

Time Efficient Co-composting of Water Hyacinth and Industrial Wastes by Microbial Degradation and Subsequent Vermicomposting

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Rec date: Apr 01, 2014, Acc date: Apr 29, 2014, Pub date: May 03, 2014

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Abstract

Time efficient co-composting of distillery and paint sludge industrial wastes with 2:1:1 ratio of water hyacinth, cow-dung and saw-dust in combination with *Trichoderma viride*, *Phanerochaete chrysosporium*, *Bacillus cereus* and earthworms was done. The decrease in MC, TOC and C:N ratio and increase in temperature, TKN, pH, TK, TP was significant. Out of five treatments, DS3 (60% distillery sludge) and DS4 (80% distillery sludge) treatments were stable and matured with C:N ratio of 18.68 ± 1.1 and 14.73 ± 1.12 respectively. Maximum earthworm numbers (26) were obtained in DS1 substrate and maximum cocoons (10) were counted in PS1 substrate. 20% (w/w) vermicompost mixed soil induced the growth in root length (4.6 ± 0.21), shoot length (5.4 ± 0.10) and 90% GI while >20% (w/w) have inhibitory effects on plant growth. Therefore these treatments can be applied in agronomic field as a soil amendment.

Keywords: Vermicompost; *Trichoderma viride*; *Phanerochaete chrysosporium*; *Bacillus cereus*; Earthworms

Introduction

Pollution is one of the major problems of 21st century. Due to ever increasing population and rapid rate of industrialization, has increased the problem of management of solid wastes [1]. The disposal of large amount of waste water and sludge produced by humans is becoming a serious problem of the world [2]. The problem of safe disposal of the sludge generated in large quantities has a negative impact on the society. Sludge is an unavoidable, hazardous and odorous by-product of conventional waste-water treatment plants which further requires safe disposal either by incineration or using landfills incurring large amount of money [3]. But today we are facing the major problems of agricultural wastes, municipal solid wastes, poultry wastes etc. as they are major contaminants of air, water and soil.

Distillery sludge is the waste from the distillery industries which are the most important economic resources for the soil fertility through improvement in soil water-holding capacity, nutrients retention, roots penetration, and soil acidity reduction [4-6]. Currently, with the ever increasing number of sugar mills and distillery units, it has now become compulsory to use the distillery sludge as a fertilizer to increase the fertility of the soil. However, distillery sludge application in soil results in environmental pollution problem [7] because apart from organic content and nutrients, sludge also contains toxic compounds, heavy metals etc. [8]. Thus, the microorganisms in the soil may be affected by the variation in soil temperature, pH, nutrient status, heavy metals, oxygen levels, which in turn can have severe effect on the ecological processes linked with nutrients cycling [9].

Paint sludge is the waste from the paint industries. The hazardous waste stream generation is estimated to be 480 million pounds per year. Paint Sludge Removal System has become very important because all manufactured goods purchased are painted. There are two

types of paint used, wet paint which is solvent-borne or water-borne and powder coat which is fluidized solid. Removing over spray through Paint Sludge Removal System is very important during manufacturing because letting out the paint directly to the atmosphere is very hazardous and cause environmental damage. Through Paint Sludge Removal System the paint that is over sprayed during the manufacturing process gets collected in the paint booth walls, on the floors of the spray booth and also on spray painting equipment. Cleaning this over sprayed paint is a very difficult task and requires extensive time to complete. The paint over spray has to be chemically treated and sent to paint sedimentation tank [10].

Reuse and recycling activities of heavy metals can reduce the demand for raw materials and energy while minimizing the impact of waste disposal [11]. Although the method of incineration is rather progressive (most of the hazardous components are transformed into less dangerous substances, combined with volume reduction), but there are some harmful side effects such as sulphur and nitrogen oxides, hydrogen chloride, dioxins, heavy metals compounds, ash and other materials are generated which causes groundwater pollution and contamination [12]. Landfills are intended to minimize the negative impact of waste on the environment, they have harmful effects on nature due to the leaching of chemicals into soil and groundwater [13].

Composting is a natural process which involves the aerobic biological decomposition of organic matter from the biodegradable wastes using microbes under controlled conditions, resulting in a final product containing stabilized carbon, nitrogen and other nutrients in the organic fraction, the stability depending on the compost maturity [14-16]. Biodegradable waste is composted with the objective of returning the waste to the plant production cycle as fertilizer and soil improver [17]. Composting helps to optimize nutrient management and the land application of compost may contribute to combat soil organic matter decline and soil erosion [18]. Furthermore, it may partially replace peat and fertilizers [19]. Composting of vegetable waste produced in horticulture may reduce the environmental impact

on climate change, at a rate of about 40% and 70% respectively, compared to land filling and incineration [20]. Composting removes biodegradable wastes that otherwise, produce germs of diseases and cause serious hazards. Composting can be used to produce useful manures, which when mixed properly in soil, increase production of different types of crops. Composting of wastes puts back the nutrients into the soil through recycling them and also reduces the expenses of farmers on raising crop-production through purchase of synthetic fertilizers [21].

Vermicomposting is a simple biotechnological process of composting, in which certain species of earthworms are used to enhance the process of waste conversion and produce a better product. Vermicomposting differs from composting in several ways [22]. It is a mesophilic process that utilizes microorganisms and earthworms that are active at 10°C to 32°C (not ambient temperature but temperature within the pile of moist organic material). The process is faster than composting; because the material passes through the earthworm gut, a significant but not fully understood transformation takes place, whereby the resulting earthworm castings (worm manure) are rich in microbial activity and plant growth regulators, and fortified with pest repellence attributes as well. In short, earthworms through a type of biological alchemy are capable of transforming garbage into “gold” [23].

Nadaf and Ghosh investigated the biodegradation of a paper and pulp effluent by *Rhodococcus sp.* NCIM 2891 and its toxicity assessments [24]. Kaushik et al. emphasized on vermicomposting as an eco-friendly option for fermentation and dye decolorization waste disposal [25]. Singh and Sharma shown significant improvement in the quality of vermicompost produced by pre-decomposing the wastes with microbes before vermicomposting [26]. The production of cellulose, hemicellulose, and lignin degrading enzymes by the inoculated microbes during pre-decomposition might have increased the degradation process. Araujo and Monteiro [27] have demonstrated that application of composted textile sludge increased significantly the microbial biomass and activity, and bacteria number of soil as compared to the untreated sludge. Solid textile mill sludge mixed with cow dung (CD) can be used as a substrate for vermicomposting [28]. Several earthworm species have been tested for vermicomposting process; however, a large number of reports are based on the utilization of *E. fetida* for vermicomposting [29-31].

Water hyacinth (*Eichhornia crassipes*) is a highly competitive plant that is capable of rapid growth and spread. It can displace native species, reduce biodiversity, limit recreation, diminish aesthetic value and decrease water quality and flow. Water hyacinth is a good source of cellulose and hemicellulose, which can be converted to biogas. Lu et al. showed that a water hyacinth system was effective in treating wastewater from an intensive duck farm [32]. Song et al. found that water hyacinth can potentially remove nitrogen and phosphorus in sewage [33]. Zheng et al. found that the contents of nitrogen, phosphorus and potassium in water hyacinth to be 3.07%, 0.46%, and 5.70%, respectively [34]. Water hyacinth can also effectively absorb some metals like arsenic, Se(VI), Cu(II), Cd (II), Cr(VI), Ni(II) [35,36]. Water hyacinth and saw dust are very useful and effective bulking agents for composting with paint/distillery sludge.

The present study mainly focused on the application of combine composting and vermicomposting for management of weeds and industrial wastes with microbes and bulking agent to reduce the time duration of composting and better productivity. The objectives of this study was to assess the physicochemical and biological parameters to

determine the stability and maturity of compost prepared by mixing cow dung and water hyacinth with industrial wastes.

Materials and Methods

Materials

Microbial source

The bacterial strain *Bacillus cereus* (MTCC 4079) and the fungus strains *Trichoderma viride* (MTCC 800), *Phanerochaete chrysosporium* (MTCC 787) were procured from IMTECH, Chandigarh, India. The fungi *Trichoderma viride* were maintained by subculturing on Malt extract agar and *Phanerochaete chrysosporium* on Malt extract agar with glucose and peptone whereas bacteria was subcultured on Luria Bertani agar medium.

Industrial wastes (IW)

Distillery sludge (IW1) was obtained from Hindustan Distillery, Pilukhedi, Bhopal, Paint industry sludge (IW2) was taken from Max coat Paint Industry, Indore. The source of saw dust was saw mill, Bhopal.

Cow dung, water hyacinth and earthworm

Water Hyacinth (WH) was collected from the Lake situated in Bhopal. Cow dung (CD) was procured from a farm at Gandhi Nagar, Bhopal. Earthworms (*Perionyx excavates*) were obtained from Plant tissue culture training centre, Bhopal.

Methods

Set-up of experiment

The water hyacinth, cow dung and sawdust were mixed in 2:1:1 ratio. Each industrial waste (IW1, IW2) was added to the mixture of water hyacinth and cow dung (Table 1). Five kilogram of mixture (dry weight) was put in each rectangular pit (1 m length×1 m width×1 m height). A combination of pure culture of *Trichoderma viride*, *Phanerochaete chrysosporium* and *Bacillus cereus* was inoculated in all the treatments on first day of composting [37,15]. Temperature was maintained around 26°C by adding water when necessary. For mesophilic aerobic digestion turning was done manually after every 4 days. The mixture was allowed to pre-decompose in triplicates for duration of 20 days.

Vermicomposting

For subsequent Vermicomposting of the pre-decomposed waste, 20 weighed adult and healthy earthworms (*Perionyx excavates*) was introduced in each pit (0th, 20th and 40th days) and was allowed for biodegradation for a period of 20 days. Parameters like temperature and humidity was maintained during the process. Stability and maturity analysis of vermicompost was done after 20 days using physicochemical parameters. If the compost is stable and mature, the analysis of earthworm's mortality, Gross Biomass and Total number of cocoons production was carried out or else the compost was allowed to degrade for few more days until the compost is matured [38]. Growth and cocoons production of earthworms in each pit was measured on the completion of vermicomposting. Earthworms and cocoons produced during experiment were separated from the

substrate material by hand sorting, after which earthworms was washed in tap water to remove the adhering material from their body and subsequently weighed on a live weight basis. Cocoons were separate and count and introduce in the same way in which their parent was reared. Total number of dead earthworms was counted and mortality was measured [28].

Treatments	Constituents
Control	Water hyacinth + cow dung + saw dust (2:1:1) (100%)
DS1	Distillery sludge (20%) + Water hyacinth + cow dung + saw dust (2:1:1) (80%)
DS2	Distillery sludge (40%) + Water hyacinth + cow dung + saw dust (2:1:1) (60%)
DS3	Distillery sludge (60%) + Water hyacinth + cow dung + saw dust (2:1:1) (40%)
DS4	Distillery sludge (80%) + Water hyacinth + cow dung + saw dust (2:1:1) (20%)
DS5	Distillery sludge (100%)
PS1	Paint sludge (20%) + Water hyacinth + cow dung + saw dust (2:1:1) (80%)
PS2	Paint sludge (40%) + Water hyacinth + cow dung + saw dust (2:1:1) (60%)
PS3	Paint sludge (60%) + Water hyacinth + cow dung + saw dust (2:1:1) (40%)
PS4	Paint sludge (80%) + Water hyacinth + cow dung + saw dust (2:1:1) (20%)
PS5	Paint sludge (100%)

Table 1: Substrates composition

Sampling of compost

Homogenized feed material samples were drawn at 0, 20 and 40 days from each pit. The 0th day refers to the time of initial mixing of the industrial wastes (IW) with water hyacinth and cow dung before preliminary decomposition.

Analysis of compost

The temperature was measure daily by placing thermometer through the plastic pipe. The pH of the compost was measured in distilled water with 1:10 (w/v) compost: water ratio. The moisture content was measured by drying in an oven at 105°C to constant weight. Total Organic Carbon (TOC) and Total Kjeldahl Nitrogen (TKN) of the pre-decomposed residue and the vermicompost were measured with the Walkely and Black [39]. Rapid titration method (1934) and Micro-Kjeldahl method [40] respectively. Total phosphorus (TP) was determined spectrophotometrically [41]. Total

potassium (TK) was detected by the flame emission technique by Flame photometer [42].

Seed germination test

Seed germination test was performed to compare the efficiency of compost with normal soil. Soybean seeds (*Glycine max*) were used for the test. Germination temperature ranges was from 22-50°C for it. Plastic containers with lids were filled with mixture of compost and garden soil in the concentration of 0, 20, 40, 60, 80 and 100 %. Container filled only with garden soil was serving as control. Cool tap water was added to maintain the moisture content. Excess water was poured off. The soaked and sterile 10 seeds were placed to the depth of 2 cm and about 2 cm apart. Incubation was given at room temperature and moisture content was maintained by adding water at constant time interval. Length of “radicle” was observed after 7 days incubation, duration for seed germination and radical length was compared to determine the best test sample [43].

Statistical analysis

Results were mean of triplicates. One way ANOVA was done using the SIGMASTAT programme. The objective of statistical analysis was to determine any significant differences among the parameters analyzed in different treatments during the composting and vermicomposting process [37].

Results and Discussion

Physico-chemical parameter analysis of substrates

To investigate the compost stabilizing characteristic of water hyacinth, the physico-chemical parameters like pH, TOC, TKN, TK, TP and C: N were analysed for all the initial substrates (IW1, IW2, WH, CD and SD) using standard protocols, so that the nutritive value of each substrate could be evaluated and compared. The details of the parameters evaluated of the initial feed used for composting have been shown in Table 2.

The determination of chemical parameters of the initial feed revealed that Paint sludge possessed higher percentage of TOC (49.3%), TKN (0.31%), TP (0.01%) and TK (0.20%) and C:N ratio (159.21) which clearly indicates that carbon to nitrogen ratio is very high. But with the co-composting with water hyacinth, cow dung and saw dust it can be maintained. Water hyacinth substrate shown very good values of C:N ratio (48.39) which indicates that it was nutrient rich substrate essential for feedstock to be decomposed. Distillery sludge was also having good chemical properties for composting C:N ratio (54.83), TP (0.49) and TK (0.48). Cow dung and saw dust was having C:N ratio (72.1) and (249.42). C:N ratio of saw dust was high but it is proved in different composting studies that cow dung and saw dust are very useful and effective bulking agents for composting.

Parameters	Distillery sludge (IW1)	Paint sludge (IW2)	Saw dust (SD)	Water hyacinth (WH)	Cow dung (CD)
pH	8.30 ± 0.01	8.40 ± 0.01	7.5 ± 0.20	7.3 ± 0.20	8.7 ± 0.20
Total organic carbon (TOC)	37.29 ± 0.01	49.31 ± 0.02	42.10 ± 0.21	41.7 ± 0.10	38.2 ± 0.21

Total Kjeldahl nitrogen (TKN)	0.68 ± 0.12	0.31 ± 0.15	0.16 ± 0.03	0.87 ± 0.05	0.53 ± 0.02
C: N ratio	54.83 ± 1.0	159.21 ± 1.10	249.42 ± 1.12	48.39 ± 1.01	72.1 ± 1.05
Total phosphorus (TP)	0.49 ± 0.01	0.01 ± 0.0	0.53 ± 0.02	0.83 ± 0.13	0.58 ± 0.14
Total potassium (TK)	0.48 ± 0.20	0.20 ± 0.18	1.73 ± 0.10	0.84 ± 0.05	0.61 ± 0.12

Table 2: Physico-chemical characteristics of the substrates; All values are mean ($n=3$) ± SD and given in percentage; *Significant ($P<0.001$)

Physical parameter analysis during decomposition

After the analysis of physico-chemical parameters of all the substrates, the industrial wastes were mixed in various combinations (20-100%) with water hyacinth, cow dung and saw dust and then subjected to 20 days composting (with the inoculation of microbial enriched culture).

Temperature (°C)

An initial temperature of 26 to 29.5°C on 5th day and final temperature of 47 to 57°C on 20th day (significant at $P<0.001$) was recorded in all the runs of different industrial waste composting (Figure 1). The increase in temperature in the early phase of composting can be attributed to the thermophilic phase of composting. The temperature rises as a consequence of the rapid breakdown of organic matter, mostly due to the presence of readily available substrates rich in carbon, by microbial metabolism [44]. Although studies of Chao-Ming Lai in 2010, suggest that in the thermophilic phase the temperature could raise to 60°C, in this study final increase in temperature of (57°C) was observed as comparable to control (55°C) after 20 days. This could be recognized due to the difference of material used and the different types of composting process adopted. The gradual increase in temperature during composting is indicative of the microbial action as a result of the inoculation of microbial enriched culture (*Trichoderma viride*, *Phanerochaete chrysosporium* and *Bacillus cereus*) in each treatment at 0th day.

pH

The pH value of compost is looked as an indicator of process of decomposition and stabilization. The change in pH value during composting is quite predictable [45], it drops slightly early in processing due to the production of organic acids derived from the intense fermentation of carbohydrates, and then rises rapidly to approximately 8.5 because of ammonification. The pH value test of compost before application is necessary since high pH (>8.5) value causes ammonia volatilization, odours production and plant root damage. The pH of the initial mixed wastes ranged 6 to 8.7, and for the composted product on 20th day ranged 6.9 to 7.6 in all the runs of both industrial wastes with different composition (Figure 2).

The final pH on the 40th day of vermicomposting ranges from 6.9 to 7.6 in all the runs which is a sign of compost stabilization as the pH value settles between 7.0-8.0 as the compost stabilizes. Sanchez-Monedero [46] found that pH value varies within 7.0-8.0 in 4 composting mixtures (wide range feedstock) in 20 weeks. Garg and Gupta [47] observed pH in the range of 7.7 ± 0.1 - 8.0 ± 0.1 in final vermicompost.

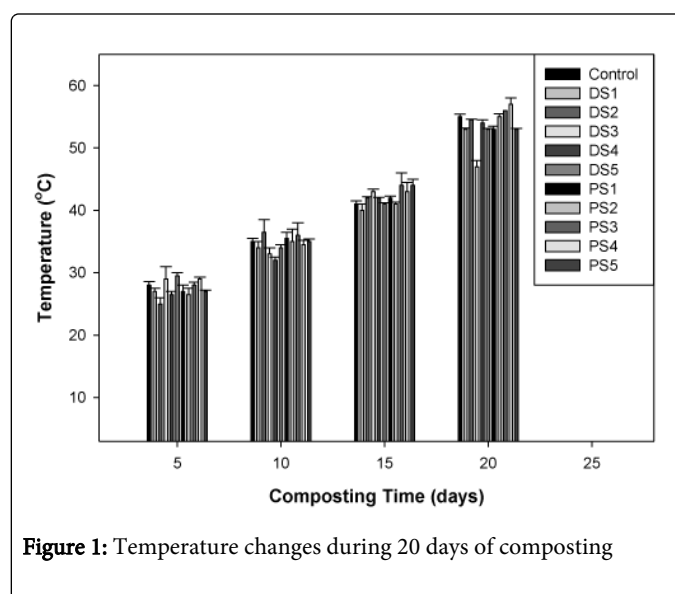


Figure 1: Temperature changes during 20 days of composting

The results obtained were in accordance with the findings of other studies of Patidar et al. [37] and Suthar et al. [48]. At the 20th day of composting period, the pH values increased from the initial values recorded at 0th day and the maximum pH value observed were 8.7 in control. The rise in pH values result from the release of ammonia due to the start of proteolytic process [49]. During the cooling down and maturation stages, the pH drops to neutral-slightly alkaline value [50] as observed in the present study at 40th day. All the values were found to be significant at $P<0.001$.

The pH of composts of Distillery sludge and Paint sludge varied from slightly alkaline to neutral after vermicomposting which is within the range of optimal pH for plant growth. The pH shift has been reported due to mineralization of the nitrogen and phosphorus into nitrites/nitrates and orthophosphates [51] and decrease in pH was due to production of acids during the decomposition process by microbial metabolism [52]. The combined effect of these two oppositely charged groups actually regulates the pH of compost leading to a shift of pH towards neutrality. Moreover, Gupta and Garg [53] pointed out that the shift in pH during vermicomposting is substrate-specific, as different substrates could result in the production of different intermediate species.

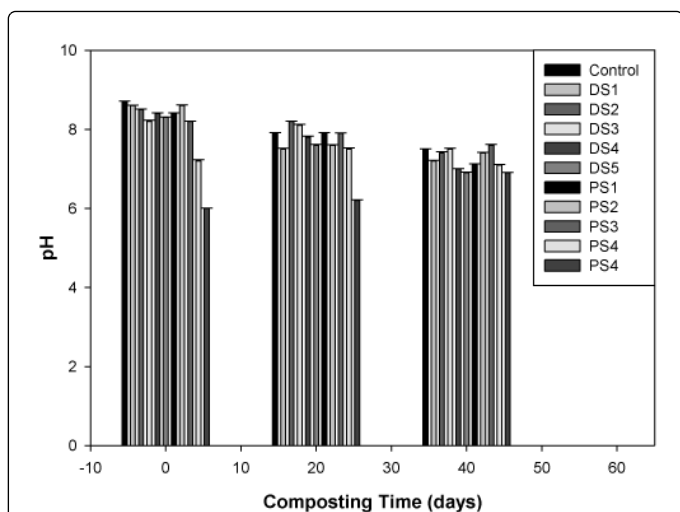


Figure 2: Changes in pH during composting and vermicomposting after 0, 20 and 40 days

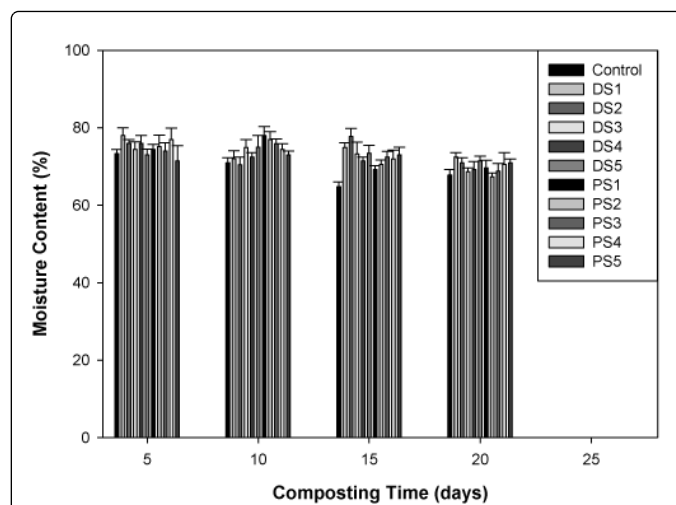


Figure 3: Changes in moisture content (%) during composting 0 to 20 days

% Moisture content

Moisture content is an important factor in degradation because it influences the structural and thermal properties of the material, as well as the rate of degradation and metabolic process of microorganisms. Previous results indicated that 1 g of organic matter releases about 25 kJ of heat energy which is sufficient to vaporize 10.2 g of water [54]. The moisture content becomes a limiting factor and the rate of decomposition decreases rapidly when the moisture content decreases below 45% to 50%. Moisture content during the 20 days composting was calculated (Figure 3) and was controlled directly by adding water during 20 days vermicomposting to maintain moisture around 60%. Initial % moisture content in all the composting treatments was in the range of 71-78%. On the 20th day of microbial composting the final moisture content was in the range of 67.35-71.94% which shows that heat generated during the degrading process helps in reducing the moisture content.

Chemical parameter analysis during decomposition

Total organic carbon

Typically, carbon provides the preliminary energy source for the microorganism's growth. Especially, bacteria, actinomycetes, and fungi need both carbon and nitrogen to grow. The breakdown of organic matter is a dynamic process achieved by microorganisms, when each group of microorganisms reaches its peak population at the optimum condition for microorganism activity. The organic matter such as protein, cellulose and hemicelluloses are easily degradable [55] by microorganisms.

Total organic carbon during the process was determined using Walkey and Black's Rapid Titration method. Data in Table 3 showed a significant decrease in total organic carbon in all the treatments of five different compositions of both industrial wastes during composting and vermicomposting.

Substrate	0 th day	20 th day	40 th day
Control	40.23 ± 0.21 [*]	31.42 ± 0.30 [*]	28.21 ± 0.20 [*]
DS1	43.65 ± 0.10 [*]	35.51 ± 0.05 [*]	31.37 ± 0.04 [*]
DS2	47.28 ± 0.10 [*]	39.25 ± 0.20 [*]	34.49 ± 0.04 [*]
DS3	49.62 ± 0.25 [*]	41.61 ± 0.10 [*]	39.61 ± 0.10 [*]
DS4	36.13 ± 0.21 [*]	35.29 ± 0.15 [*]	33.29 ± 0.02 [*]
DS5	37.29 ± 0.02 [*]	32.37 ± 0.02 [*]	30.39 ± 0.20 [*]
PS1	56.92 ± 0.10 [*]	48.31 ± 0.31 [*]	42.25 ± 0.01 [*]
PS2	59.76 ± 0.10 [*]	52.19 ± 0.01 [*]	49.22 ± 0.10 [*]
PS3	68.65 ± 0.03 [*]	62.65 ± 0.03 [*]	56.31 ± 0.20 [*]
PS4	45.43 ± 0.10 [*]	41.28 ± 0.20 [*]	36.12 ± 0.03 [*]
PS5	49.31 ± 0.20 [*]	43.37 ± 0.03 [*]	38.05 ± 0.15 [*]

Table 3: Total Organic Carbon (%TOC) during composting and vermicomposting; All values are mean (n=3) ± S.D. and given in percentage; ^{*}Significant (P<0.001)

The best results were obtained in DS1, DS2 and PS3 treatments and also in control. Decline in TOC during composting was expected due to degradation of more labile fractions of the organic matter by microbes [56]. As the waste concentration increased above 20% in the feed mixture of cow dung and water hyacinth, the loss in TOC content decreased as compared to control during composting and vermicomposting process.

During composting and subsequent vermicomposting of distillery sludge and paint sludge mixed with 2:1:1 ratio of water hyacinth, cow dung and saw dust in five different proportions, total organic carbon was decreased significantly in all the treatments on 20th and 40th day (Figure 4). The maximum percent loss in TOC was 13.16% noted in DS2 treatment as compared to control in which 12.02% TOC was noted. More pronounced loss of 12.28% was seen in DS1 treatment

and as the TOC was decreased in subsequent treatments. Significant percent loss was observed in TOC during the distillery sludge and paint sludge composting on 20th day and vermicomposting on 40th day due to degradation of more labile fractions of the organic matter by inoculated micro-organisms [57].

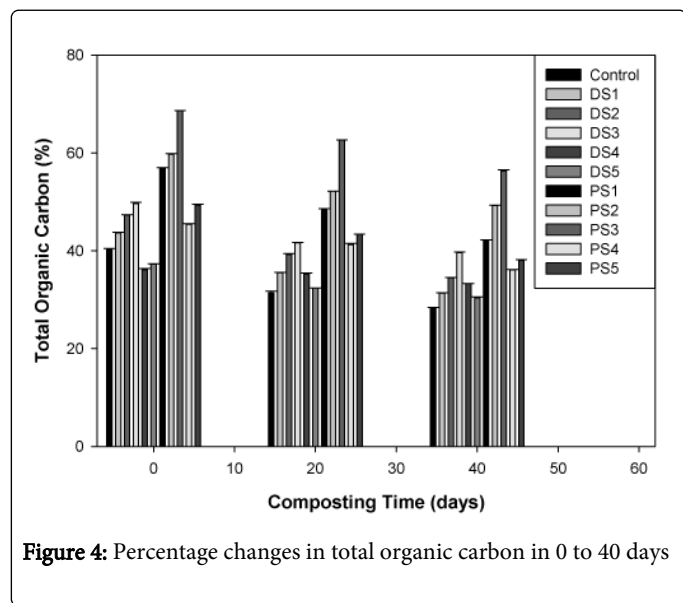


Figure 4: Percentage changes in total organic carbon in 0 to 40 days

Total kjeldhal nitrogen

Nitrogen is an important nutrient for composting process since the quantity of nitrogen determines the microorganism population growth. During composting process, microorganisms oxidize organic matters, and release essential minerals for plants such as nitrogen, phosphorus, and sulphur. Therefore, amount of nitrogen increases during the process. Total Kjeldhal nitrogen (TKN) was determined using Macro Kjeldhal method. Data on the percentage of TKN during microbial composting and subsequent vermicomposting. During initial composting at 20 days, the nitrogen content increased in all treatments. The initial TKN content of the different treatments was in the range of 0.31-1.57%. After 20 days of composting TKN was in the range of 0.64-1.9%. Final TKN on 40th day in vermicomposts was in the range of 0.39 to 2.26% after vermicomposting [58].

Final TKN content in DS2, DS3 and DS4 is maximum as compared to other treatments and control. Similar observations have also been reported by other workers [59,60]. Kaushik and Garg [43] have observed 2.0-3.2 fold increase in TKN in textile mill sludge vermicomposting. It has also been suggested that the final nitrogen content of vermicompost is dependent on the initial nitrogen content present in the waste and the extent of decomposition [61]. During vermicomposting there was slight increase in TKN in all treatments and significant difference was noted in DS3 and DS4 as compared to control (Figure 5). In different studies subsequent vermicomposting results in decrement in TOC which may have been due to ammonification, NH₃ volatilization and denitrification [14, 62].

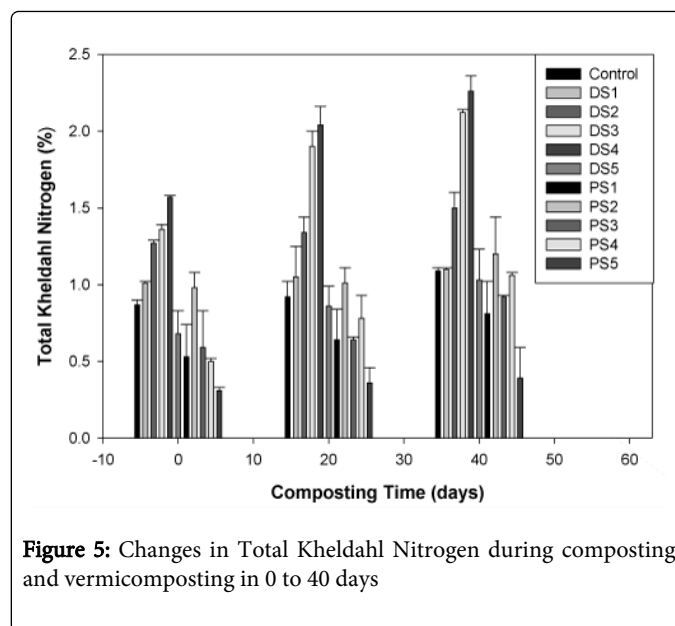


Figure 5: Changes in Total Kjeldahl Nitrogen during composting and vermicomposting in 0 to 40 days

C: N ratio

The C:N ratio of a substrate material reflects the organic waste mineralization and stabilization during the process of composting and vermicomposting. The loss of carbon as CO₂ through microbial respiration and simultaneously addition of nitrogen by worms in the form of Mucus and nitrogenous excretory material lowers the C:N ratio of the substrate [57]. C:N ratio is considered as a parameter to establish the degree of maturity of compost and its agronomic quality [63]. Decline of C:N ratio to less than 20 indicates an advanced degree of organic matter stabilization and reflects a satisfactory degree of maturity of organic wastes [64].

It is evident that C:N ratios decreased with time in all treatments after 20 days of microbial inoculated composting and at the end of vermicomposting period. Initial C: N ratio was in the range of 23-159 before the inoculation of microbes in all the feed mixture and 46 in control (at 0th day). C:N ratios after composting on 20th day were in the range of 17 to 120 and 34 in control. Final C: N ratios were in the range of 14-97 and 25 in control after vermicomposting. The decreasing trend of C:N ratio during vermicomposting of distillery sludge is in good agreement with other authors [53,65].

During composting C:N ratio decreases in all treatments as well as in control (Figure 6). Max percent decrement was noted in paint sludge. The C:N ratio decreased during vermicomposting in less percentage as compared to composting. The relevance of the C: N ratio relies on the fact that a decrease in the ratio implies an increase in the degree of humification of organic matter. There were decrease of C:N ratio in all the treatments but best ratio were found in DS3 and DS4. Gupta and Garg [47] observed final C:N ratios of paper waste vermicompost in the range of 11.3-35.6 and the ratio was higher in those feed mixtures which had higher percentage of paper waste. Our findings corroborate with the findings of other studies [53,66]. DS3 and DS4 showed maximum degradation and so biological parameter of germination index were carried out for only these two treatments.

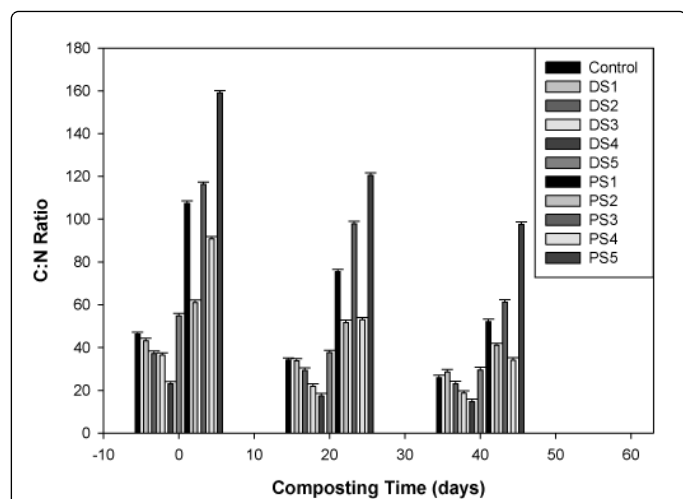


Figure 6: Variations in C: N ratio during composting and vermicomposting in 0 to 40 days

Total phosphorus

In this process Total phosphorus (TP) was determined using spectrophotometer.

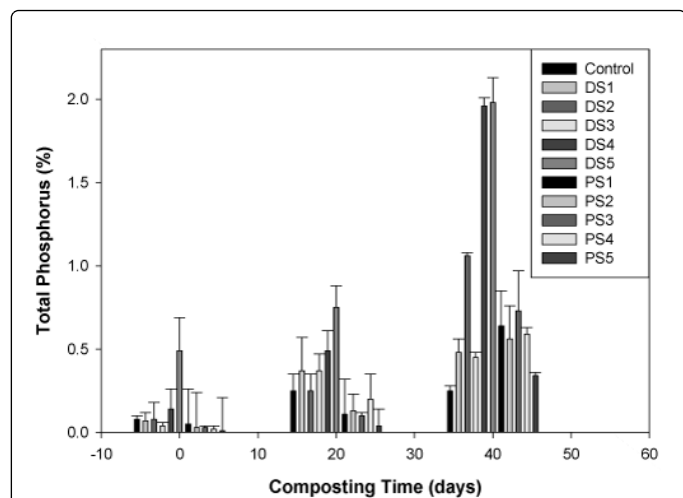


Figure 7: Changes in total phosphorus during composting and vermicomposting in 0 to 40 days

Percentage of total phosphorus increased in all five treatments during composting and vermicomposting of distillery sludge (Figure 7). The overall increase in TP content was maximum in DS2, DS4 and DS and minimum increase was in PS5. According to Lee [66] if the organic materials pass through the gut of earthworms then unavailable forms of phosphorus are being converted to such forms that are available to plants.

Percent increase in total phosphorus was noted in distillery sludge and paint sludge degradation (Figure 7). Total Phosphorus increment in distillery sludge was more than the paint sludge compost. Singh and Sharma [14] also reported rapid decomposition of wheat straw with a mixture of cellulolytic fungi, *P. sajorcaju*, *Trichoderma reesei*, *A. niger*

along with nitrogen fixing bacteria *Azotobacter chroococcum*. The increasing trend in TP content during vermicomposting is consistent with the findings of other researchers [53,58,65].

Total Potassium

Fundamentally, potassium is a macronutrient in plants and animals. Recent studies have proved that potassium can increase the plant height, fresh plant weight and also increase herbage and oil yield on the patchouli [65]. Total potassium (TK) was determined using flame photometry. It is reported that total potassium during microbial composting increased significantly in all the treatments. Out of five treatments of three waste maximum increments in TK was noted in DS1, DS2, PS4 and control. This increment in potassium was due to acid produced by the microorganisms which plays major role in solubilisation of insoluble potassium [68]. It has been suggested that earthworm processed residue contains higher concentration of exchangeable K due to enhanced microbial activity during vermicomposting, which consequently enhances the rate of mineralization [68].

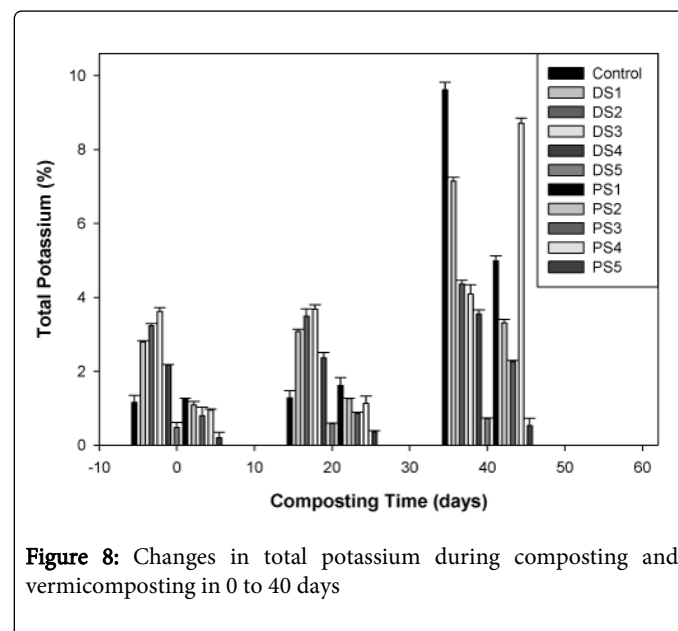


Figure 8: Changes in total potassium during composting and vermicomposting in 0 to 40 days

During composting of distillery sludge percent increase in total potassium was noted maximum in all five treatments after 20th day (Figure 8). Maximum increment was noted in DS2 after vermicomposting. This increment in potassium was due to acid produced by the microorganisms which plays major role in solubilisation of insoluble potassium [58]. However, there are no decrease in potassium during vermicomposting was observed.

Total no. of earthworms, cocoons and biomass

In vermicomposting process, earthworms mineralize the organic matter, converting a part of it in worm biomass and respiratory products, and, the rest is converted as nutrient rich vermicompost. The appropriate feed composition for the earthworms could optimize the manual value of the vermicompost [66]. Suthar et al. [67] emphasized that in addition to the biochemical properties of waste, the microbial biomass and decomposition activities during vermicomposting are also important in determining the worm biomass and cocoon production.

A noticeable increase in the number of earthworms as well as the cocoons was observed during vermicomposting. Various studies have shown that earthworm utilize micro-organisms in their substrates as a food source and can digest them selectively [53]. Data suggested that maximum number of earthworm increases in DS1, DS2, DS3, DS4 and DS5 well as in control but as the concentration of waste increase, number was significantly decrease in all the five different trials of three waste containing different waste composition (Figure 9). There was less number of earthworms recorded in paint sludge vermicomposting.

Seed germination test

The evaluation of industrial wastes toxicity by chemical characterization and biological testing is extremely important for screening the suitability of vermicompost for land application. To evaluate the biological parameter of vermicompost, the amendments of most stable and matured compost on the basis of C: N ratio less than 20 i.e. treatment DS3 and DS4 was mixed with garden soil at different concentrations (0, 20, 40, 60, 80 and 100%). The soaked and sterile ten seeds of Soybean (Glycine max) were evenly sown in each pot to a depth of 2 inch watered daily till seed germination.

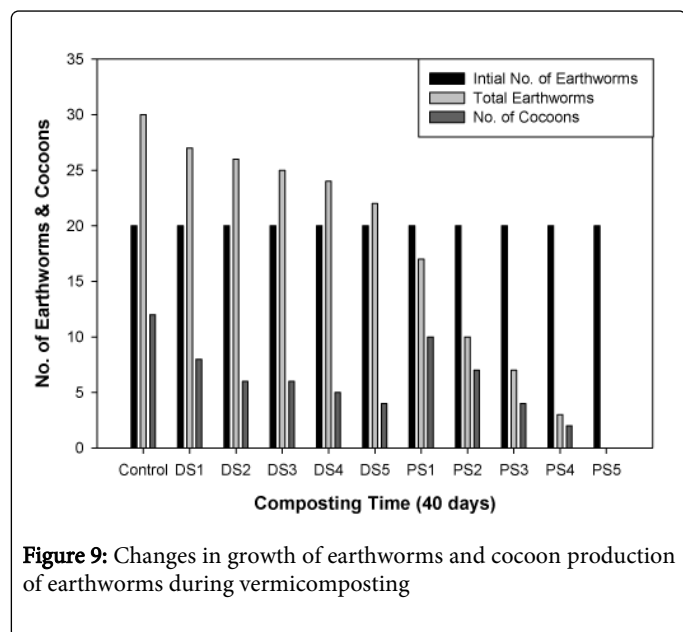


Figure 9: Changes in growth of earthworms and cocoon production of earthworms during vermicomposting

In general, percentage of seed germination decreased with increase in vermicompost concentrations in all the trials of distillery sludge (Table 4). In present study, it was observed that up to 20% (w/w) concentration of vermicompost has no inhibitory effect on seed germination while at higher concentration (>40%), decrease in percent germination was recorded as 40, 60, 80, and 100% (w/w) concentration of vermicompost has shown seed germination in the range of 86-96%, 70-86%, 60-70% and 50-60% respectively in all the trials of DS3 and DS4. This decrease in percent germination except in control and 20% vermicompost was due to phytotoxic effect produced by combination of several factors, rather than one [69]. These factors include the presence of heavy metals, ammonia [70] salts and low molecular weight organic acids all of which have been shown to have inhibitory effects.

Hence, the results obtained in the present investigation can be concluded that DS3 and DS4 were most stable and quite significant

during vermicomposting of 20 days. DS3 and DS4 treatments were degraded stabilized during vermicomposting. Therefore these treatments can be applied in field for agronomic use as a soil amendment.

Conclusions

It was concluded that out of five treatments (20-100%) two wastes, DS3 (60% distillery sludge) and DS4 (80% distillery sludge) treatments were stable and mature after combined composting and vermicomposting for a span of 40 days as indicated by reduction in C: N ratio and presence of high nutrient levels, viz., nitrogen (TKN), phosphorus (TP) and potassium (TK). Remaining treatments containing higher percentage of wastes require further days for degradation. For such treatments application of additional bulking agents and multistage microbial inoculation can be carried out in future for the enhancement of degradation process. In the present investigation use of distillery sludge in composting and vermicompost was found cost and time efficient. Paint sludge needed more time to convert into good compost. The C:N ratio of paint sludge compost was also not good for a compost.

Vermicompost concentration (%)	GI (%)	Root length (cm)	Shoot length (cm)
Control (0)	60	4.6 ± 0.30	5.2 ± 0.10*
DS3 (20)	80	4.2 ± 0.31	6.2 ± 0.20*
DS3 (40)	75	4.4 ± 0.21	5.1 ± 0.10*
DS3 (60)	60	3.9 ± 0.10*	4.4 ± 0.12*
DS3 (80)	58	3.8 ± 0.13*	4.4 ± 0.14
DS3 (100)	50	3.4 ± 0.12*	4.2 ± 0.13*
DS4 (20)	90	4.6 ± 0.21	5.4 ± 0.10
DS4 (40)	85	3.4 ± 0.10*	5.8 ± 0.12*
DS4 (60)	80	3.1 ± 0.10*	4.4 ± 0.13
DS4 (80)	70	3.2 ± 0.21*	4.2 ± 0.10
DS4 (100)	60	3.1 ± 0.10*	4.1 ± 0.12*

Table 4: Effect of vermicompost DS3 and DS4 amended soil on germination of soybean seed; All values are mean (n=3) ± S.D.; *Significant (P<0.001)

It further suggests that, wastes cannot be used in composting and vermicomposting in 100% composition but they need some bulking agents like water hyacinth, cow dung and saw dust for making good compost. It was observed that up to 20% (w/w) concentration of the trials of DS3 and DS4 vermicompost has no inhibitory effect on seed germination and helpful in growth and development of plants. Further it was concluded that water hyacinth enhances nutritive value of final compost and if blended in appropriate quantity with industrial wastes it accelerates the degradation process as well the quantity of solid waste dumped in the landfills would be appreciably reduced. Finally the objectives of the study in terms of biodegradation of industrial wastes, time efficient composting and compost stabilizing with the help of microbial inoculation was quite significant.

Acknowledgement

Author would like to thank the Indian institute of Soil Science, Bhopal for help in the compost analysis. With a deep sense of gratitude, I wish to express my sincere thanks to my supervisor, Dr. Archana Tiwari, HOD, School of Biotechnology, RGPV, Bhopal.

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