

Effect of some edaphic factors on microbial decomposition of leaf fiber biomass of *Toona ciliata* Roxb. and *Trema orientalis* Bl.

Sunanda Chanda, Kakali Ghosh, Priyanka Majumdar and Swapan K. Bhaduri¹

Plant Chemistry Unit, Biological Sciences Division, Indian Statistical Institute,
203, B.T. Road, Calcutta 700 035, India.

¹Quality Evaluation & Improvement Division, National Institute of Research on
Jute and Allied Fibre Technology, Indian Council of Agricultural Research, 12,
Regent Park, Calcutta 700 040, India.

ABSTRACT

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Leaf fiber residue generated as a by-product during the bulk production of leaf protein from forest tree leaves poses disposal problems in the absence of any proper utilisation. The fiber residue can be returned to the forest floor for mineralization as a soil amendment. A short term *in vitro* study was conducted to determine the microbial population and activity as well as the rate of decomposition of the mixture of leaf fiber residues from two perennial plants viz. *Toona ciliata* Roxb. and *Trema orientalis* Bl. in the forest soil in relation to moisture, temperature and pH. The most favourable condition for microbial association and fiber decomposition was found to be 25% moisture content at 35°C and 6.5 pH for microbial association and fiber decomposition. Effects of temperature, moisture and pH on the species composition of fungal flora associated with decomposition were also investigated. *Aspergillus*, *Mucor*, *Rhizopus*, *Trichoderma*, *Penicillium*, *Cladosporium*, etc. were the most common genera isolated from decaying fibrous residues, which ultimately governed the decomposition of leaf fiber in the soil. Some species of *Aspergillus* such as *A. flavus*, *A. fumigatus*, *A. nidulans*, *A. terreus* and some species of *Penicillium*, *Sporotrichum* and *Trichoderma* were found to survive at 50°C.

Keywords: *Toona ciliata*, *Trema orientalis*, leaf fiber biomass, litter, chemical composition, microbial decomposition, edaphic factors, fungal flora

Correspondence: S. Chanda. **Address:** Plant Chemistry unit, Biological Sciences Division, Indian Statistical Institute, 203, B.T. Road, Calcutta 700 035, India

INTRODUCTION

Lignocellulose is one of the most abundant components of biomass on earth and is the chief structural component of the plant cell wall. Biodegradation of various lignocellulosic wastes has been studied by a number of workers (Sein, 1984; Christensen, 1985; Shukla *et al.*, 1990). Decomposition of leaf litter is influenced by a combination of environmental factors including physico-chemical status of the substrate, the external environment coupled with the various enzyme-producing organisms active under particular substrate and environmental conditions (Daubenmire & Prusso, 1963; William & Grey, 1974). Several workers (Dwivedi & Shukla, 1977; Rai & Srivastava, 1982; Rai & Kumar, 1988; Senthilkumar *et al.*, 1993) have studied decomposition of litter in relation to environmental factors.

During bulk production of protein from lopped leaves collected carefully from trees in social forestry circles, a vast quantity of fibrous by-product is thrown to the soil as a soil amendment. Soil contains a variety of heterotrophic microorganisms which influence soil fertility and plant growth by breaking down organic matter and releasing nutrients. The physical structure and chemical composition of the substrate and the climate of the site determine the quality and quantity of microorganisms colonising organic matter. A change in temperature, moisture and pH alters the species composition of active decomposer flora and at the same time has a direct influence on each organism within the community in the terrestrial ecosystem (Alexander, 1977). In the present study the influence of certain edaphic factors like temperature, moisture and pH of the soil environment on the decomposition of leaf fiber residues from two woody perennials viz. *Toona ciliata* Roxb. and *Trema orientalis* Bl. was studied over a period of five months. The study evaluated the relationship between the edaphic factors and the microbial population, activity and the rate of decomposition of the leaf litter.

MATERIALS AND METHODS

Foliage in lush green stage from two trees viz. *Toona ciliata* (Meliaceae) Roxb. and *Trema orientalis* (Ulmaceae) Bl. were carefully collected from the crown following strictly the guidelines of lopping for preparation of leaf protein

meal. Both the trees grow in humid tropical conditions of India. Green foliage of the trees was collected from Hijuli forest in the social forestry circle of West Bengal, India.

Chemical analysis of leaf fiber residues

Freshly prepared leaf fiber residues were collected from the two plant species after extraction of leaf protein. The fiber residues were air-dried and defatted with chloroform - methanol (2:1) in Soxhlet apparatus for 6 h for the estimation of crude fat. The α -cellulose, holo-cellulose, lignin and pentosan contents were estimated from the defatted sample using standard methods (TAPPI, 1971). Ash values were determined by heating samples at 550°C in a muffle furnace and nitrogen was estimated by Kjeldahl method. Modified acid detergent fiber (ADF) was determined by standard method (Anonymous, 1986). The fiber residues were hydrolysed with sulphuric acid (Jeffery *et al.*, 1960) and neutral sugars in the hydrolysate were analysed by gas liquid chromatography as their alditol acetates. A Hewlett-Packard Gas Chromatograph (Model 5830A) equipped with flame ionization detector and stainless steel column (1800 mm x 5 mm diameter) containing 3% ECNSS-M on Supelco-port (80-100 mesh) was used for analysis at 190°C using nitrogen as carrier gas. Molar proportions of sugars were determined conventionally from the peak area. For mineral analysis, the sample was dried at 105°C for 4 h, cooled in a desiccator and then ground in a Wiley-Mill to minimise minor element contamination. The sample was digested in nitric-perchloric acid mixture and analysed for elements with an atomic absorption spectrophotometer (Perkin-Elmer, Model 370).

Soil collection and analysis

Soil samples were collected randomly from ten different sites in the Hijuli forest floor and beneath the soil floor (15-20 cm. deep) and mixed together intimately by grinding and passing through 2 mm mesh sieve. Organic carbon, total nitrogen, and available phosphorus in the soil were determined by standard methods (Jackson, 1973). Exchangeable potassium was determined following the method by Black (1965). Ten g soil was placed in a beaker, stirred with 50 ml of 60% ethanol and filtered. The residue soil was

transferred into a conical flask and shaken with 100 ml of 1N ammonium acetate. The experiment was repeated until 250ml leachate was collected from the soil. The total leachate was analysed for exchangeable potassium cation by flame photometer (EEL, UK) using potassium filter and compared with standard curve for potassium. The pH was measured in a soil-water mixture (1:5, v/v) using a pH meter. Moisture content was determined by oven drying of sample at 105°C for 4h.

Determination of microbial association

Eleven earthen pots (30 cm diameter x 20 cm height) were filled each with 2 kg of fresh forest soil, in which 50g of dried leaf fiber biomass of the two plant species which were thoroughly mixed in equal proportion was dispersed. The pots were incubated at 15°C, 25°C, 35°C 40°C and 50°C to study the effect of temperature whilst maintaining adequate moisture level (28 - 30%) by manual addition of sterile distilled water at regular intervals. The effect of moisture was separately studied by keeping moisture content of the pots at approximately 10%, 25% and 40% level of dry weight at ambient temperature (28° - 33°C) and pH 6.5. The effect of pH was examined by adjusting the pH of the litter at 4.5, 6.5 and 8.5 using dilute acid and alkali. The pots kept as control were treated with the same amount of sterile distilled water. The pots were incubated at room temperature (28°C – 35°C) and regularly supplied with sterile distilled water to maintain 28 – 30% moisture levels.

Rate of litter decomposition

The litter bag technique was used to study the rate of litter decomposition and microbial growth (Shukla *et al.*, 1990). Plastic nets of 1mm mesh were stitched into bags (20cm x 20cm). Fifty bags each containing 10g of air dried mixture of the two leaf fiber residues in 1:1 ratio were placed randomly in 11 different pots having 5 bags in each pot; a few were buried under the soil at a depth of 5 cm. The collection of litter bags was done at monthly intervals for five months. At each sampling time eight replicate bags of litter were collected aseptically of which four were used for isolation of microbes and four were

used for determination of weight loss. Adhering soil particles were removed with care and each litter sample was dried at 70°C to a constant weight.

Isolation of microbes

The litter from four replicate bags was used for the isolation of bacteria, fungi and actinomycetes. The litter was washed with sterile water to remove surface contaminants. One g of litter in 100 ml sterile water was placed in a horizontal shaker to form a homogeneous suspension. The dilution plate method (Warcup, 1950) was used for the isolation of microflora. A minimum dilution of 10^{-4} was used for bacteria, 10^{-3} for fungi and 10^{-4} for actinomycetes. One ml portion of suitably diluent litter suspension was spread in each of the sterile media such as nutrient agar, rose bengal agar and casein agar media supplemented with streptomycin sulphate at 150 mg ml⁻¹ and griseofulvin at 650 mg ml⁻¹ level for screening of total bacteria, fungi and actinomycetes respectively. The bacteria and actinomycetes colonies were counted after 3 and 10 days of incubation at $37 \pm 1^\circ\text{C}$ and $28 \pm 1^\circ\text{C}$, respectively. Fungal colonies were assessed qualitatively by microscopic examination and quantitatively by counting the number of clones appearing in the plate after one week of incubation at $27 \pm 1^\circ\text{C}$. Each fungal colony was subcultured in potato dextrose agar medium and examined by light microscope. Fungi were identified according to Gilman (1965) and Subrahmanian (1971).

Statistical analysis

The two-way analysis of variance technique considering duration and each of the edaphic factors was adopted for statistical analysis of data on the effects of temperature, moisture and pH of soil environment on microbial population in decomposing leaf fiber residue.

RESULTS AND DISCUSSION

Chemical analysis of leaf fiber residues

The chemical composition and mineral make-up of the leaf fiber residues from the two plants viz. *Toona ciliata* Roxb. and *Trema orientalis* Bl. are reported in Tables 1 and 2, respectively. The nitrogen contents of the two samples were similar while ash content of *Toona* (9.5%) was much higher than that of *Trema* (5.6%). This was also indicated by the higher mineral composition of *Toona*. Extractives (crude fat) of both leaf fiber residues were quite high. Analysis of major constituents showed higher lignin (40%) and pectin (6.4%) in *Trema* in comparison to those in *Toona* while holocellulose and α -cellulose contents of both the samples were almost same and comparable to those of straw and grasses (Mc Govern, 1967). Analysis of neutral sugars of the leaf fibers by gas liquid chromatography (Table 3) showed arabinose, xylose and glucose as major sugars with trace amount of rhamnose in both *Toona* and *Trema* while small amount of galactose was found in *Toona* only.

Effect of temperature, moisture and pH on microbial population and weight loss of fiber

The physico-chemical parameters of soil during the study were as follows: pH (water), 5.0; moisture, 25%; organic carbon, 2.5%; total nitrogen, 0.15%; phosphorus, 500 mg/kg; and potassium, 19.31 me./kg. The effect of temperature, moisture and pH on the microbial population viz. bacteria, fungi, and actinomycetes grown on decomposing leaf fibers are shown in Tables 4, 5 and 6, respectively. Fungal population was recorded minimum at 15°C and maximum at 50°C in the fifth sample collected after five months. Bacterial population was low at 15°C but gradually increased with length of exposure. A fair number of bacteria could harbour on litter at 35°C and 50°C. The maximum bacterial count was noted at 35°C in the second and third samples. The population of actinomycetes increased with exposure of samples and recorded highest at 35°C in the fourth and fifth samples and lowest at 50°C. The fungal and actinomycetes populations showed maximum at 25% moisture level while the bacterial count increased with increase in moisture content.

Table 1. Proximate composition of leaf fibre biomass from *Toona ciliata* Roxb. and *Trema orientalis* Bl.

Composition	<i>Toona ciliata</i>	<i>Trema orientalis</i>
Ash	9.54 \pm 2.22	5.63 \pm 0.79
Crude fat	8.97 \pm 1.05	10.16 \pm 1.99
Nitrogen	3.55 \pm 0.45	3.92 \pm 0.37
Lignin	33.95 \pm 4.55	40.05 \pm 5.20
Holocellulose	48.16 \pm 6.66	47.22 \pm 5.16
α - cellulose	27.53 \pm 3.72	27.44 \pm 4.28
Pentosan	12.16 \pm 0.88	15.17 \pm 0.92
Pectin	2.85 \pm 0.07	6.42 \pm 0.08
ADF*	46.00 \pm 7.02	52.11 \pm 8.06

Values are expressed as percent of oven dry weight.

Values are mean \pm S. D. of 5 replicates.

* Acid Detergent Fibre

Table 2. Mineral composition (mg/100g) of leaf fibre biomass of *Toona ciliata* Roxb. and *Trema orientalis* Bl.

Element	<i>Toona ciliata</i>	<i>Trema orientalis</i>
Sodium	103.55 \pm 20.05	62.14 \pm 15.22
Potassium	7.75 \pm 1.05	7.46 \pm 1.31
Calcium	6.63 \pm 0.85	3.62 \pm 0.66
Magnesium	4.55 \pm 0.63	2.92 \pm 0.75
Iron	1.40 \pm 0.07	0.64 \pm 0.06
Zinc	0.02 \pm 0.00	N. D.

Values are mean \pm S. D. of 5 replicates.

N.D. - not determined

Table 3. Neutral sugars (mole percent) of leaf fibre biomass of *Toona ciliata* Roxb. and *Trema orientalis* Bl.

Sugar composition	<i>Toona ciliata</i>	<i>Trema orientalis</i>
Rhamnose	1.22 + 0.16	0.50 + 0.07
Arabinose	14.21 + 2.88	25.09 + 4.66
Xylose	31.99 + 4.96	28.76 + 4.02
Galactose	4.39 + 0.07	0
Glucose	48.19 + 7.33	55.49 + 8.16

Values are mean \pm S. D. of 5 replicates.

Table 4. Effect of temperature on the number of bacteria, fungi and actinomycetes (average number $\times 10^4$ per gram dry weight) on decomposing leaf fibre of *Toona ciliata* Roxb. and *Trema orientalis* Bl.

Temperature (°C)	Incubation period				
	1st month	2nd month	3rd month	4th month	5th month
Bacteria					
15	7.6+1.60	9.5+0.58	11.6+0.97	13.2+1.66	15.8+1.28
25	13.6+2.14	20.1+2.67	25.8+1.91	28.7+2.14	30.7+3.54
35	25.8+1.94	31.2+3.34	32.5+2.38	20.5+1.73	11.8+1.70
40	24.0+0.82	30.8+3.06	31.6+3.13	20.8+1.17	12.0+0.82
50	15.7+2.18	19.9+1.19	22.0+1.83	19.0+1.15	17.1+1.45
Fungi					
15	20.9+2.16	31.9+6.84	50.6+1.98	51.8+9.99	51.0+8.83
25	41.6+6.42	50.6+4.81	65.8+5.61	72.4+2.47	69.0+7.35
35	49.2+7.71	53.5+5.07	79.4+5.63	70.6+5.63	60.5+3.11
40	78.5+1.96	96.4+2.95	125.8+13.73	225.0+46.4	175.0+13.52
50	106.6+11.22	150.8+8.44	265.0+36.55	230.0+12.14	280.0+15.87
Actinomycetes					
15	1.85+0.19	12.60+1.14	14.29+1.25	20.60+3.05	19.55+0.53
25	4.00+0.62	9.50+1.96	15.75+0.96	21.65+1.25	21.00+1.63
35	4.47+0.41	10.95+1.43	16.25+2.06	23.75+2.63	22.66+1.43
40	3.50+0	5.60+0.70	10.66+0.92	17.20+1.04	16.00+1.58
50	1.25+0.20	4.49+0.95	6.76+0.63	3.99+0.62	3.25+0.35

Data are mean values of four replications + S. D. $\times 10^4$

Table 5. Effect of moisture on the number of bacteria, fungi and actinomycetes (average number $\times 10^4$ per gram dry weight) on decomposing leaf fibre of *Toona ciliata* Roxb. and *Trema orientalis* Bl.

Moisture (%)	Incubation Period				
	1st month	2nd month	3rd month	4th month	5th month
Bacteria					
10	0.49 ± 0.05	0.78 ± 0.06	3.60 ± 0.42	2.82 ± 0.44	1.98 ± 0.22
25	14.39 ± 0.71	16.45 ± 0.90	20.05 ± 2.87	14.39 ± 0.55	10.71 ± 0.89
40	22.65 ± 2.02	27.27 ± 1.82	35.56 ± 2.37	22.05 ± 1.89	19.55 ± 1.38
Fungi					
10	13.6 ± 3.61	20.5 ± 1.94	45.6 ± 4.50	40.8 ± 5.55	38.7 ± 4.24
25	27.7 ± 3.84	45.8 ± 4.60	87.0 ± 5.03	70.3 ± 6.80	43.0 ± 9.02
40	26.7 ± 2.52	39.8 ± 3.79	61.4 ± 10.15	52.8 ± 7.48	32.6 ± 5.26
Actinomycetes					
10	2.49 ± 0.05	8.55 ± 0.67	18.39 ± 1.38	17.55 ± 0.82	15.63 ± 0.70
25	3.06 ± 0.11	12.88 ± 1.62	30.53 ± 1.48	20.77 ± 2.18	17.37 ± 2.65
40	0.95 ± 0.14	2.25 ± 0.61	2.90 ± 0.49	1.95 ± 0.38	1.00 ± 1.51

Data are mean values of four replications \pm S. D. $\times 10^4$

Microbial growth was found maximum in the third sample at all moisture levels. Significant increase in fungal population was observed with exposure of samples at pH 4.5 and 6.5. Though pH 8.5 was not favourable for fungal growth it was found ideal for bacterial population. Actinomycetes preferred pH 6.5 to propagate. The count was reduced by acid and alkali treatment.

The data of weight loss of fiber residues and total microbial count at different temperature, moisture and pH conditions are recorded in Figures 1, 2 and 3, respectively. The maximum weight loss of fiber residues was recorded at 35°C (75%), 25% moisture level (74%) and pH 6.5 (72%) while the minimum weight loss was observed at 15°C (40%), 40% moisture level (45%) and pH 8.5 (45%).

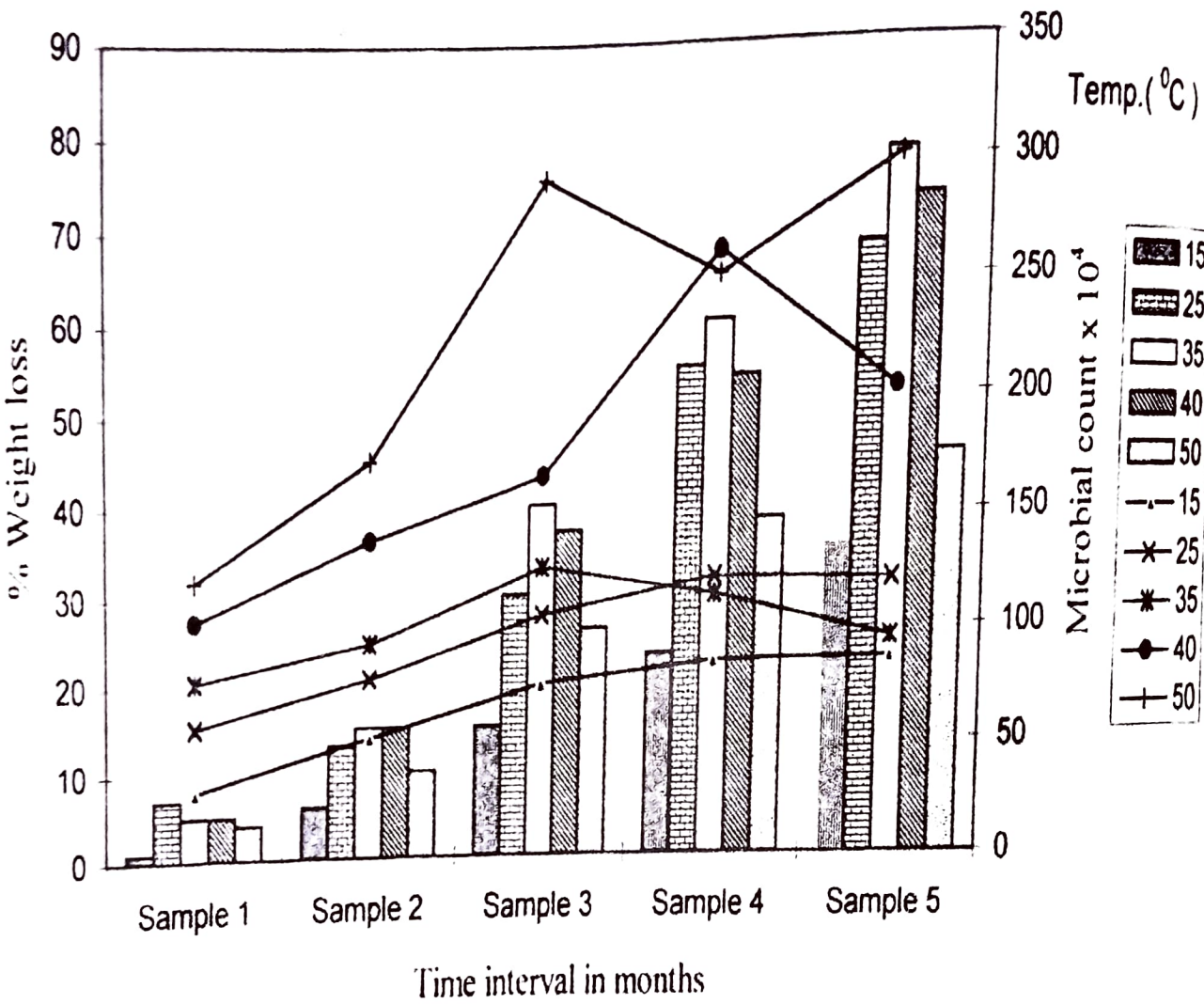


Figure 1. Effect of temperature on weight loss of decomposing leaf fibre and microbial count

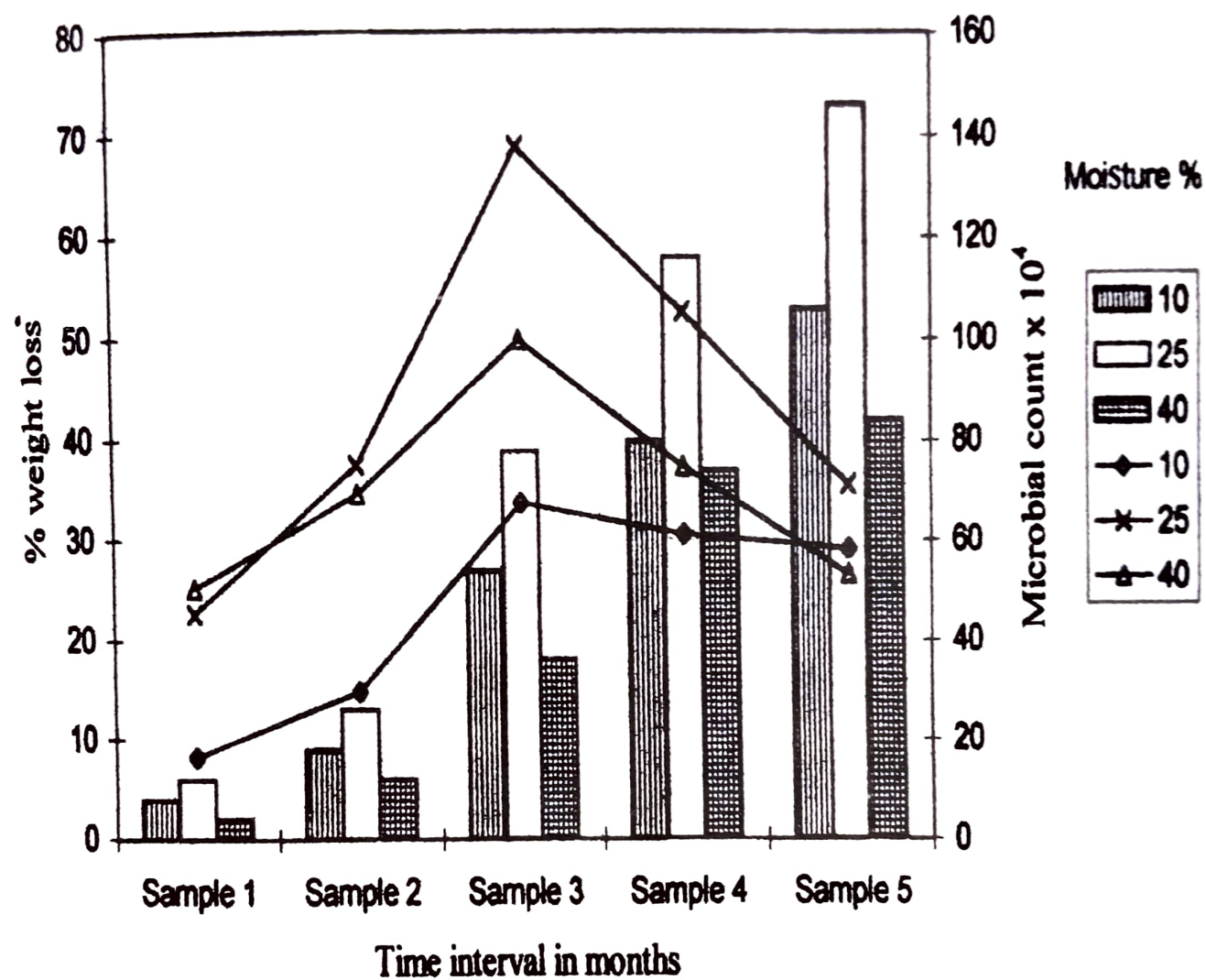


Figure 2. Effect of moisture on weight loss of decomposing leaf fibre and microbial count

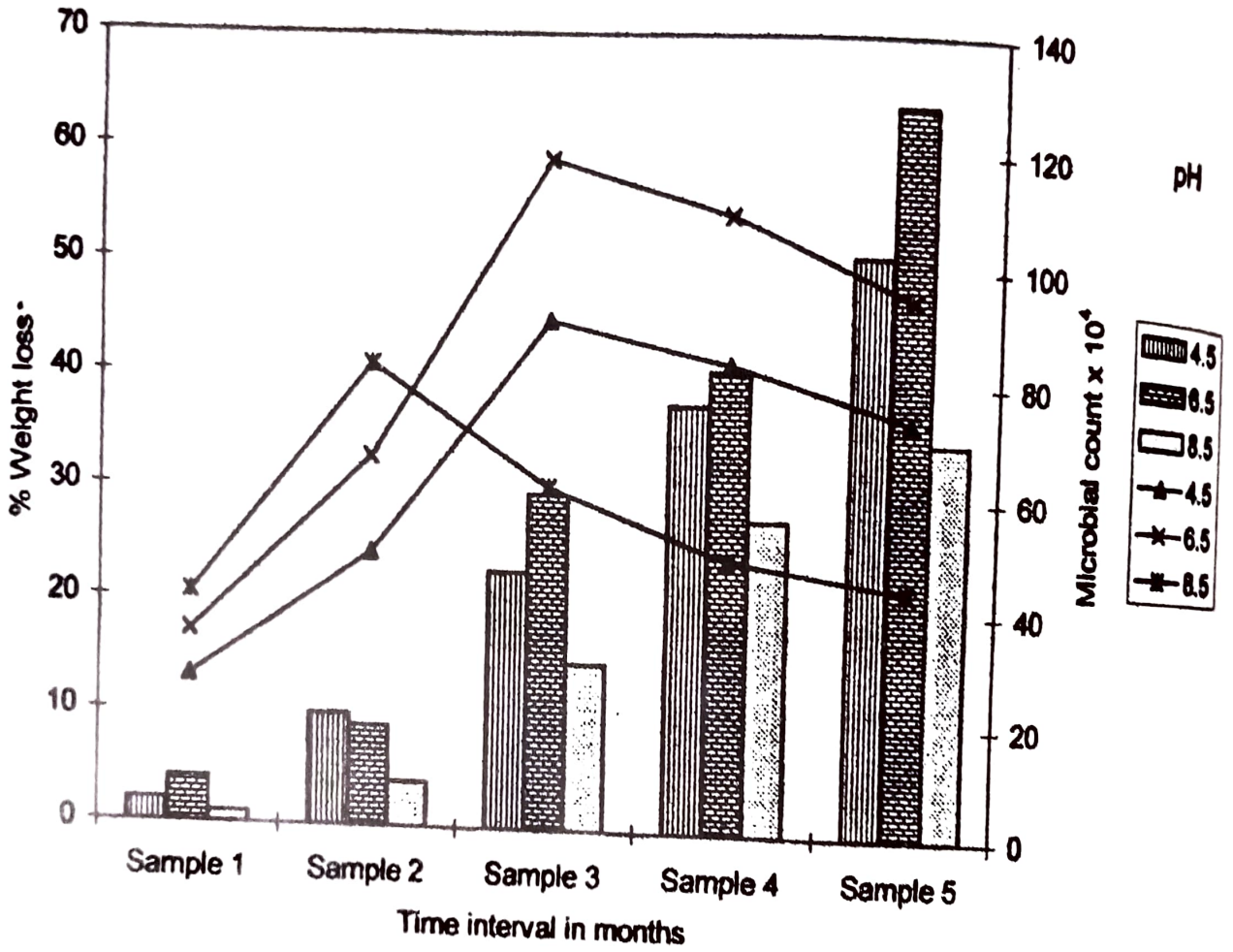


Figure 3. Effect of weight loss on pH of decomposing leaf fibre and microbial count

Table 6. Effect of pH on the number of bacteria, fungi and actinomycetes (average number $\times 10^4$ per gram dry weight) on decomposing leaf fibre of *Toona ciliata* Roxb and *Trema orientalis* Bl.

pH	Incubation Period				
	1st month	2nd month	3rd month	4th month	5th month
Bacteria					
4.5	3.75 ± 0.16	4.40 ± 0.82	5.25 ± 0.50	6.28 ± 0.26	7.65 ± 0.60
6.5	10.90 ± 0.90	13.68 ± 1.04	16.66 ± 2.10	13.95 ± 1.54	11.00 ± 1.41
8.5	21.10 ± 1.35	26.25 ± 1.50	18.96 ± 1.42	10.55 ± 1.90	7.90 ± 0.92
Fungi					
4.5	21.72 ± 1.24	43.66 ± 2.28	83.52 ± 2.05	74.79 ± 1.95	63.20 ± 3.30
6.5	20.17 ± 1.75	42.86 ± 2.06	79.95 ± 4.28	69.73 ± 3.09	61.90 ± 2.39
8.5	19.10 ± 1.67	29.90 ± 3.11	39.95 ± 2.41	35.65 ± 2.67	33.46 ± 2.47
Actinomycetes					
4.5	0.86 ± 0.62	0.92 ± 0.04	1.47 ± 0.11	2.10 ± 0.32	2.55 ± 0.50
6.5	3.35 ± 0.03	9.60 ± 1.34	21.65 ± 2.49	25.32 ± 2.98	21.70 ± 2.34
8.5	0.95 ± 0.06	1.36 ± 0.16	2.50 ± 0	2.67 ± 0.05	2.50 ± 0.20

Data are mean values of four replications \pm S. D. $\times 10^4$

Analysis of variance on the growth of bacteria, fungi and actinomycetes on decomposing leaf litter in relation to different temperature, moisture and pH conditions of soil environment are shown in Tables 7, 8 and 9. Test of significance of differences in microbial populations in the decomposing leaf litter between different edaphic factors and their interaction with duration are shown in the Tables. Results showed that significant differences in microbial populations existed between edaphic factors viz. temperature moisture and pH as well as with duration and their interaction.

Table 7. Analysis of variance of effect of temperature, moisture and pH on growth of bacteria on decomposing leaf fibre of *Toona ciliata* and *Trema orientalis*

Source of variation	DF	SS	MSS	F-ratio	SEM	CD1%	CD5%
Between temperature	4	2407.13	601.78	149.63*	0.2011	0.7534	0.5674
within	4	799.76	199.94	49.71*	0.2011	0.7534	0.5674
Duration							
Interaction (Temperature x Duration)	16	2412.68	150.79	37.49*			
Error	75	301.64	4.02				
Between Moisture	2	5542.52	2771.26	1405.59*	0.4053	1.5436	1.1555
within							
Duration	4	564.22	141.05	71.54*	0.3140	1.1959	0.8952
Interaction (Moisture x Duration)	8	288.85	36.11	18.31*			
Error	45	88.72	1.97				
Between pH	2	1389.63	694.81	460.14*	0.3547	1.3509	1.0113
within							
Duration	4	271.99	68.00	45.03*	0.2748	1.0466	0.7835
Interaction (pH x Duration)	8	783.10	97.89	64.83*			
Error	45	67.74	1.51				

*Significant

Table 8. Analysis of variance of effect of temperature, moisture and pH on growth of bacteria on decomposing leaf fibre of *Toona ciliata* and *Trema orientalis*

Source of variation	DF	SS	MSS	F-ratio	SEM	CD1%	CD5%
Between temperature	4	388997.62	97249.41	486.57*	3.1612	11.8425	8.9188
within	4	81879.35	20469.84	102.41*	3.1612	11.8425	8.9188
Duration							
Interaction (Temperature x Duration)	16	73552.71	4597.04	23.00*			
Error	75	14989.95	199.87				
Between Moisture	2	5026.07	2513.04	78.21*	1.6364	6.2321	4.6654
within							
Duration	4	13360.94	3340.23	103.95*	1.2676	4.8276	3.6140
Interaction (Moisture x Duration)	8	2148.92	268.62	8.36*			
Error	45	1446.00	32.13				
Between pH	2	8024.73	4012.36	613.51*	0.7382	2.8114	2.1046
within							
Duration	4	16788.00	4197.00	641.74*	0.5718	2.1777	1.6302
Interaction (pH x Duration)	8	2906.39	363.30	55.55*			
Error	45	294.34	6.54				

*Significant

Table 9. Analysis of variance of effect of temperature, moisture and pH on growth of actinomycetes on decomposing leaf fibre of *Toona ciliata* and *Trema orientalis*

Source of variation	DF	SS	MSS	F-ratio	SEM	CD1%	CD5%
Between temperature	4	445.79	1783.17	252.95*	0.2969	1.1123	0.8377
within	4	718.39	2873.56	407.62*	0.2969	1.1123	0.8377
Duration							
Interaction							
(Temperature x Duration)	16	43.29	692.65	25.11*			
Error	75	1.76	132.18				
Between Moisture	2	2365.18	1182.59	654.09*	0.3882	1.4784	1.1868
within							
Duration	4	1581.98	395.49	218.75*	0.3007	1.1452	0.8573
Interaction							
(Moisture x Duration)	8	804.03	100.50	55.59*			
Error	45	81.36	1.80				
Between pH	2	2807.00	1403.50	917.08*	0.3571	1.3600	1.0181
within							
Duration	4	617.28	154.32	100.84*	0.2766	1.0534	0.7886
Interaction							
(pH x Duration)	8	801.61	100.20	65.47*			
Error	45	68.87	1.53				

*Significant

Effect of temperature, moisture and pH on qualitative nature and relative abundance of fungal flora

Microscopic observation revealed that fungal hyphae effectively colonised on the outer surface of the fiber. Insignificant number of fungal colonies were isolated from fibrous residues incubated at 15°C and 35°C. The fiber was incubated at room temperature (25°-33°C) instead of 35°C as there was little difference between them. The relative abundance of fungal species are recorded in Table 10. Fungal colonies like *Cunninghammella ehinulata*, *Mucor racemosus*, *M. mucedo*, *Rhizopus nigricans*, and *Penicillium* colonies were observed in the first two months followed by *Cladosporium cladosporoides*, *C. herbarum* and *Fusarium pallidoroseum*. Lastly colonies of *Aspergillus* spp. viz. *A. niger*, *A. flavus*, *A. terreus*, *A. japonicas*, along with *Chaetomium globosum*, *Phoma nebulosa*, *Trichoderma pseudokoningi*, *T. lignorum*, *Coprinus* spp. etc. were quite luxuriantly formed. While growth of *Aspergillus* spp. were retarded at 15°C, *A. fumigatus*, *A. terreus*, *A. nidulans*, *A. flavus*, *A. niger*, *Sporotrichum*, and *Chaetomium olivaceum* were most dominant species at 50°C observed at 10% moisture level. A unique range of fungal diversity viz. *Mucor racemosus*, *M. Mucedo*, *Rhizopus arrhizus*, *R. nigricans*, and *Aspergillus* spp. like *A. niger*, *A. flavus*, *A. terreus*, and *Penicillium* spp. Small number of fungal species were observed at 25% moisture content while a sharp decline in fungal species was observed when the moisture increased to 40%. Fungal species like *Fusarium pallidoroseum*, *Alternaria tenuis*, *Trichoderma viride*, *T. koningi*, *Humicola*, *Chaetomium globosum*, *Paecilomyces* etc. were abundant at pH 4.5. Alteration of pH caused sharp decline in the number of fungal species. The most significant effect was recorded at pH 8.5 and the least at pH 6.5. At higher pH *Aspergillus* species such as *A. niger*, *A. flavus*, *A. terreus*, *A. fumigatus*, as well as *Penicillium frequentus* and *P. chrysogenum* were also observed.

Table 10. Relative abundance (%) of fungal species isolated from decomposing leaf fibre of *Toona ciliata* Roxb. and *Trema orientalis* Bl. at different edaphic conditions

Fungal species	Temperature (0 C)					Moisture (%)			pH		
	10	25	35	40	50	10	25	40	4.5	6.5	8.5
<i>Alternaria alternata</i>		05	02				01			01	
<i>A. tenuis</i>		05	02				01		05	01	15
<i>Aspergillus flavus</i>			03	03	07	05	03	10	03	02	15
<i>A. fumigatus</i>			03	10	10		05		10	04	
<i>A. japonicus</i>	04	07	02	02	02	08	04		07	05	
<i>A. nidulans</i>			02	03	03		02			05	
<i>A. niger</i>	04	08	01	02	01	07		10	05	06	10
<i>A. terreus</i>			02	02	05					06	10
<i>Chaetomium globosum</i>			03	10			05		05	08	
<i>C. olivaceum</i>		10	02	10	10					05	
<i>Cladosporium Cladosporoides</i>	12	04	03			10	02	06	05	02	
<i>C. herbarum</i>	10	03	04			10	01	06		02	
<i>Coprinus sp.</i>			05	08			03			05	
<i>Cunninghamella echinulata</i>	06	03					04	05		05	07
<i>Fusarium pallidroseum</i>	10	10	05			10	06	08	03	04	
<i>Humicola sp.</i>			08	05	10		03		07	04	
<i>Mucor mucedo</i>	15	08	03				03	05	02	03	
<i>M. racemosus</i>	15	07	03			10	04		02	03	
<i>Paecilomyces sp.</i>			07	07					06	05	05
<i>Penicillium chrysogenum</i>	02						05	03	02	10	
<i>P. frequentas</i>		03	05	02	05	10	07	07	03	07	
<i>P. funiculosum</i>			05			05	01	10	03		
<i>P. javanicum</i>	10	04				08					
<i>Phoma nebulosa</i>				10	02		04		05	05	
<i>Rhizopus nigricans</i>			05			02	05	05	02		
<i>R. arrhizus</i>		07	05	01		15	07	05	02	02	
<i>Sporotrichum sp.</i>	12	05	05	07	15		05		04	05	
<i>Trichoderma koningi</i>		05	05	05	08		02	05	10	01	
<i>T. pseudokoningi</i>		03	05	05			03	05	10	01	08
<i>T. viride</i>			05	08	10		05	10	10	03	08

DISCUSSION

The results indicate that the most favorable temperature, moisture and pH for microbial growth in the litter are 35°C, 25% and 6.5 respectively. The optima of the three factors for growth of microorganisms govern the optima of microbial metabolic activity. In recent years a number of research workers (Divevre & Horwath, 2000; Henriksen & Breland, 1999) have focused their attention on the biodegradation of lignocellulosic residues on the soil surface under various environmental parameters to enrich soil fertility status. Edward (1975) noted that microbial activity studied in terms of microbial respiration declined at temperature above 35°C except in the case of thermophilic microbial activity. Frey *et al.* (1999) while working on bacterial and fungal abundance and biomass in the conventional and non-tillage agro-ecosystems found that soil moisture was positively correlated with fungal biomass. Berg *et al.* (1998) worked on the dynamics of bacterial and fungal biomass and their relationship with moisture and temperature. Their results indicated the existence of an environmental stress factor affecting the abundance of microbes in phases of decomposition. The pH influences the type of microorganisms associated with the carbon cycle of any habitat (Alexander, 1977). The rapid decomposition of litter occurred at pH 6.5 which is also optimum for microbial count. According to Paulsamy *et al.* (1995) a pH value of 6.5 is the ambient one for microbial association and biodeterioration of leaf fiber at 32% litter moisture. The lowest rate of decomposition was recorded at pH 8.5 which might be attributed to the adverse effect of alkaline reaction on fungi.

Several species of *Aspergillus* such as *A. flavus*, *A. fumigatus*, *A. terreus* and some species of *Penicillium*, *Sporotrichum*, *Trichoderma*, are the pioneer colonizers of the litter and can survive at a high temperature (50 °C). Survival of some *Aspergillus* species under adverse situations at higher temperatures has been reported earlier (Upadhyay, 1987). Other species like *Alternaria*, *Humicola*, *Cladosporium*, *Chaetomium*, *Rhizopus*, *Mucor* were also abundant in the primary as well as in later stages of decomposition, which ultimately govern the biodeterioration of leaf fibre in the soil. Occurrence of more or less similar species of fungi were reported by several workers (Paulsamy *et al.*, 1995; Shukla & Mishra, 1992; Griffith & Boddy, 1990) where the dominance of *Fusarium*, *Aspergillus*, *Penicillium*, *Mucor*, *Trichoderma*, *Rhizopus*, *Chaetomium*, *Cladosporium* and *Alternaria* was cited.

Mishra & Kanaujia (1973) isolated more diverse microflora from cultivated soil as compared to grassland and forest soils and they found that *Mucor*, *Aspergillus*, *Pencillium*, *Cladosporium* were the most dominant fungi. These species are also dominant in maize (Dkhar, 1983) and rice (Baruah, 1983) fields and isolated relatively high frequencies. The overall study reveals that *Aspergillus* is extremely common in subtropical soils and frequently forms colony in all decomposing fibers as reported by previous workers (Saxena & Sarbhoy, 1963).

It may be assumed that microbial assemblage obtained from decomposition of litter from leaf fiber is a process that replenishes the forest ecosystem's nutrient pools. Litter on the forest floor acts as an input-output system for nutrients (Das & Ramkrishnan, 1985). Because of the important role of litter decomposition in regulating nutrient fluxes, factors influencing litter decomposition have important implications for long-term productivity of forest ecosystems (Adams and Angradi, 1996). Therefore, litter dynamics of the humid tropical forest ecosystems in relation to microbial biomass have received considerable research attention.

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