME resulted in the reduction of pH, slight increase in EC and considerable increase in organic carbon and soil available N and K content. The increase in K content was significant. Banerjee et al. (2004) studied the effect of various concentrations (20, 40, 60, 80 and 100%, v/v) of distillery effluent on different growth parameters of Casuarina equisetifolia. A significant increase was found in all the parameters up to 60%concentration of the effluent and beyond that there was decline. It was concluded that the distillery waste water can be used for irrigation purpose after appropriate dilution.

Sukanya and Meli (2004a) conducted an experiment on maize using different dilution levels (1:5, 1:10, 1:25 and 1:50) of effluent and reported higher grain yield, higher protein and reducing sugar content of maize with 1:5 dilution levels of effluent. The electrical conductivity and organic carbon content of soil increased with increase in concentration of effluent (lower dilutions of effluent) but pH of soil remained unchanged (Sukanya and Meli, 2004b). These workers also studied the effect of different dilutions of effluent on performance of wheat and reported that higher dilution level of 1:50 gave significantly higher yield than other dilution levels, (Sukanya and Meli, 2004c). Using varying nitrogen substitution levels Sukanya and Meli (2004d, 2004e) investigated the role of distillery effluent as nitrogen source in maize as well as in wheat. The substitution level of 50% through inorganic source recorded significant higher grain yield followed by 25% through solid distillery + 75% through inorganic source. Thus there is possibility of saving 50% of the cost of inorganic nitrogen fertilizer. The nitrogen substitution levels did not bring significant influence on soil properties like bulk density, electrical conductivity, organic carbon and micro-nutrient status of the soil. However, the major nutrients like N, P and K in soil were increased significantly at 100% substitution through distillery effluent. These workers also studied the soil fertility status in wheat irrigated with spent wash. Results revealed that the electrical conductivity and organic carbon content of soil increased with an increase in concentration of spent wash in irrigation water. The reverse trend was observed with bulk density and reduction in bulk density is a favorable trait for the sandy loam soil. However, pH of soil did not show marked change. Availability of N, P, K, Zn, Cu, Fe and Mn in soil was more in plot irrigated with undiluted effluent which decreased progressively with an increase in dilution level (Sukanya and Meli, 2005).

Mahimairaja and Bolan (2004) found that higher doses (> 250 m³ha⁻¹) of spent wash application are detrimental to crop growth and soil fertility but its use at lower doses (125 m³ha⁻¹) remarkably improves germination, growth and yield of dry land crops due to the presence of considerable plant nutrients and organic matter.

Hati *et al.* (2005) studied the influence of added spent wash on soil physical properties and yield of wheat under soybean–wheat system. They reported that application of spent wash at lower dilution level caused noticeable improvement in the soil physical properties. However, increase in EC was a cause for concern at high level of effluent application. Bhalerao *et al.* (2005) showed that continuous pre-sowing application or fertigation with 100 m³ secondary treated biomethanated effluent

(STBME) ha⁻¹ to dhaincha (*Sesbania aculeata*) and 200 m³ STBME ha⁻¹ to sugarcane improved the growth and yield of the crops. Application of STBME reduced soil pH, salinity increased the soil EC, considerably increased organic carbon and soil available nitrogen and potassium content. The increase in potassium content was prominent.

Biswas et al. (2005) found that soil application of untreated spent wash decreased extractable nitrate content with increase in level of spent wash application in black, alluvial and red soil. In contrast, extractable nitrate content increased with the increase in level of spent wash application in all the three soils. Qizhan et al. (2006) mixed sugar mill based distillery effluent with other fertilizer and observed a significant economic benefit of applying the liquid fertilizer in sugarcane. Gahlot *et al.* (2011) observed that lower concentration (5%) of digested spent wash had no adverse effect on shoot and root growth and maximum shoot and root growth was observed at 2.5% concentration of the digested spent wash. Likewise, lower concentrations up to 50 m³ha⁻ ¹ of digested spent wash had beneficial effect on plant growth. Nevertheless crop performance was overall less than recommended dose of fertilizers probably on account of the failure of the spent wash to supply balanced nutrition to the plants for achieving their potential growth capacity. With increase in concentrations of spent wash, there was decrease in N and P uptake when plants were irrigated with different levels of digested spent wash. Similarly with pre-sowing amendment of digested spent wash, nutrient uptake increased up to 50 m³ha⁻¹ but at higher doses it decreased. There was no significant increase in the soil pH with the application of distillery spent wash. However, increase in soil EC was observed with increased spent wash application and this increase was much higher with repeated (irrigation) application than one time application (pre-sowing amendment) of digested spent wash. Percent organic carbon, total N and P contents of post harvest soil also increased with increasing doses of spent wash in both parts of the experiments (irrigation as well as amendment). Deora *et al.* (2008) in our laboratory studied the effect of different concentrations of digested distillery effluent on plant growth of wheat. They found that lower concentrations (10 and 20%) of effluent were not inhibitory to plant growth when irrigated after

germination. But plant growth was affected drastically at higher concentrations of effluent when applied after germination, while complete suppression of plant growth was observed when 50% effluent was applied throughout the experiment. Gahlot *et al.* (2011) also studied the effect of graded doses and method of application of digested spent wash on nodulation, nutrient uptake and photosynthetic activity of chickpea and found that lower concentrations of spent wash were not detrimental for all the parameters studied while higher concentrations were inhibitory.

CONCLUSIONS

Distilleries generate large volumes of highstrength wastewater that is of serious environmental concern. Spent wash disposal even after conventional treatment is hazardous and has high pollution potential due to the accumulation of recalcitrant compounds. Various physico-chemical and biological treatment options are available for the treatment of distillery wastewaters, but most of them could not find their application in the industry due to their technical limitations or exorbitant cost. This review presents an account of the problem and a detailed overview on the existing scenario on the treatment of distillery wastewater and its proper utilization in agriculture and also issues requiring further research in the field of distillery wastewater.

Today as we are aware most of the Indian distilleries are treating this distillery waste water by anaerobic digestion process of biomethanation to recover methane gas (Best technology so far). This technology saves 50% of the coal requirement by replacing it by methane generated during anaerobic generation. Due to high biochemical oxygen demand of raw spent wash, application of anaerobic treatment technology with biogas recovery has been reported to be highly effective (Nandy *et al.*, 2002). Anaerobic treatment is an accepted practice and various high rate anaerobic reactor designs have been tried at pilot and full-scale operation. However, anaerobically treated effluent still contains high concentrations of organic pollutants and as such cannot be discharged directly (Nandy et al., 2002). The recalcitrant nature of effluent is due to the presence of brown polymers melanoidin, caramel and alkaline degradation products. Melanoidin pigments are formed by the non-enzymatic amino carbonyl reaction i.e. Millard reaction. It possesses antioxidant property causing toxicity to many microorganisms involved in conventional wastewater treatment processes. Reverse phase thin layer chromatography identified gallic and vanillic acid present in spent wash. Microbial decolourization and degradation is an environment friendly and cost competitive alternative to chemical decomposition processes (Kumar et al., 1997). In order to reduce the colour and COD, it is considered highly desirable to exploit the biodegradation potential of soil microorganisms from polluted sites exposed to recalcitrant compounds of distillery spent wash for prolonged periods. As such polluted soils can facilitate selection of biodegradative capability in microorganisms and may act as reservoir of fungal communities capable of degrading pollutants. Large volumes of spent wash, characterized by high Biochemical Oxygen Demand (BOD), low pH, obnoxious smell, high Chemical Oxygen Demand (COD), and melanoidin polymers and extremely dark brown color, are generated from these distilleries. Since the quantity of spent wash released/day/ distillery is about one lakh liter, it cannot be consumed in total for biocompost production. So far, there has been limited success in search for dual system of fungi for biodegradation and alga for decolourization. This study is centered on the concept that Biomethanated Distillery Spent Wash (BMSW) could be used for the irrigation purpose after suitable dilution with water and its utility as a nutrient source without adversely affecting soil and crop health.

REFERENCES

- Aoshima, I., Tozawa, Y., Ohmomo, S. and Ueda, K. (1985). Production of decolourizing activity for molasses pigment by Criolus versicolor.Ps4a. *Agric. Biol. Chem.* **49** : 2041-2045.
- Babu, R. S., Saralabai, V. C., Muralidharan, K. S. and Vivekanandan, M. (1996). Foliar application of distillery spent wash as a liquid fertilizer for betterment of growth of *Sorghum vulgare* and *Cajanus cajan. J. Appl. Biochem. Biotechnol.* Totowa, N.J. Human press. **59** (1): 87-91.
- Banerjee, A. C., Bajwa, I. and Behal, K. K. (2004). Effect of distillery effluent on growth of *Casuarina equisetifolia*. J. Indus. Poll. Cont. **20** (2): 199-204.

AGRICULTURAL REVIEWS

- Baskar, M., Kayalvizhi, C. and Bose, M. S. C. (2003). Eco-friendly utilization of distillery effluent in agriculture review. *Agri. Rev.* **24** (1) : 16-30.
- Bhalerao, V. P., Jadhav, M. B. and Bhoi, P. G. (2005). Effect of distillery's biomethanated effluent on yield, quality of adsali sugarcane and soil properties. *Co-operative Sugar.* **36** (8) : 653-659.
- Bhalerao, V. P., Jadhav, M. B., Power, A. B. and Bhoi, P. G. (2004). Effect of pre-sowing application and fertigation of secondary treated biomethanated effluent on soil property and growth of green manuring crop. *Co-operative Sugar.* **36** (2) : 155-159.
- Biswas, A. K., Kundu, S., Saha, J. K. and Raman, S. (2005). Loss of nitrate from soil treated with spent wash. J. Environ & Ecol. 23 (1):84-87.
- Chandra, R., Kumar, K. and Singh, J. (2004). Impact of anaerobically treated and untreated (raw) distillery effluent irrigation on soil microflora, growth, total chlorophyll and protein contents of *Phaseolus aureus* L. *J. Environ. Bio.* **25** (4) : 381-385.
- Das, A. K. (1990). Energy from distillery effluent. Chemical Engg. World (CEW). 15:49.
- Deora, A., Kukreja, K. and Suneja, S. (2008). Effect of digested distillery effluent on seed germination and plant growth of wheat. *Research on Crops.* **9** (1) : 110-115.
- Devarajan, L., Rajannan, G. and Oblisami, G. (1998). National seminar on application of treated effluent for irrigation held at Regional Engineering College, Trichy, 23rd March.
- Dikshit, A. K. and Chakraborty, D. (2006). A techno-economic feasibility study on removal of persistent colour and COD from anaerobically digested distillery effluent: a case study from India. *Clean Techn. Environ. Policy* **8**:273-285.
- Gahlot, D. K., Kukreja, K., Suneja, S. and Dudeja, S. S. (2011). Effect of digested distillery spent wash on nodulation, nutrient uptake and photosynthetic activity in chickpea (Cicer arietinum) Acta Agronomica Hungarica. **59**(1): 73-85.
- Gopal, H., Kayalvizhi, C., Baskar, M., Bose, M. S. C. and Sivanandham, M. (2001). Effect of distillery effluent on changes in soil properties, microbial population, yield and quality of sugarcane in sandy soils. Proceedings of the 63rd Annual convention of the sugar Technologist Association of India, Jaipur, India 25th 27th August, 2001. pp. 52-58.
- Goyal, S., Chander, K. and Kapoor, K. K. (1995). Effect of distillery wastewater application on soil microbiological properties and plant growth. *J. Environ. & Ecol.* **13** (1): 89-93.
- Gupta, S., Pandey, A. K., Sharma, N. C., Pandey, P. and Sharma, C. B. (2001). Studies on the biological treatment of digested spent wash effluent using mutant strain of *Phanerochaete chrysosporium*. *Proc. Natl. Acad. Sci. India*. **71B** : 259-267.
- Hadas, A. (1976). Water uptake and seed germination of leguminous seeds under changing external water potential in osmoticum solution. J. Exp. Bot. 27 : 480-489.
- Hati, K. M., Biswas, A. K., Bandyopadhyay, K. M., Mandal, K. G. and Misra, A. K. (2005). Influence of added spent wash on soil physical properties and yield of wheat under soybean-wheat system in a vertisol of central India. J. Environ. & Ecol. 23 (3): 631-634.
- Jain, R., Shahi, H. M., Srivastav, S., Madan, V. K., Jain, R. and Hogarth, D. M. (2002). Impact of distillery effluent on growth attributes, chlorophyll content and enzyme activity of sugarcane. International society of sugarcane technologist. Proceeding of 14th congress, Brisbane, Australia 2 : 155-157.
- Jeyabaskaran, K. J., Pandey, S. D. and Gomadhi, G. (2003). Effect of potassium rich cement kiln flue dust and distillery effluent as substitute for potassium fertilizer on growth, yield and quality of 'Ney Poovan banana (Musa x paradisiacal). *Indian J. Agri. Sci.* **73** (12) : 645-648.
- Joshi, H. C. and Kalra, N. (1995). Distillery waste utilization in agriculture. Yojana. 39: 8-9.
- Joshi, H. C., Chaudhary, A., Pathak, H., Kalra, N., Chaudhary, R. and Kumar, S. (1996). Proceeding of national symposium on use of distillery and sugar industry waste in Agriculture. 28th and 29th October, 1996. AC and RI, Trichy. pp. 97-107.
- Juwarkar, A., Dutta, S. A. and Juwarkar, A. (1990). Impact of distillery effluent application to land on soil microflora. *J. Environ. Monitoring and Assessment.* **15** (2) : 201-210.
- Kitts, D. D., Wu, C. H., Stich, H. F. and Powrite, W. D. (1993). Effect of glucose lysine maillard reaction products on bacterial and mammalian cell mutagenesis. *J. Agric. Food Chem.* **41**: 2353-2358
- Kumar, V., Wati, L., FitzGibbon, F., Nigam, P., Banat, I. M., Singh, D. and Marchant, R. (1997). Bioremediation and decolorization of anaerobically digested distillery spent wash. *Biotechnol. Lett.* 19: 311-314.
- Mahida, U. N. (1981). Water pollution and disposal of waste water on land. Tata McGraw Publishing Co. Ltd., New Delhi.

- Mahimairaja, S. and Bolan, N. S. (2004). Problems and prospects of agricultural use of distillery spent wash in India. In: Singh, B. Super soil 2004: Proceedings of the 3rd Australian New Zealand Soils Conf. University of Sydney Australia, 5-9 Dec. 2004. S3 (www.regional.org.au/au/assoi/supersoil 2004).
- Miyata, N., Mori, T., Iwahori, K. And Fujita, M. (2000). Microbial decolourization of melanoidin- containing wastewaters: combined use of activated sludge and the fungus *Coriolus hirstus. J. Biosci. Bioeng.* **89** (2) : 145-150.
- Mukherjee, U. and Sahai, R. (1988). Effect of distillery waste on seed germination, seedling establishment and early seedling growth of *Cajanus cajan* L. (Var. 5-16). *Acta Bota. Indica.* **16** (2) : 182-185.
- Murata, M., Terasawa, N. and Homma, S. (1992). Screening of microorganisms to decolourize a model melanoidin and chemical properties of a microbially treated melanoidin. *Biosel. Biotechnol. Biochem.* **56** : 1182-1187.
- Murugaragavan, R. (2002). Distillery spentwash on crop production in dryland soils. M.Sc. (Environmental Sciences) Thesis, Tamilnadu Agricultural University, Coimbatore, India.
- Nandy, T., Shastry, S. and Kaul, S. N. (2002). Wastewater management in cane molasses distillery involving bioresource recovery. *J. Environ.Manage*. **65** : 25-38.
- Ohmomo, S., Yoshikawa, H., Nozaki, K., Nakaijma, T., Daengusubha, W. and Nakamura, W. (1988b). Continuous decolourization of molasses waste water using immobilized *Lactobacillus hilgardii* cells. *Agri. Biol. Chem.* 52 : 2437-2441.
- Ohmomo, S., Kainuma, M., Kamimura, K., Sirianutapiboon, S. and Aoshima, I. (1988a). Adsorption of melanoidin to the mycelia of *Aspergillus oryzae* Y-2-32. *Agri. Biol. Chem.* **52** : 381-386.
- Ohmomo, S., Kaneko, Y. and Sirianutapiboon, S. (1987). Decolourization of molasses waste water by thermophillic strain, *Aspergillus fumugatus* G-2-6. *Agri. Biol. Chem.* **51** : 3339-3346.
- Pandey, A. (1989). Anaerobic waste treatment, two phase reactor concept. *Encol.* **3** : 25.
- Pandey, S.K., Pallavi, T. and Gupta, A.K. (2007). Physico-chemical analysis and effect of distillery effluent on seed germination of wheat, pea and lady's finger. ARPN *J. Agri. and Biol. Sci.* **2** : 35-40.
- Pathak, H., Joshi, H. C., Chaudhary, A., Chaudhary, R., Kalra, N. and Dwiwedi, M. K. (1998). Distillery effluent as soil amendment for wheat and rice. *J. Indian Soc. Soil Sci.* **46** (1) : 155-157.
- Pathak, H., Joshi, H. C., Chaudhary, A., Chaudhary, R., Kalra, N. and Dwiwedi, M. K. (1999). Soil amendment with distillery effluent for wheat and rice cultivation. *Water Air Soil Pollution*. **113** (1-4) : 133-140.
- Patil, G. D., Pingat, S. M. and Yelwanda, A. J (2000). Effect of spent wash on soil fertility, uptake, quality and yield of fodder maize. J. Maharasttra Agri. Uni. 25 (2):168-170.
- Pawar, R. B., Desai, B. B., Chavan, U. D. and Naik, R. M. (1993). Effect of spent wash on growth parameters of sugarcane. *Indian Sugar.* **43** (2) : 107-114.
- Pawar, R. B., Desai, B. B., Chavan, U. D. and Naik, R. M. (1992). Effect of spent wash on physico-chemical properties of saline calcareous soil. J. Maharasttra Agri. Uni. 17(1):1-3.
- Pushpavalli, R., Kotteeswaran, P., Krishnamurthi, M. and Parmeshwaran, P. (2002). Effect of treated distillery effluent application on soil and yield of sugarcane at Nellikuppam in Tamil-Nadu, India. 17th WCSS, 14th -21th August, Thailand. Paper no. 1826: 1-5.
- Qizhan, T., Zhongxiao, T., Shubio, Z. and Deng, Y. (2006). Effect of liquid fertilizer made from sugar mill based distillery effluent on sugarcane. Sugar. Tech. 8 (4): 303-305.
- Radosevich, S., Holt, J. and Ghersa, C. (1997). Weed ecology implication for management. Pub. Wiley. New York.
- Rajannan, G., Parvinbanu, K. S. and Ramaswami, P. P. (1998). Utilization of distillery effluent based compost for crop production and economization of fertilizer use. *Indian J. Environ. Health.* **40** (3): 289-294.
- Rajaram, N. and Janardhanan, K. (1988). Effect of distillery effluent on seed germination and early seedling growth of soybean, cowpea, rice and sorghum. *Seed Research*. **16** (2) : 173-177.
- Ramana, S., Biswas, A. K. and Singh, A. B. (2002c). Effect of distillery effluent on some physiological aspects in maize. J. Bioresour. Technol. 84 (3): 295-297.
- Ramana, S., Biswas, A. K., Kundu, S., Saha, J. K. and Yadava, R. B. R. (2002a). Effect of distillery effluents on seed germination in some vegetable crops. *J. Bioresour. Technol.* **82** : 273-275.
- Ramana, S., Biswas, A.K., Singh, A. B. and Yadava, R. B. R. (2002b). Relative efficacy of different distillery effluents on growth, nitrogen fixation and yield of groundnut. *J. Bioresour. Technol.* **81** : 117-121.
- Ramteke, D. S. (1989). Comparative adsorption studies on distillery waste on activated carbon. *Indian J. Environment Health* **29** : 105.
- Rembowski. (1974). Investigation on the application of reverse osmosis for treatment and utilization of wastes from yeast industry. *Proc. IV, Int. Cong. Food Sci. Technol., Madrid* **4** : 306.
- Rich, L. G. (1986). Improved lagoon performance through retention time control. Public Works 117 (12): 38.

- Sachiz, R., Corboda, P. and Sinoriz, F. (1985). Use of UASB reactor for the anaerobic treatment of stillage from sugarcane molasses. *Biotechnol. Bioengg.* **27** : 1710.
- Sahai, R., Jabeen, S. and Saxena, P. K. (1983). Effect of distillery waste on seed germination, seedling growth and pigment content of rice. *Indian J. Ecol.* **10** (1) : 7-10.
- Sahm, H. (1984). Anaerobic waste water treatment. Advances Biochemical Engg. Biotechnol. 28:83.
- Sharma, Vinod, Sharma, Rajeev, Sharma, K. D., Sharma, V. and Sharma, R. (2002). Distillery effluent effect on seed germination, early seeding growth and pigment content of sugarbeet (*Beta vulgaris* Linn. Var.mezzanau-poly). *J. Environ. Bio.* 23 : 77-80.
- Singh, A. B., Biswas, Sivockoit, Ramana, R. (2003). Effect of distillery effluents on plants and soil enzymatic activities and groundnut quality. *J. Plant Nutrition and Soil Sci.* **166** (3) : 345-347.
- Singh, D., Dahiya, J. S., Nigam, P. and Marchant, R. (1995). Decolourization of molasses waste water by *Pseudomonas* fluorescens. Gene. **60** : 2253.
- Singh, P. K., Sharma, K. P., Sharma, S., Swami, R. C. and Sharma, Subhasini (2010). Polishing of biomethanated spent wash (primary treated) in constructed wetland: A bench study. *Indian J. Biotechnol.* **9** : 313-318.
- Singh, Y. and Bahadur, R. (1995). Germination of field crop seeds in distillery effluent. Indian J. Ecol. 22: 82-85.
- Singh Y. and R. Bahadur. (1997). Effect of distillery effluent irrigation on maize grain crop and soil fertility. *Indian J. Ecol.* **24** : 53-59
- Somawanshi, R. B. and Yadav, A. M. (1990). Effect of spent wash on soil chemical properties and composition of leachate. Agricultural papers of 14th Annual convention of the Deccan Sugar Technologist Association **1** : 101-108.
- Speece, R. E. (1983). Anaerobic biotechnology for industrial waste water treatment. Environ. Sci. Technol. 17: 416.
- Sukanya, T. S. and Meli, S. S. (2004a). Effect of distillery effluent irrigation on growth, yield and quality of maize grown on sandy loam in northern transitional zone of Karnataka. *Karnataka J. Agri. Sci.* **17** (3) : 405-409.
- Sukanya, T. S. and Meli, S. S. (2004b). Conjunctive use of liquid distillery effluent (spentwash) and water on maize yield and soil properties. *Crop Research*. **27** (2/3): 182-187.
- Sukanya, T. S. and Meli, S. S. (2004c). Response of wheat to graded dilution of liquid distillery effluent (spent wash) on plant nutrient contents, nutrient uptake, crop yield and residual soil fertility. *Karnataka J. Agri. Sci.* **17** (3) : 417-420.
- Sukanya, T. S. and Meli, S. S. (2004d). Distillery effluent as a nitrogen source on maize yield and soil properties. *Karnataka J. Agri. Sci.* **17** (3) : 421-427.
- Sukanya, T. S. and Meli, S. S. (2004e). Effect of distillery effluent as nitrogen source on wheat yield and soil properties. *Karnataka J. Agri. Sci.* **17** (3) : 428-433.
- Sukanya, T. S. and Meli, S. S. (2005). Soil fertility status as affected by use of distillery effluent as irrigation water in wheat. *Mysore J. Agri. Sci.* **39** (2):151-157.
- Wedzicha, B. L. and Kaputo, M. T. (1992). Melanoidins from glucose and glycine: composition characteristics and reactivity towards sulphite ion. *Food Chem.* **43** : 359-367.
- Zalawadia, N. M. and Raman, S. J. (1994). Effect of distillery waste water with graded fertilizer levels on sorghum yield and soil properties. *J. Indian Soc. Soil Sci.* **42** : 575-579.
- Zalawadia, N. M., Raman, S. and Patil, R. G. (1997). Influence of diluted spent wash of sugar industries application on yield and uptake by sugarcane and changes in soil properties. *J. Indian Soc. Soil Sci.* **45** (4) : 767-769.