

## Microbial Count and Succession, Soil Chemical Properties as Affected by Organic Debris Decomposition

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**Abstract:** Soil organic matter has been considered an important indicator of soil quality because it is a nutrient sink and source, not only enhances soil physical and chemical properties and promotes biological activity but also maintains environmental quality. Two laboratory and one greenhouse experiments were conducted to study the decomposition of different organic debris and their effects on soil chemical, microbial count and succession and plant growth. In both the experiments, six treatments; 1-turf (T), 2-mixed leaves (ML), 3-mixed leaves+stem and wood chips(ML+CW), 4-stem and wood chips(CW), 5-mixed leaf and stem of trimmed *Evonymus latifolia* (ELS) and 6-Control (soil only), each with three replications were used. The organic debris used in treatments 2-5 belonged to common trees of city and recreational parks (*Plantanus orientalis*, *Morus alba*, *Acer pseudoplatanus*, *Robinia pseudacacia*, *Populus* spp., *Salix* sp., *Alianthus altissima*, *Catalpa speciosa*, *Fraxinus rotundifolius*) which were mixed on an equal w/w bases and kept between the two soil layers in plastic pots undisturbed (soil: organic debris ratio was 8:1). The results from laboratory experiment showed that the maximum bacterial counts in treatments were T>ELS>ML+CW>ML>CW, whereas, in case of fungal counts the trend was ML+CW>ML>CW>ELS>T. The ratio of bacteria/fungi and bacterial population were found to be similar as T>ELS>ML+CW>ML>CW. Comparing the initial and final ratios of C/N, OM/TN and lignin/TN in treatments, the initial and final lignin/TN ratios found to be similar (CW>ML+CW>ELS>ML>T) compared to fungi/bacteria ratio (CW>ML+CW>ML>ELS>T). In this study, the ratio of lignin/TN in treatments found to be the most suitable ratio to estimate microbial succession. The microbial succession was fungi/bacteria/actinomycetes. Phytotoxins produced by microbes in two months old (under greenhouse) treatments totally inhibited germination of snap dragon. Water extracts prepared at different concentrations from treatments induced poor germination, plumule and radicle growth of wheat.

**Key words:** Organic debris % decomposition % microbial succession % soil

### INTRODUCTION

Minimum time required for an organic substance to undergo decomposition in order to have a stable end product compost is variable. It depends on several factors, including the size of the materials, aeration, moisture, C:N ratio of the materials, heat levels and range during composting.

Using immature compost as a growing media because of high microbial activity in it can cause biological blockage of N from the crop or due to certain phytotoxic chemicals such as short chain fatty acids produced by the microbes can be a real treat for inhibiting seed germination

and deformity or death of plants for composts with a high C:N ratios [1].

A large proportion of the organic carbon residues utilizing by the micro flora consists of simple sugars, their derivatives or their appropriate polymers. Although water-soluble carbohydrates may predominate in young plants, mature plants have a higher percentage of cellulose, hemicellulose and lignin. Barbarika *et al.* [2] found that C:N ratio was one of the primary factors controlling biosolids N mineralization. The sole N source for the compost is grass clippings that are only available in large quantities [3]. Sullivan *et al.* [4] reported that when food residuals mixed with wood chips and sawdust under field

conditions, N mineralization was 7.3 percent of total N. Several studies report that  $\text{NO}_3\text{G}$  is a final product of composts and  $\text{NO}_3\text{G}$  is sometimes used as an indicator of stability [5]. The amount of  $\text{NH}_4^+$  as a percentage of total N is usually less than 10 percent.

Phytotoxic chemical compounds in composted waste materials can injure plants. Toxic substances obtained from water extracts of composted waste materials inhibited germination and growth of seedlings of test plant and induced darkening and necrosis of root cells. Organic acids such as acetic, propionic and butyric acids can accumulate in compost with a high C:N ratio and high concentrations of ammonia can accumulate in compost with a low C:N ratio [1, 6]. Crop injury due to compost toxicity has been linked to immature compost use and also composting methodology [7, 8]. The most phytotoxic organic acid is acetic, which can completely inhibit *Lepidium sativum* seed growth at concentrations above 300 mg kg<sup>-1</sup> [9] and *Cucumis sativus* seed germination at concentrations above 30 mg kg<sup>-1</sup> [10]. Keeling *et al.* [11] suggested that phytotoxicity rather than C:N ratio was primarily responsible for poor seed germination and growth inhibition. Additionally, application of immature compost can reduce soil  $\text{O}_2$ ; increasing soil temperature to levels that are incompatible with normal root function and causing N immobilization by the soil microbial population because of a high C:N ratio [6]. The extracts of immature and mature (1-year-old) composts showed that the 8-week-old compost extract was the most phytotoxic as it decreased percentage germination, root growth and germination index [12, 13]. They concluded that plant seed germination was inhibited largely by 8-week-old compost extract possibly due to a higher acetic acid concentration.

Many reports on the impact of composting feedstock and process indicated that woody debris and some yard trimmings composts with C:N ratio greater than 25-30:1 and  $\text{NO}_3\text{-N} < 100$  mg kg<sup>-1</sup> are usually deficient in nitrogen and need additional time for compost stabilization. Ammonia toxicity from unstable composts with  $\text{NH}_4\text{-N} < 100$  ppm and C:N < 20:1 is also reported [7, 8, 11, 14, 15, 16, 17]. Gagnon *et al.* [18] concluded that pulp fiber, either applied raw or composted, greatly increased soil organic matter content, the C:N ratio, C mineralization and microbial biomass C, but had no effect on its total N of soil under continuous potato for three years.

Epstein [19] and Walke [20] concluded that the microbial population of bacteria, fungi and actinomycetes changes during composting. According to Finstein and Morris [21] bacteria thrive during all the stages of composting. In most cases bacteria are about

100 times more prevalent than fungi [22]. Actual bacteria populations are dependent upon the feedstock, local conditions and amendments used. Burford [23] observed that at the start of the composting process a large number of species are present including *Streptococcus sp.*, *Vibrio sp.*, *Bacillus sp.* Corominas *et al.* [24], identified species belonging to the genera *Bacillus*, *Pseudomonas*, *Arthrobacter* and *Alicigenes*, all in the mesophilic stage. Two general growth forms in fungi exist molds and yeasts. The most commonly observed species of cellulolytic fungi in composting materials are *Aspergillus*, *Penicillium*, *Fusarium*, *Trichoderma* [25]. While cellulose and hemicellulose are slower to degrade than either sugars or starch, lignin is the most resistant organic waste and as such is usually the last in the food chain to be degraded [19]. *Aspergillus fumigatus* is a mold and has a special significance as a cellulose and hemicellulose degrader [26]. Usually actinomycetes are not present in appreciable numbers until the composting process is well established. Species of the actinomycetes genera *Micromonospora*, *Streptomyces* and *Actinomyces* can regularly be found in composting material. They play an important role in the degradation of the cellulosic component. To some extent they can also decompose the lignin component of wood [18, 27]. The objective of this study was to compare decomposition of different organic wastes and their effects on soil chemical, microbial population and succession and plant growth.

## MATERIALS AND METHODS

Two laboratory and one greenhouse experiments was conducted to study the decomposition of different organic debris and their effects on microbial count and succession, soil chemical properties and plant growth. In both the experiments, six treatments, 1) turf (T), 2) mixed leaves (ML), 3) mixed leaves+stem and wood chips (ML+CW), 4) stem and wood chips (CW), 5) mixed leaf and stem of trimmed *Evonymus Latifolia* (ELS) and 6) control(soil) were used. The organic debris used in this experiments, belonged to common trees of city and recreational parks, such as, *Plantanus orientalis*, *Morus alba*, *Acer pseudoplatanus*, *Robinia pseudacacia*, *Populus spp.*, *Salix sp.*, *Alianthus alitissima*, *Catalpa speciosa*, *Fraxinus rotundifolius* which were crushed separately and mixed on an equal w/w bases.

**Laboratory study:** Six treatments mentioned above used for laboratory experiments to study microbial count and succession and their impact on organic litter

decomposition with respect to different physicochemical nature of the treatments used. Equal amount of soil (100g) added to each plastic pot (approx. 500 cm<sup>3</sup>) and 20g of crushed organic litter from each treatment mixed with soil of each pot. An additional 60g of soil added to pots as a covering layer. Eighteen pots (6 treatments×3replications) were prepared, irrigation with distilled water and incubated at 25±1°C for six months. During incubation period, pots irrigated with distilled water to maintain soil moisture at an optimum level. Sampling was done in 60, 90 days and at the end of incubation, from each pot and transferred into nutrient broth (saline solution) for further microbial studies. Bacterial count was determined on plate count agar by standard spread plate method. Fungi count and identification was done by using saboroud dextrose agar along with Chloramphenicol and Eosin Methylene Blue agar (EMB) medium culture for coli forms. Actinomycetes was counted and identified after usually 4-5 days incubation with Enrichment plate count culture medium and in many cases with EMB culture medium [28-30]. Total microbial count, number of bacteria, fungi and actinomycetes data, were analyzed as a complete randomized blocks by using M STATC [31] and treatment means were compared using Duncan's Multiple Range Test (DMRT) at p=0.05.

**Growth chamber study 1:** The pots used for laboratory study, after sampling for microbial tests was examined for plant growth. After thorough mixing of soil and organic litter in each pot, 20 seeds of snap dragon was sown, irrigated with distilled water and then transferred into growth chamber for germination test. However, no germination, or poor seedling growth observed in all treatments and therefore no results obtained, probably due to the release of toxic substances such as phenolic or aliphatic compounds inhibiting germination and death of young seedlings.

**Laboratory study for germination and seedling growth:** Germination and seedling growth of wheat (tolerant) was performed under laboratory conditions to investigate the inhibitory effects of organic litters used in previous study. For extract preparation from treatments mentioned earlier (except turf), the organic litters of each treatment powdered using a mixer cum grinder. An aqueous extract was prepared by soaking 50 g dried ground sample from each treatment in 250 ml distilled water at 25°C for 36h and the extracts were filtered through muslin cloth, Whatman filter paper No. 1 and then centrifuged at 3000 rpm for 20 min (original extract). Extracts of 1, 2 and

4% concentrations were prepared by diluting original extract of each treatment with distilled water. 30 seeds of wheat placed in each sterilized Petri-dish lined with two Whatman No. 1 filter papers and germination was determined. 5.0 ml of each treatment extract or distilled water (control) added to each Petri-dish on the first day. Thereafter, extracts or distilled water added as needed. Petri-dishes incubated at 20±1°C. Germination count recorded daily for 10 days and final germination, seedling radicle and plumule lengths recorded on day 10<sup>th</sup>. Data collected, was analyzed in completely randomized design with three replications using MSTAT-C [31] and treatment means were compared using Duncan's Multiple Range Test (DMRT) at p=0.05.

**Greenhouse study:** This research was conducted to study the decomposition of different organic litters arranged in six treatment combinations used earlier under greenhouse conditions. Pots with 850 cm<sup>3</sup> volume were filled with 480 g soil (Silty Clay Loam in texture)+100 g of crushed organic litters belonging to each treatment. Two identical sets, each with 24 pots were prepared (6 treatments×4 replications). Pots irrigated daily for 5 days just to maintain optimum moisture levels. Thereafter, 320 g soil added to all pots as a covering layer about 2 cm thick. pots irrigated during the experiment to keep moisture content at 60% field capacity, with day and night temperatures of 25 and 15°C respectively. After 2 months, a set of 18 pots (6 treatments×3 replications) was selected to determine carbon, nitrogen and C:N ratios of the samples. Soil and organic litters of each pot mixed thoroughly, undecomposed litters removed and the grounded mixture passed through a 2 mm sieve. Carbon determined by Walkley and Black method and total nitrogen by kjeldahl method [32]. C: N ratios of all treatments were high (except turf treatment). All pots kept in green house for more 8 months. During this period, optimum moisture maintained by irrigating the pots. At the end of the experiment, soil of each pot was dried, crushed and after sieving with 2 mm mesh, analyzed for organic carbon, nitrogen, total nitrogen, N-NO<sub>3</sub>G, N-NH<sub>4</sub><sup>+</sup> and C: N was calculated for each treatment [32]. The results obtained after 2 months and 10 months were analyzed statistically as a complete randomized design using MSTAT-C [31] and treatment means were compared using Duncan's Multiple Range Test (DMRT) at p=0.05.

**Growth chamber study II:** The pots used for carbon and nitrogen determinations two months after the start of green house study were examined for plant growth. For

Table 1: Chemical analysis of organic litters used in each treatment before the start of experiment

Treatments	C (%)	Total N (%)	C:N ratio	OM:TN ratio	Crude fiber (%)	ADL (%)	ADF (%)	Cellulose
T	47.3	2.19	22	37	24.40	8.9	40.40	31.5
ML	51.1	0.98	52	90	17.95	13.4	31.86	18.5
ML+CW	53.0	0.81	65	113	32.00	20.7	47.60	26.9
CW	57.0	0.66	86	149	36.80	24.0	54.00	30.0
ELS	54.2	1.17	46	80	23.80	18.0	31.90	13.9

T = turf, ML = mixed leaves, ML+CW = mixed leaves+stem and wood chips, CW= stem and wood chips, ELS =mixed leaf and stem of trimmed *Evonymus latifolia*

preparation of pots, same procedures followed as mentioned earlier in growth chamber study I. 20 seeds of snap dragon was sown in each pot, irrigated with distilled water and transferred to growth chamber for germination test at 25°C. Here again, no germination, or poor seed germination were observed in almost all treatments and no results obtained for further analysis.

**Chemical analysis:** Organic litters used in treatments as stated earlier were analyzed for crude protein by determining N in micro Kjeldahl and then multiplied by 6.25. Ash was determined at 550°C for 6h. Acid Detergent Fiber (ADF) and Acid Detergent Lignin (ADL) were determined by the method of Van Soest and Robertson [33] and cellulose by subtraction (Table 1).

To analyze the relationship between microorganisms' total count and lignin: total nitrogen ratios at initial and after 10 months of composting, regressions were fitted to the data for actinomycetes: bacteria and fungi: bacteria vs. lignin: total nitrogen ratios and the results were assessed based on obtained  $r^2$  by using Sigma Stat for Windows V.3.

## RESULTS AND DISCUSSION

**Treatment effects on pH of growing medium, total microbial count, bacteria, fungi and actinomycetes numbers:** Different treatment effects on pH of growing medium (Table 2) indicated significant difference between control with ML+CW and ELS treatments.

Effect of different treatments on total microbial count per gram of soil (Table 2) showed no differences among ML, ML+CW and CW treatments, but significant changes observed between these with other treatments. Maximum total microbial count noted in T and ELS treatments.

Total number of bacteria in T treatment was significantly higher compared to others. No significant changes noted between ML, ML+CW with ELS and ML with CW treatments (Fig. 1). Different treatments with respect to total number of bacteria, Classified as T>ELS>ML+CW>ML>CW.

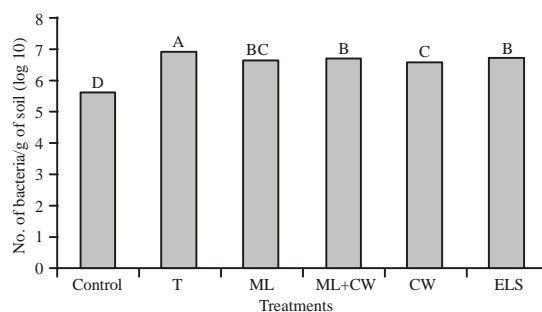


Fig. 1: Total number of bacteria per gram of soil in different treatments (p = 0.05)

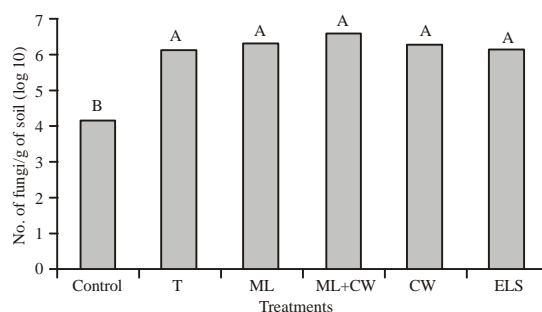


Fig. 2: Total number of fungi per gram of soil in different treatments (p = 0.05)

Results obtained with respect to percent increase in number of bacteria of each treatment when compared with control (Table 2), showed highest significant increase in T compared to ELS, ML+CW, ML (insignificant to each other) and CW treatments. Marked differences also noted between CW and ELS treatments. Maximum percent increase in number of fungi compared with control noted in ELS, ML, CW treatments (Table 2). The differences between ELS with T and ML+CW treatments were significant (Fig. 2).

Maximum number of actinomycetes per gram of soil noted in ELS followed by ML+CW, T, ML and CW treatments, respectively (Fig. 3). Percent - ratio of bacteria: fungi numbers, with respect to, treatments with different organic litters noted to be 67% (T), 57% (ELS), 48% (ML), 46% (ML+CW), 45.7% (CW) respectively.

Table 2: pH of growing medium, total microbial count, percent increase in fungi and bacteria numbers compared with control

Treatments	pH	Total microbial count Per g. soil (log 10)	Increase in fungi/control (%)	Increase in bacteria/control (%)
T	7.97 c	7.00 a	12302.7 bc	2102.0 a
ML	8.43 ab	6.81 c	18197.0 ab	1013.0 bc
ML+CW	8.48 a	6.79 c	8128.3 c	1249.0 bc
CW	8.44 ab	6.74 c	14454.4 ab	895.5 c
ELS	8.50 a	6.90 b	21379.6 a	1320.0 b
Control	8.20 bc	5.61 d	-	-

T = turf, ML= mixed leaves, ML+CW = mixed leaves+stem and wood chips, CW= stem and wood chips, ELS =mixed leaf and stem of trimmed *Evonymus latifolia*

Values in the same column followed by the same letters are not significantly different according to DMRT (at  $p = 0.05$ )

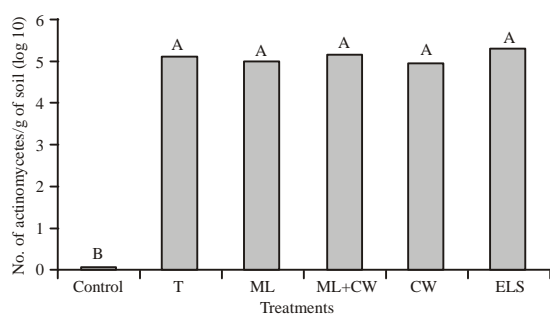


Fig. 3: Total number of actinomycetes per gram of soil in different treatments ( $p = 0.05$ )

The trend in number of bacteria observed earlier when compared with the ratio of bacteria: fungi were more or less similar. Bacteria, fungi and actinomycetes total count in different treatments when compared with each other, indicated maximum number of bacteria in T followed by ELS and minimum in CW treatments, respectively.

In case of fungi population, it followed the opposite trend as  $CW > ELS > T$ .

Maximum number of actinomycetes found in ELS followed by T and minimum in CW treatments respectively. Indicating, thereby, the microbial succession as fungi/ bacteria/actinomycetes.

Comparing the different treatments, it is concluded that the rate of decomposition of plant materials depends on their nitrogen content and C:N ratios. Other factors such as plant parts and maturity are important in addition to nitrogen content. Tissue of young plants compared to mature one are metabolized faster, because the immature plants have a higher nitrogen and maturation is accompanied by lignifications and related alterations. Organic litters rich in lignin are less readily utilized by microorganisms and the quantity of lignin in residues may be of greater importance in predicting decomposition velocity than the C:N ratio. The residues with high

nitrogen, proteins and water-soluble substances are readily decomposed. While increase in the contents of cellulose, hemicelluloses, increase the persistence but is not too great. The lignin is highly resistant.

Simpson *et al.* [34] identified two genera of *Alternaria* and *Cladosporium* in decomposing organic litters under natural soil ecosystems. In this research *Alternaria alternaria* was found in ML and *Alternaria solani* in ELS and CW treatments. Wicklow *et al.* [35] proposed the succession of four groups of fungi, started with *Cladosporium cladosporium* followed by *Alternaria alternaria* and then replaced by systemic fungi such as *Fusarium spp.* finally *Aspergillus*, *Penicillium* and *Trichoderma spp.*

In this research, the fungal succession started from second stage with *Alternaria solani*, which followed by *Fusarium graminearum*, *Aspergillus flavus* and *A. parasiticus* and *penicillium spp.* in ELS and ML treatments. While in CW treatment, the trend was *Alternaria solani*, *Fusarium graminearum* and *Aspergillus flavus* respectively. Turf treatment had *Fusarium spp.*, *Aspergillus niger*, *A. Flavus* and *Penicillium spp* respectively. *Geotrichum candidum* identified in CW and ML+CW treatments, which is responsible for degradation of more complex compounds such as lignin along with cellulose and hemicellulose. Brodie [36] and Carr *et al.* [37] proposed *Geotrichum* and *Aspergillus spp.* as the more effective fungi for organic litter decomposition, which found in plenty in both CW and ML+CW treatments in this study.

Mature plant materials or organic litters with more complex compounds need more active micro flora in order to decompose the stable residues [38]. In this study, the most dominant fungi was identified in CW, ML+CW and ML treatments, while bacteria was more dominant in T and ELS treatments respectively. The presence of fungi such as *Aspergillus*, *Fusarium* and *Trichoderma* in organic

litters can be attributed to high contents of cellulose and hemicelluloses in most of organic litters used in this study. *Aspergillus*, *penicillium* and *Fusarium* are responsible for lignin decomposition. The numbers of aerobic, mesophilic bacteria metabolizing cellulose are those belonging to genera *Bacillus*, *Closteridium*, *Pseudomonas*, *Cytophaga*, which were abundant in most of treatments in this study, for example *Bacillus letrosporus*, *B. cereus*, *Closteridium putrifaciens* in T treatments. *Bacillus spp.*, *Closteridium putrifaciens* and *Closteridium nigrificans* in ML treatment. In ELS treatment, the species of *Bacillus* and *Closteridium* were abundant. *Bacillus cereus* and *closteridium nigrificans* and *Pseudomonas cerpens* and *Pseudomonas flourscense* in CW treatment and in ML+CW treatment, the most predominant ones belonged to *Bacillus subtilis* and *Closteridium putrifaciens*. Bacteria utilizing hemicellulose as the sole sources of C and energy are of the genera *Bacillus*, *Pseudomonas* that were identified in almost all treatments in this research.

Aerobic bacteria capable of decomposing lignin, are strains of *Arthrobacter*, *Flavobacterium*, *Micrococcus*, *Pseudomonas* which were mostly present in ELS, ML, CW treatments in this study [38]. Actinomycetes are commonly present in later stages of decomposition. In this study, their presence in ELS and T treatments found maximum respectively. In T treatments, the genera *Streptomyces* and *Nocardia* and to some extent *Lactobacils* were abundant. While in ELS, the genera *Streptomyces* and *Nocardia* were more active. Bacteria present in CW treatment, covered about 55% of total microbial count, which ranked last among treatments under study in this regard. Bacteria present in CW treatments belonging to the genera *Bacillus* and *Closteridium* eliminated the coli form populations especially pathogenic ones. The microbial succession observed in this study as fungi/bacteria/actinomycetes were mostly related to the nature of organic litters, their chemical characteristics and more complex compounds such as lignin. Actinomycetes were generally the last term of the succession. They stabilize the organic remains also by producing antibiotics [39-41].

#### Treatment effects on soil chemical properties:

Treatments carbon to nitrogen ratio, 2 months after the start of experiment revealed that all treatments showed higher significant C:N ratios compared to control ( $P \neq 0.05$ ). Differences in C: N ratio of treatment T, ML, ELS compared to each other and with two treatments of

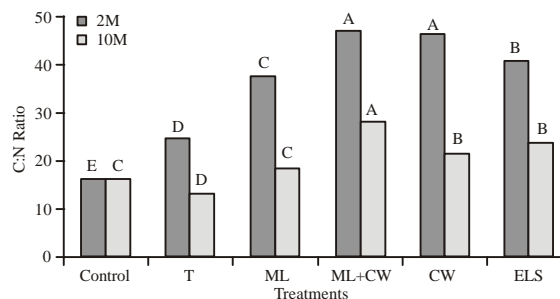


Fig. 4: Carbon to nitrogen ratio after 2 and 10 months of experiment in different treatments ( $p = 0.05$ )

ML+ CW and CW found significant ( $P \neq 0.05$ ). At the end of experiment ELS and CW treatments had more or less same C:N ratios, but there was marked differences between these two with other treatments (Fig. 4).

Differences in C:N ratios in each treatment revealed that, the maximum percent reduction in C: N ratio were observed in ML (50%) and T (45%) respectively and CW (33%) treatment had significantly lowest percent reduction in C: N ratio compared to ELS (42%) and ML + CW (40.5%).

In order to identify the relative toxicity of different organic litters present in treatments under study, total nitrogen was determined after 2 and 10 months in all treatments and organic matter: total nitrogen ratios were calculated and analyzed statistically. Results indicated that after 2 months the OM:TN ratios of all treatments were very high. Comparing treatments with each other, indicated that OM: TN ratios of CW(177) and ML+ CW (150) were significantly higher than ELS (119), ML(109), T(57) treatments. No difference was observed between ML and ELS, but both had significantly different OM: TN ratio compared to other treatments.

OM:TN ratios obtained at the end of experiment, show marked differences between the treatments. CW (101.5), ML + CW (67.4) and ELS (56) still had high OM:TN ratios respectively. Significant differences noted among treatment and all with T (21) and ML (39) treatments (data not shown). Indicating, thereby, decomposition in CW, ML+CW and ELS treatments are not completed and they are immature, probably due to the presence of more complex and resistant organic litters and or higher lignin content in CW, ML+CW and ELS treatments.

He *et al.* [42] proposed that the percent loss in organic litter during composting process ranged from 39 to 75 depending upon source and type of plant materials, which is similar with finding of this study.

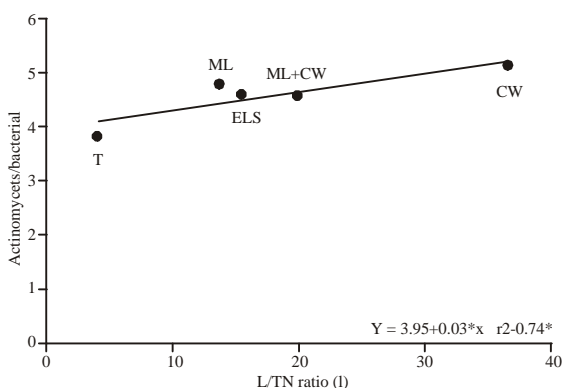


Fig. 5: The relationship between actinomycetes: Bacteria ratios to initial lignin: Total nitrogen ratio of different treatments (P = 0.05)

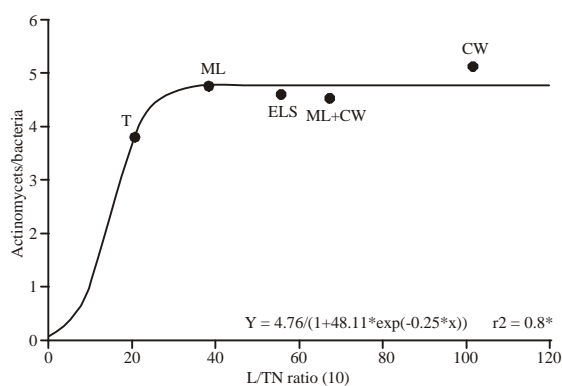


Fig. 7: The relationship between actinomycetes: bacteria ratios to final lignin: Total nitrogen ratio of different treatments (P = 0.05)

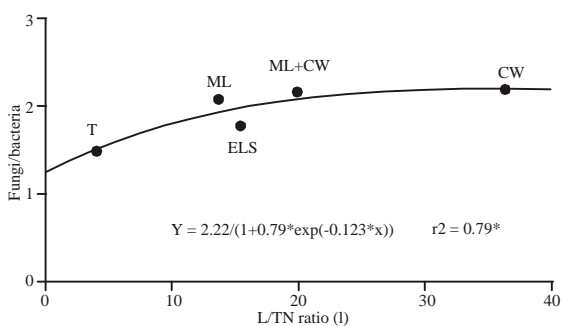


Fig. 6: The relationship between fungi: bacteria ratios to initial lignin: Total nitrogen ratio of different treatments (P = 0.05)

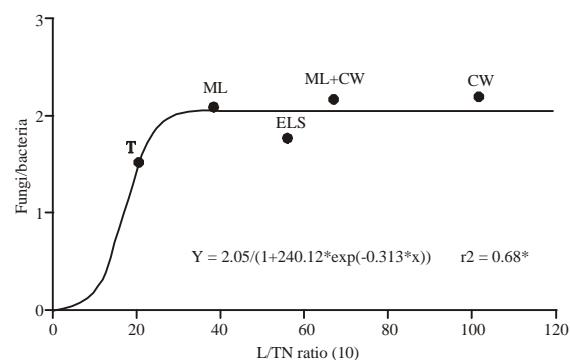


Fig. 8: The relationship between fungi: bacteria ratios to final lignin: Total nitrogen ratio of different treatments (P = 0.05)

Brown *et al.* [43] and Tseng *et al.* [44] indicated that mean percent loss of organic litters when compared the treatments during decomposition was about 30%. In this study, the organic litters' loss (except in ML treatment) was in the range 23 to 37% with mean reduction of 30%, similar to those proposed by Brown *et al.* [43] and Tseng *et al.* [44]. During composting, humic acid concentration increases [45]. They concluded that the aromatic, phenolic and carboxylic structures increased. While polysaccharides and carbohydrates decreased [46]. The results obtained with respect to increase in OM remains and persistence may probably be the result of increased structural stability of compounds as indicated by Proyenzano *et al.* [46]. Maximum OM was observed in CW, ML+ CW and ELS, respectively and was about 57%, 57% and 40% more organic matter contents compared to T and ML treatments (Fig. 7 and 8). Although organic litters with C: N ratios ranging 17 to 87 can be used for

composting, but ideal C: N ratios are in range of 25 to 35 [47]. The results obtained in this research indicating slower decomposition rates in ML+ CW, CW and ELS treatments, probably, due to the presence of higher lignin content or higher lignin: total nitrogen ratios [48, 49]. Golueke [50] concluded C: N ratios higher than 20:1 have deleterious effects on seeds germination and plant growth. The same results observed in this study under green house and growth chamber studies. Total nitrogen of all treatments in this study increased, being highest in T (121%) and lowest in CW (59%) treatments as reported by Day *et al.* [51].

**Treatment concentration effects on germination and seedling growth:** Germination, Plumule and radicle lengths of wheat was reduced with increasing concentration of treatment solutions under study (Table 3). Significant reduction in germination of each solution concentration revealed 26, 41 and 69% reductions at 1, 2 and 4%

Table 3: Solution concentration effects on germination, plumule and radicle lengths and ratio of plumule to radicle lengths

Solution conc (%)	Germination (%)	Plumule length (cm)	Radical length (cm)	Plumule length Radicle length
1	58.2 a	4.15 a	9.0 a	0.49 b
2	49.5 b	3.24 ab	6.9 b	0.59 ab
4	33.1 c	2.26 b	5.1 c	0.64 a

Values in the same column followed by the same letters are not significant (p= 0.05)

Table 4: Effect of different treatments on germination, plumule and radicle lengths and ratio of plumule to radicle lengths

Treatments	Germination (%)	Plumule length (cm)	Radicle length (cm)	Plumule length Radicle length
ML	32.2 c	2.35 b	3.93 d	0.614 a
ML+CW	53.9 b	3.14 b	8.21 b	0.369 c
CW	47.2 b	2.82 b	6.74 c	0.404 bc
ELS	37.8 bc	2.89 b	6.61 c	0.438 bc
Control	78.3 a	5.18 a	11.0 a	0.470 b

T = turf, ML = mixed leaves, ML+CW = mixed leaves+stem and wood chips,

CW= stem and wood chips, ELS =mixed leaf and stem of trimmed Evonymous latifolia. Values in the same column followed by the same letters are not significant (p= 0.05)

solution concentrations, respectively compared with control (P#0.05).

Plumule length although showed a reducing trend with increasing solutions, but the significant reduction observed at 4% compared with 1% solution concentration. Changes in wheat plumule length revealed a significant reductions of 19.4, 44.4 and 73% at 1, 2 and 4% solution concentrations, respectively compared to control (P#0.05).

Changes in wheat radicle length indicated a significant reductions of 15, 40 and 61.5% at 1, 2 and 4% solution concentrations, respectively compared to control (P#0.05). The ratio of plumule to radicle lengths calculated at each solution concentration (Table 3), indicated decreasing trend but the differences between 1 and 4% solution concentrations were significant at P#0.05 only. Comparing relative changes in plumule length and radicle length, more reductions occurred in plumule length than radicle length indicating, thereby, increasing toxicity of organic compounds with increasing solution concentrations and their deleterious effects more in radicle length of wheat.

**Effects of different treatments on germination and seedling growth:** Germination, plumule and radicle lengths of wheat significantly reduced in all treatments compared to control. Wheat germination in ML treatment was significantly lowest compared to ML+CW, CW and control (Table 4). Percent reduction in germination of wheat in each treatment compared with control, showed a significant reductions in the treatments under study.

Plumule length of wheat in all treatments showed marked reductions compared with control (Table 4). However, the differences in plumule length of wheat in treatments when compared with each other was not significant (P#0.05).

Radicle length in CW and ELS was similar but the differences in radicle length of both compared to others were significant at P#0.05 (Table 4). The changes in radicle length of treatments compared to control indicated marked reductions in ML(-64%) and ML+CW (-25%) compared to each other and when both compared with CW(-39%) and ELS (-40%).

The ratio of plumule to radicle lengths of treatments compared to control revealed a significant differences in ML and ML + CW treatments when compared with each other and both compared to CW, ELS and control.

Brinton [52] concluded that the most important organic acids imposing negative impacts on germination and seedling growth are; acetic, butyric, propionic and valeric acids. Zucconi *et al.* [7, 8] concluded that high OM: TN ratio in composts is indication of immaturity in composts. They defined composts maturity with considering OM: TN ratios as; OM: TN<60 for semi mature and OM: TN<50 for mature composts. Increase in solution concentration resulted in drastic reduction in germination of wheat in this study, which is in accordance with findings of Zucconi *et al* [7, 8] and Brinton [52]. Lignin compounds or mixing organic litters with high lignin content, have a vital role in reducing degradation and decomposition process. In this study it was observed that high percentage of lignin in CW, ELS treatments

caused an increase in lignin: TN ratio and had a negative impact on germination and plant growth of snap dragon tested under greenhouse and growth chamber conditions. Mixing CW, with high lignin content with, ML to create ML+CW treatment had an increase of about 75% when compared with ML. It is obvious, that high lignin content retards activity of microorganisms producing polysaccharide enzymes (Fig. 6 and 7). The retardation may be due to the presence of aliphatic compounds as phenolic compounds, etc. [53]. In this study, the presence of 54.5% more lignin in ML+ CW compared to ML caused a drastic reduction in biological degradation in ML+ CW. After 10 months C: N ratio of this treatment increased about 52% than that found in ML treatment resulting in immobilization of NO<sub>3</sub>-N about 77% compared to ML treatment, which is in agreement with findings of Haider and Martin [54].

The regression equations obtained between fungi: bacteria and lignin: total nitrogen ratios at initial and after 10 months indicated that the fungi: bacteria ratio showed an increasing trend with increasing lignin: total nitrogen ratios in T, ML, ELS, ML + CW and CW treatments, respectively (Fig. 6). Whereas, after 10 months, with increasing lignin: TN ratios, the increasing trend was observed up to ML treatments and thereafter it showed a steady state trend which is the indication of slower decomposition rate with increased lignin: TN ratio above 40 (Fig. 8).

The regression equations obtained between actinomycetes: Bacteria and lignin: total nitrogen ratios at initial and after 10 months is presented in Fig. 5 and 7 respectively, indicated an increasing trend with initial L: TN ratios in different treatments, but after 10 months, the increasing trend was observed up to ML and thereafter it proceeds at constant rate as it was observed in fungi: bacteria ratios after 10 months which is again, an indication of slower decomposition rate due to higher lignin: TN ratios above 40 and reducing or inhibiting microbial activity (Fig. 7).

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