

MICROBIOLOGICAL AND BIOCHEMICAL CHANGES DURING THE COMPOSTING OF OIL PALM EMPTY-FRUIT-BUNCHES. EFFECT OF NITROGEN SUPPLEMENTATION ON THE SUBSTRATE

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Abstract

The composting of oil palm empty-fruit-bunches and of oil palm empty-fruit-bunches in supplementation with either goat dung, cow dung or chicken manure differed in the resulting C:N ratios. The initial C:N ratios (52:1, 35:1, 48:1, 47:1) for the four compost heaps were significantly reduced to 24:1, 14:1, 18:1 and 12:1, respectively, after 60 days of composting, resulting in the production of a stable humus that is suitable for crop production. A temperature of 70°C was maintained for 3 days at the onset of composting. Both mesophilic and thermophilic bacteria showed consistent activity throughout the process, whereas fungal activity was completely suppressed during the peak heating phase. The rate of utilization of cellulosic material showed a positive correlation with the increase in the nitrogen content of the compost.

Key words: Oil palms, empty-fruit-bunches, manure additions, compost, cellulose, carbon:nitrogen, microorganisms.

INTRODUCTION

The Malaysian palm oil industry generated 1.16 million tonnes of empty-fruit-bunches (EFB) in 1985 alone, the volume predicted to increase to 2.16 million tonnes by 2000 (Ministry of Primary Industries, Malaysia, 1991). Mulching currently accounts for only a fraction of the EFB that are discarded; these are normally burnt in incinerators for the ash as fertilizer. Currently there is much interest in utilizing palm oil waste in general (Thambirajah & Kuthubutheen, 1989). Composting has been suggested as an alternative to incineration of the waste as the process converts the waste, which is essentially organic in nature, into a humus that is suitable for crop production (Gomez & Park, 1983; Gray *et al.*,

1971). In composting, the higher-plant material breaks down under the influence of aerobic thermophilic microorganisms present in the waste to a material rich in organic nutrients.

On one hand, the EFB component of palm-oil waste presents particular difficulties both in its transportation and in its incineration, owing to its extensive bulk. On the other hand, EFB contain a high proportion of cellulosic matter which is easily decomposed by a combination of physical, chemical and biological processes. The bunch consists of 70% moisture and 30% solids; of which holocellulose accounts for 65.5%, lignin 21.2%, ash 3.5%, hot-water-soluble substances 5.6% and alcohol-benzene solubles 4.1% (Husin *et al.*, 1985). In an extension of an earlier study (Thambirajah & Kuthubutheen, 1989), on the composting of palm press fibre, we report in this paper the results of a similar study on EFB.

METHODS

Materials

Oil palm empty-fruit-bunches were shredded into loose fibrous material by using a shredder manufactured by B & W Engineering, Angel Drove, Ely, Camb. (Serial No. R/N 30/12). Goat dung, cow dung and chicken manure (poultry broiler floor litter) were collected from the research farm of the Institute of Advanced Studies.

Compost

Four heaps were prepared as follows: the first (Heap 1) had 90 kg EFB, the second (Heap 2) 90 kg EFB and 25 kg goat dung, the third (Heap 3) 90 kg EFB and 25 kg cow dung and the fourth (Heap 4) 90 kg EFB and 25 kg chicken manure (broiler floor litter). Water was added to each of the heaps to a final moisture content of 65% w/w. The heaps were turned once every week for 8 weeks. However, during the first week the heaps were turned on the third

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day as well, when the temperature in the heaps rose rapidly.

Sampling

Grab samples (approx. 200 g) were collected from the interior of the heaps that were turned weekly. The samples were bulked and portioned into two lots. Microbial enumeration was performed on one lot. The other was dried to constant weight (60°C for 2 days) for the chemical analyses. Microbial counts were calculated on the basis of dry-matter weight.

Temperature

The temperature at the top, middle and bottom layers of the interior of the heaps was read daily at 09.30 by using a thermocouple thermometer (Suntex ST-52K type). Ambient temperature was also recorded daily throughout the process.

Chemical analysis

Carbon contents were determined on dried, finely-ground (1 mm) samples by using the method of Tinsley (Allen *et al.*, 1974). The cellulose content of the fibres was determined by using the method of Updegraff (1969). Total nitrogen was determined by using the Kjeldahl method (AOAC, 1975). Changes in lignin content were followed by using the method of Zadrazil and Brunnert (1980). Changes in pH were monitored by using 10 g of dried, finely-ground samples suspended in 50 ml distilled water.

Microbial counts

The dilution plate method was used to enumerate the microorganisms in the compost heaps. A sample (10 g) from the grab lot, together with sterile distilled water (90 ml), was homogenized in a stomacher (model SF 400) for 2 min. Serial dilutions of this stock were made and used to inoculate plates in triplicate (Cappuccino & Sherman, 1983). Nutrient agar, corn meal agar, yeast extract agar and actinomycete agar were used in the counting of the number of bacteria, mesophilic fungi, thermophilic fungi and actinomycetes, respectively. The plates were incubated at 35 and 45°C for mesophilic and thermophilic bacterial counts and at 30 and 45°C for the mesophilic and thermophilic fungal and actinomycetic counts. Bacterial counts were recorded 48 h after plating, whereas fungal and actinomycetic counts were made 72 h after plating.

Statistical analysis

Statistical analyses of the data were performed by using the subroutine of the Statgraphics package (Statistical Graphics Corporation, 1987), with response variables being C:N ratio, carbon concentration, nitrogen levels, cellulose, lignin and pH. The Multifactor Analysis of Variance Program gave the effect of each treatment on the response variables with the day of collection as a co-variant.

RESULTS AND DISCUSSION

The composting of EFB resulted in a significant reduction in the C:N ratios when manure was included in the substrate. This correlated with the microbial activity which was evident from the increases in the temperature of the heaps. In composting, the C:N ratio indicates the maturity of the product (Gray *et al.*, 1971; Chanyasak & Kubota, 1981; Jimenez & Garcia, 1989). An initial ratio ranging between 30:1 and 50:1 that converges to a final value of 10:1 to 15:1 is often taken as a measure of a stable humus (Taiganides, 1977). The ratio for most agricultural waste is often as high as 70:1, but can be lowered to a more appropriate level by the addition of nitrogenous supplements such as manure.

The initial C:N ratios of Heap 1 (control), Heap 2 (EFB and goat dung), Heap 3 (EFB and cow dung) and Heap 4 (EFB and chicken manure) (52:1, 35:1, 48:1 and 47:1, respectively) were reduced to 24:1, 14:1, 18:1 and 12:1 after 60 days (Fig. 1). These values were interpreted in terms of a mature compost and they approximated the limiting ranges of 15:1 to 20:1 (Poincelot, 1974). Supplementation with chicken manure afforded a compost with the lowest ratio when the proportion of EFB to manure was fixed at 3:1.

The number of mesophilic and thermophilic microorganisms fluctuated with respect to changes in temperature of the heaps; higher temperatures can destroy pathogenic microorganisms and other plant material. The temperature of the control (Heap 1) increased rapidly to about 75°C within 1 day and then dropped gradually to 40°C after 21 days, after which time the heap slowly attained ambient temperature (Fig. 2). This trend was also observed for the heaps supplemented with the dung (Figs 3–5) but the three exhibited a higher overall temperature increase for the same period. Microbial counts (Figs 6–11) were higher for these heaps. A maximum temperature of 75°C appears sufficient to effect the destruction of pathogens (Spaggiari & Spigoni, 1986).

Three main groups of microorganisms — bacteria, fungi and actinomycetes — determine the biodegradation pattern of cellulosic plant material (Fergus, 1964; Chang & Hudson, 1967) and are responsible for the physical and chemical changes during composting. In this study on EFB, the bacterial and actinomycetic counts were low during the peak heating phase, but increased subsequently, remaining without change for the duration of the process. However, fungi appeared to be completely absent during the peak heating phase (Figs 8 and 9), an observation similar to that made on wheat straw compost (Chang & Hudson, 1967).

Depending on the conditions of growth, the substrate and the organisms involved, the end products

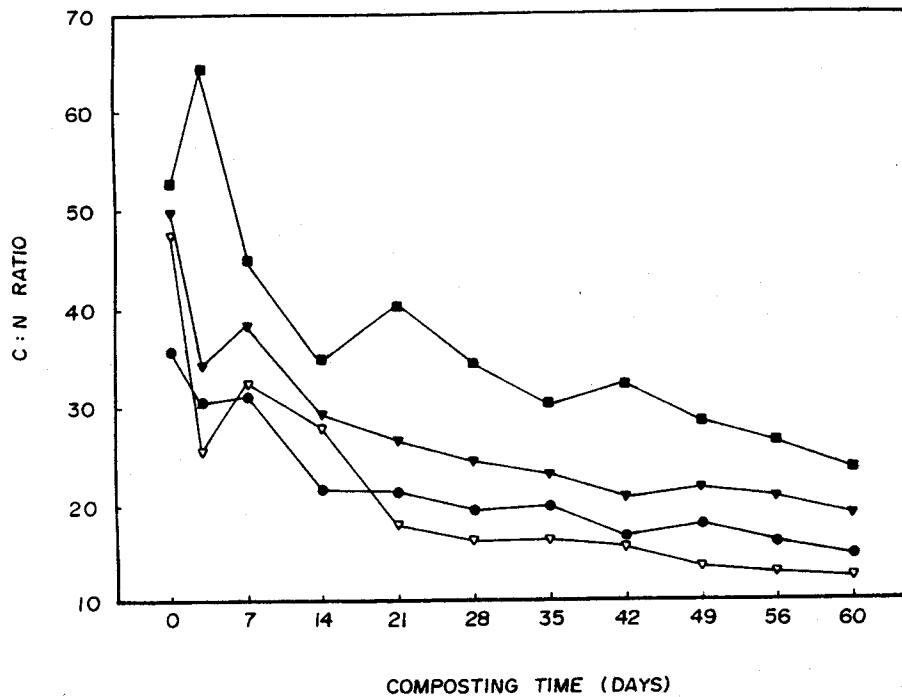


Fig. 1. Carbon versus nitrogen plot of the four heaps. ■, Heap 1; ●, Heap 2; ▼, Heap 3; ▽, Heap 4.

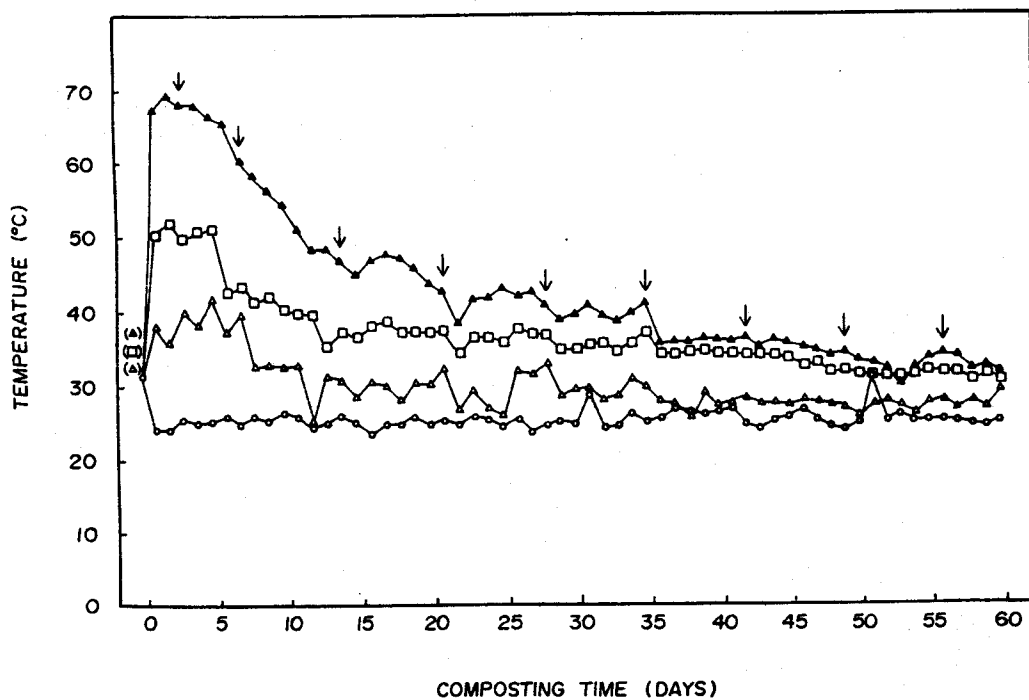


Fig. 2. Temperature-time plot of the top Δ , middle \blacktriangle and bottom \square layers of Heap 1. \circ Indicates ambient temperature. Arrows (\downarrow) indicate turning intervals of the heap.

of fermentation vary greatly. Microbial fermentation of carbohydrates generally results in an increase in acidity (Garg & Neelakantan, 1982). *Clostridium* species commonly ferment glucose to yield butyl and other alcohols and certain acids. *Lactobacillus lactis* yields almost entirely lactic acid, while *Lactobacillus brevis* yields lactic and acetic acids, ethyl alcohol and carbon dioxide (Frobisher *et al.*, 1974).

In composting, carbohydrates are also broken down to humic and fulvic acids (Spaggiari & Spigoni, 1986). However, the fulvic acid is subsequently degraded. This, together with ammonification of inorganic nitrogen, accounts for the neutral pH which is generally attained at the end of the process (Chang & Hudson, 1967; MacGregor *et al.*, 1981). The initial pH levels in the present study averaged

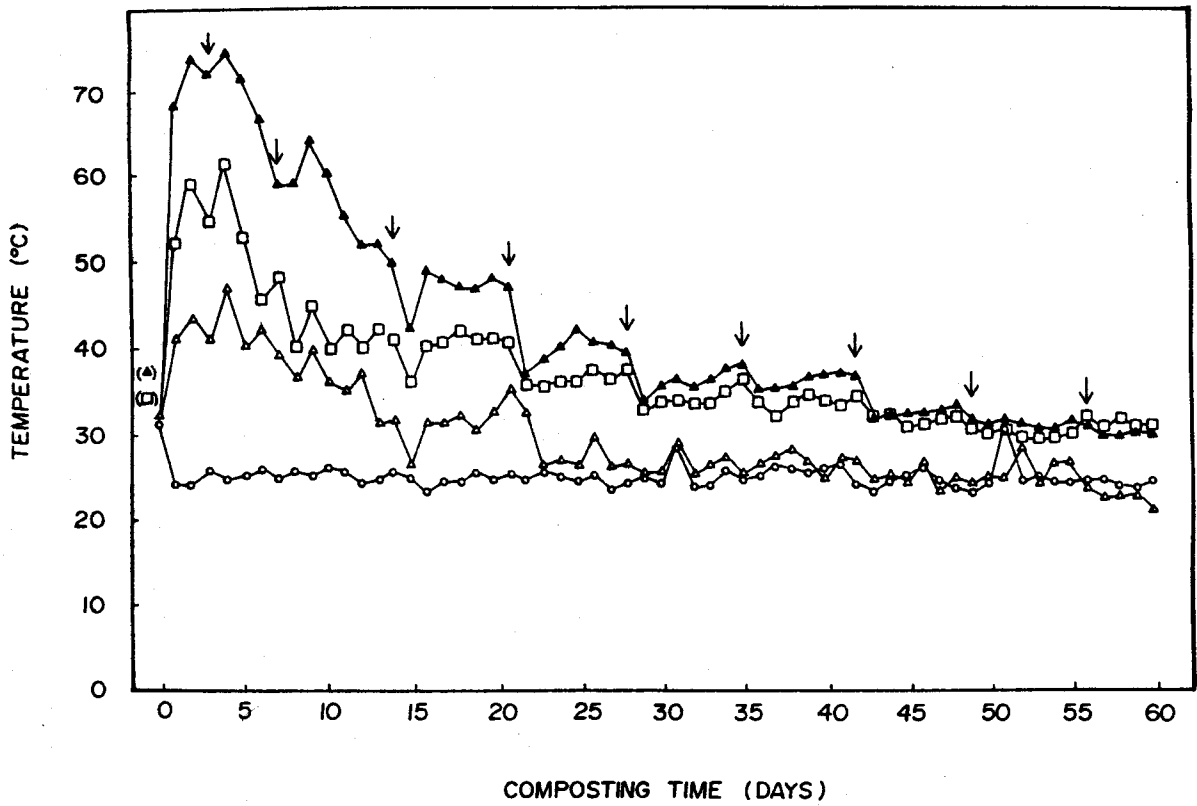


Fig. 3. Temperature-time plot of the top Δ , middle \blacktriangle and bottom \square layers of Heap 2.

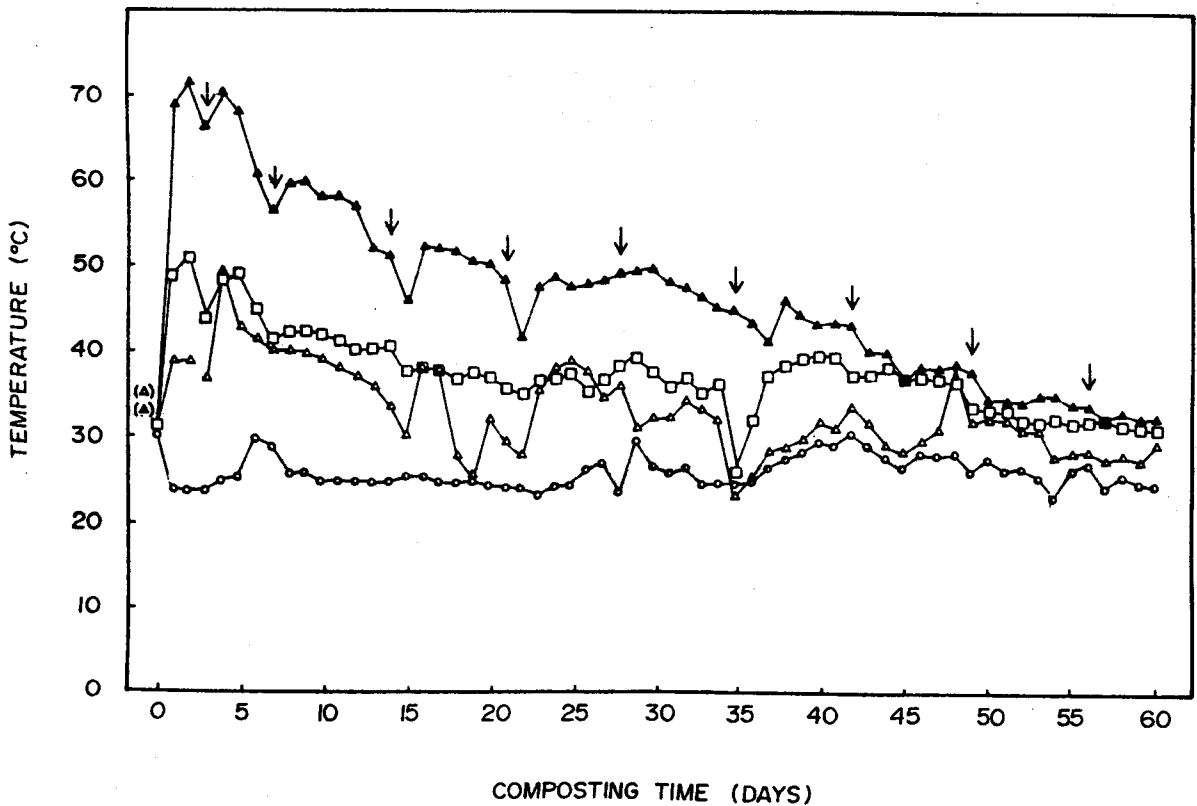


Fig. 4. Temperature-time plot of the top Δ , middle \blacktriangle and bottom \square layers of Heap 3.

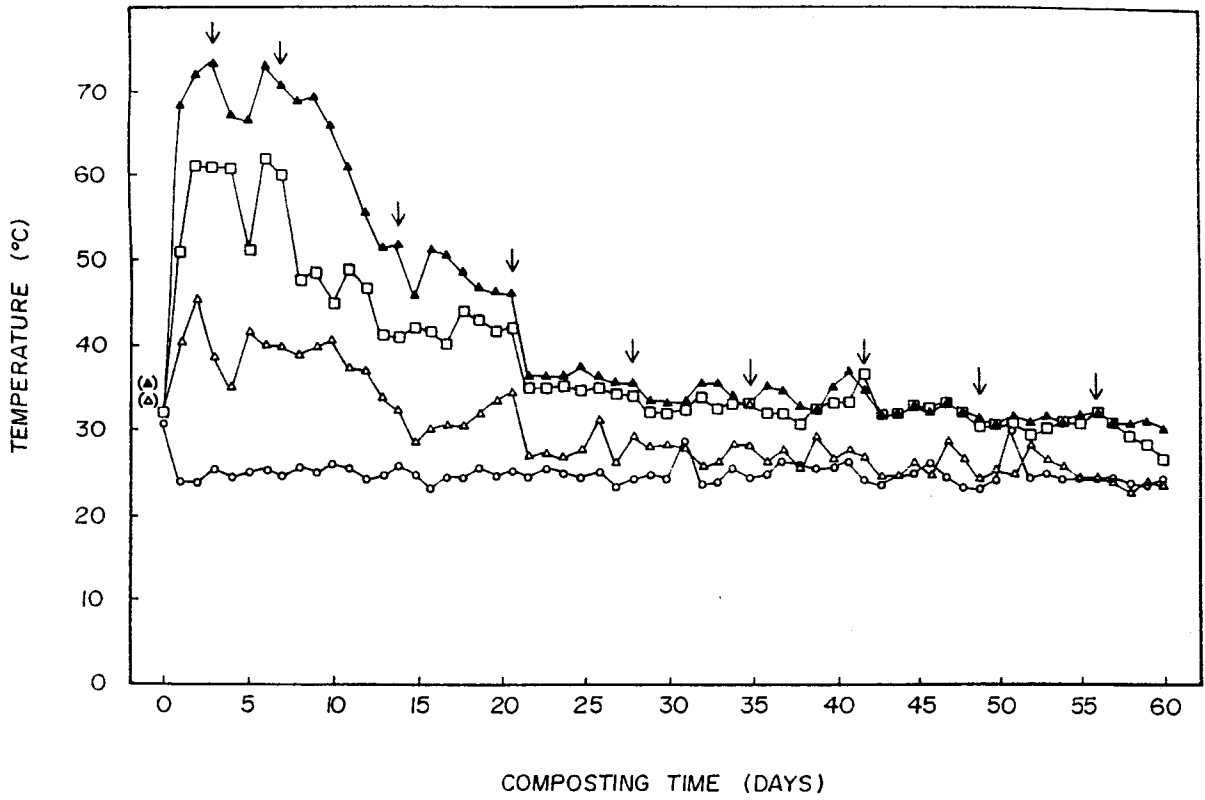


Fig. 5. Temperature-time plot of the top Δ , middle \blacktriangle and bottom \square layers of Heap 4.

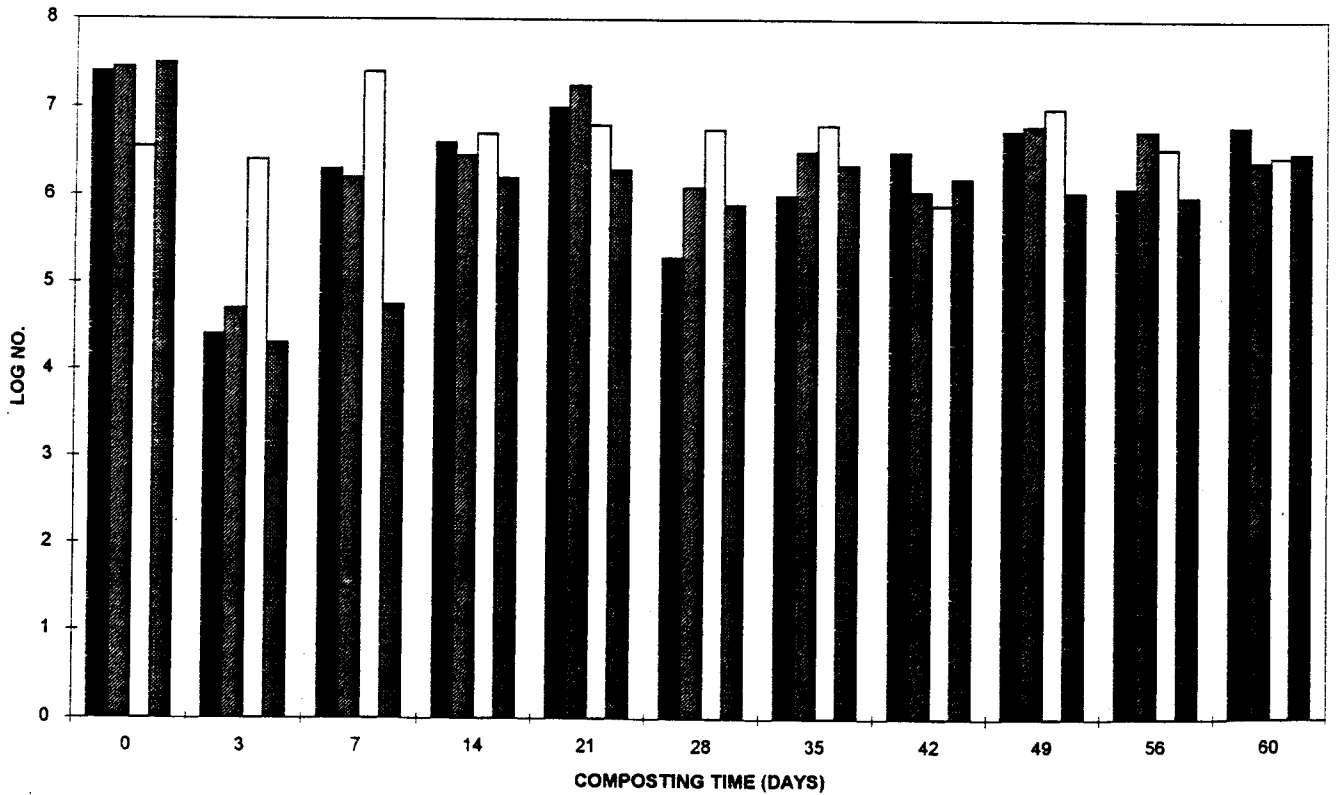


Fig. 6. Log numbers of mesophilic bacteria during the composting process. \blacksquare , Heap 1; \square , Heap 2; \square , Heap 3; \square , Heap 4.

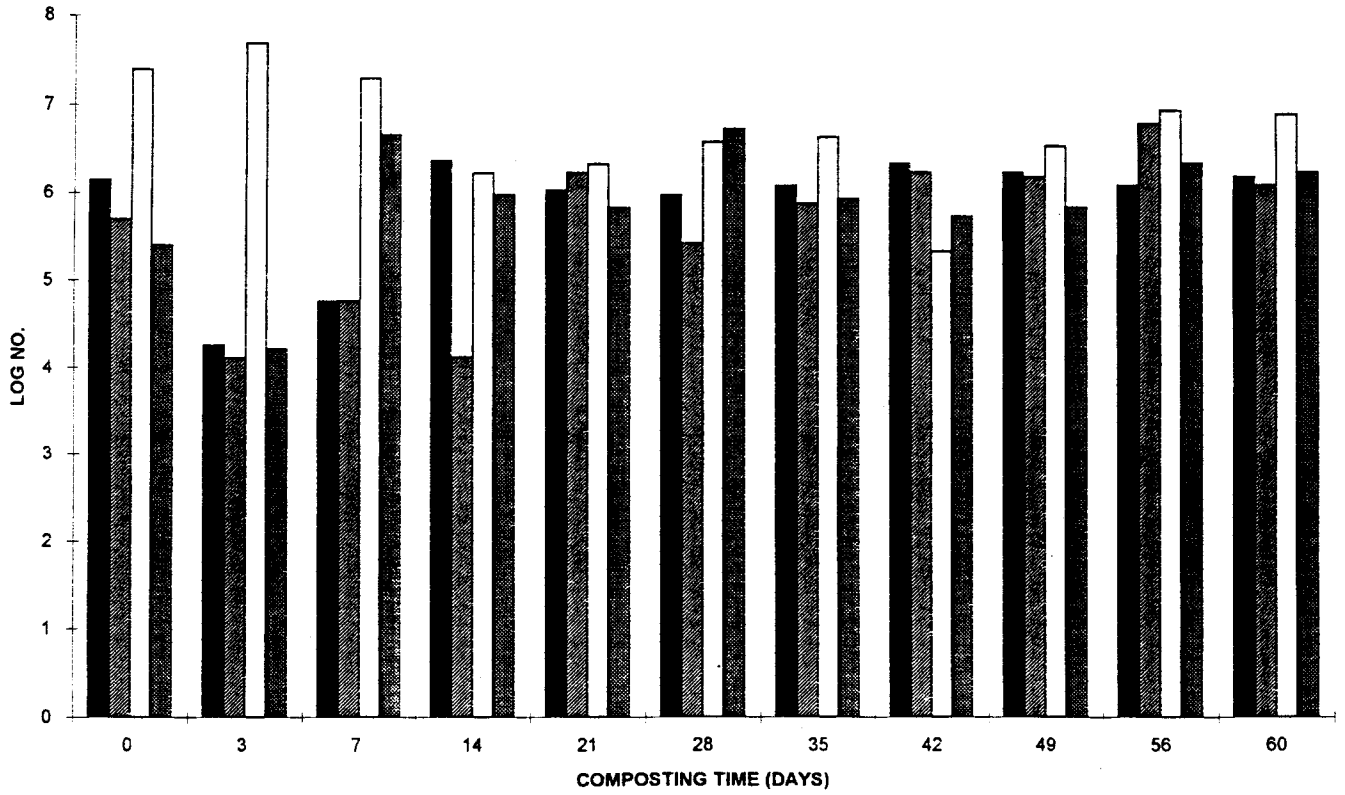


Fig. 7. Log numbers of thermophilic bacteria during the composting process. ■, Heap 1; □, Heap 2; □, Heap 3; ▨, Heap 4.

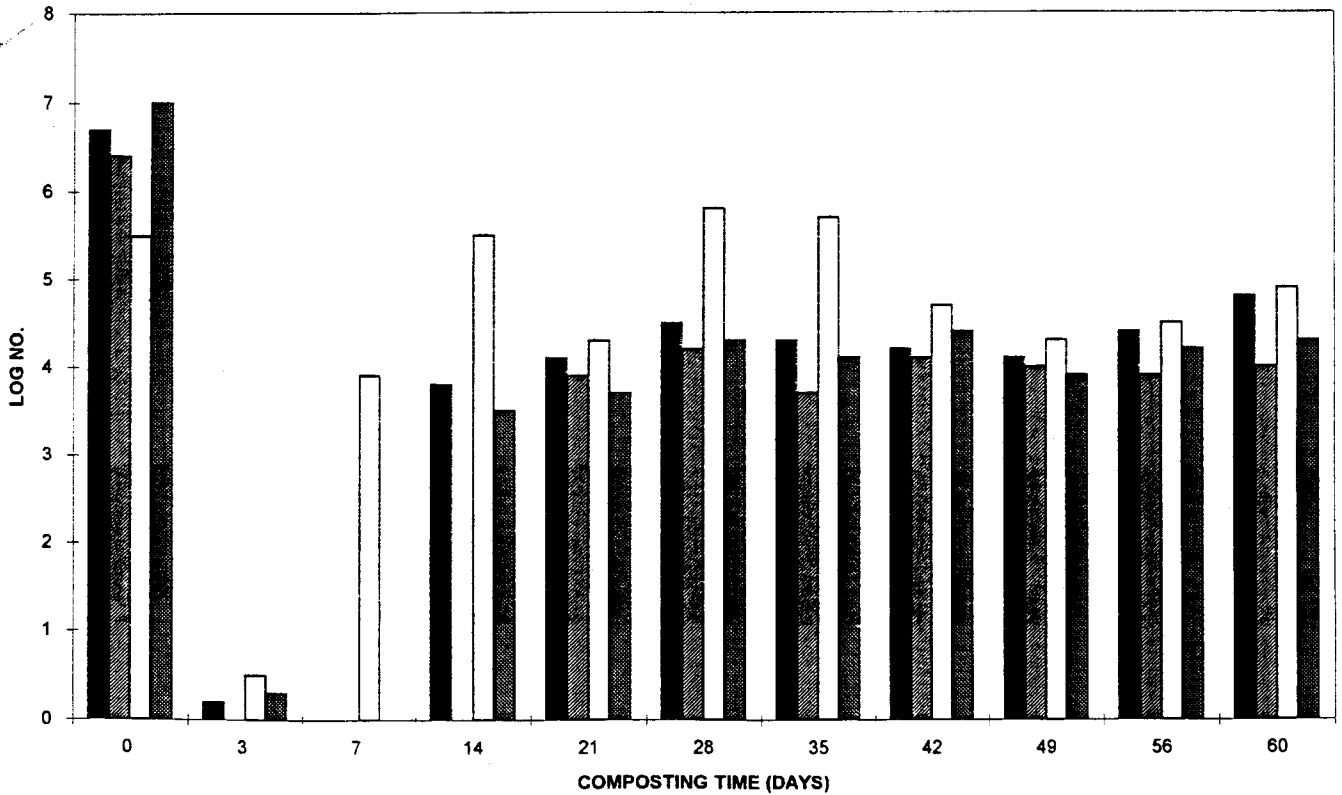


Fig. 8. Log numbers of mesophilic fungi during the composting process. ■, Heap 1; □, Heap 2; □, Heap 3; ▨, Heap 4.

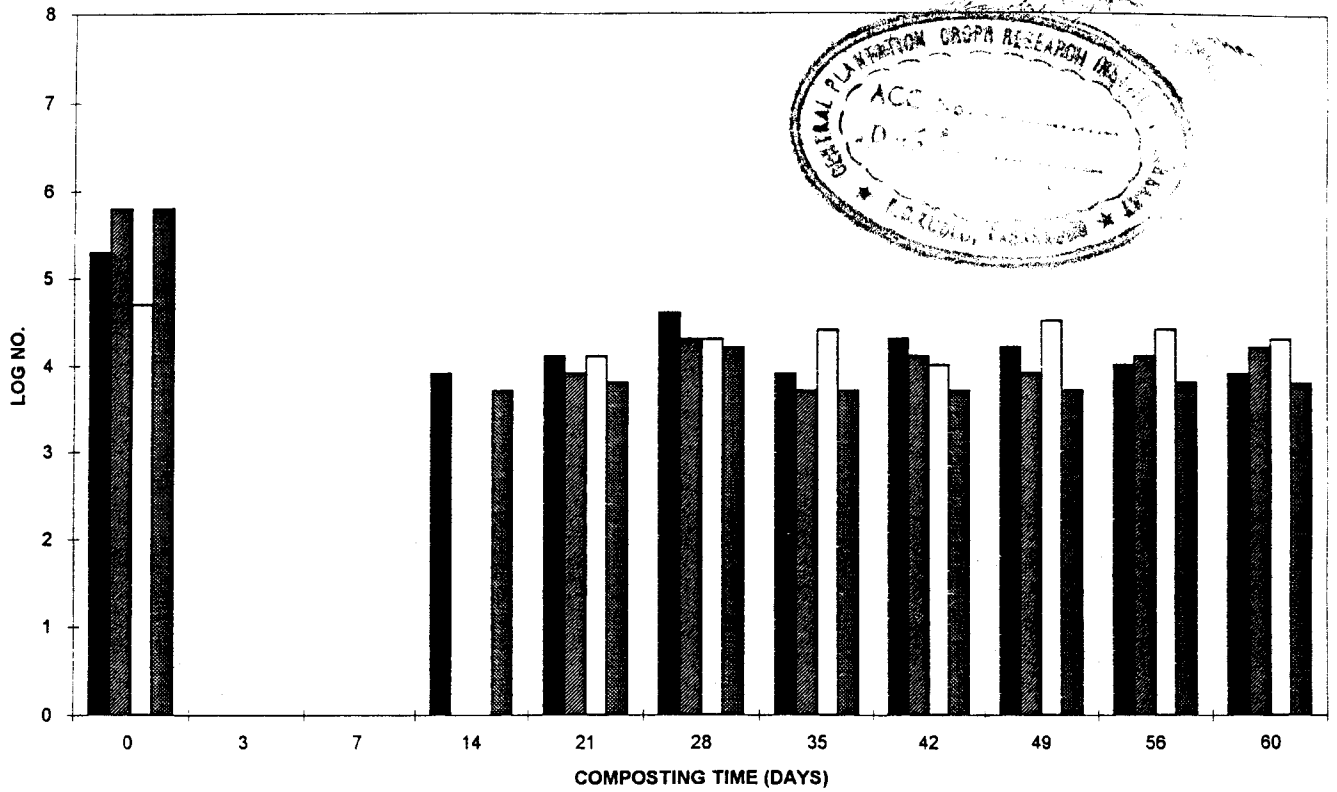


Fig. 9. Log numbers of thermophilic fungi during the composting process. ■, Heap 1; ▨, Heap 2; □, Heap 3; ▩, Heap 4.

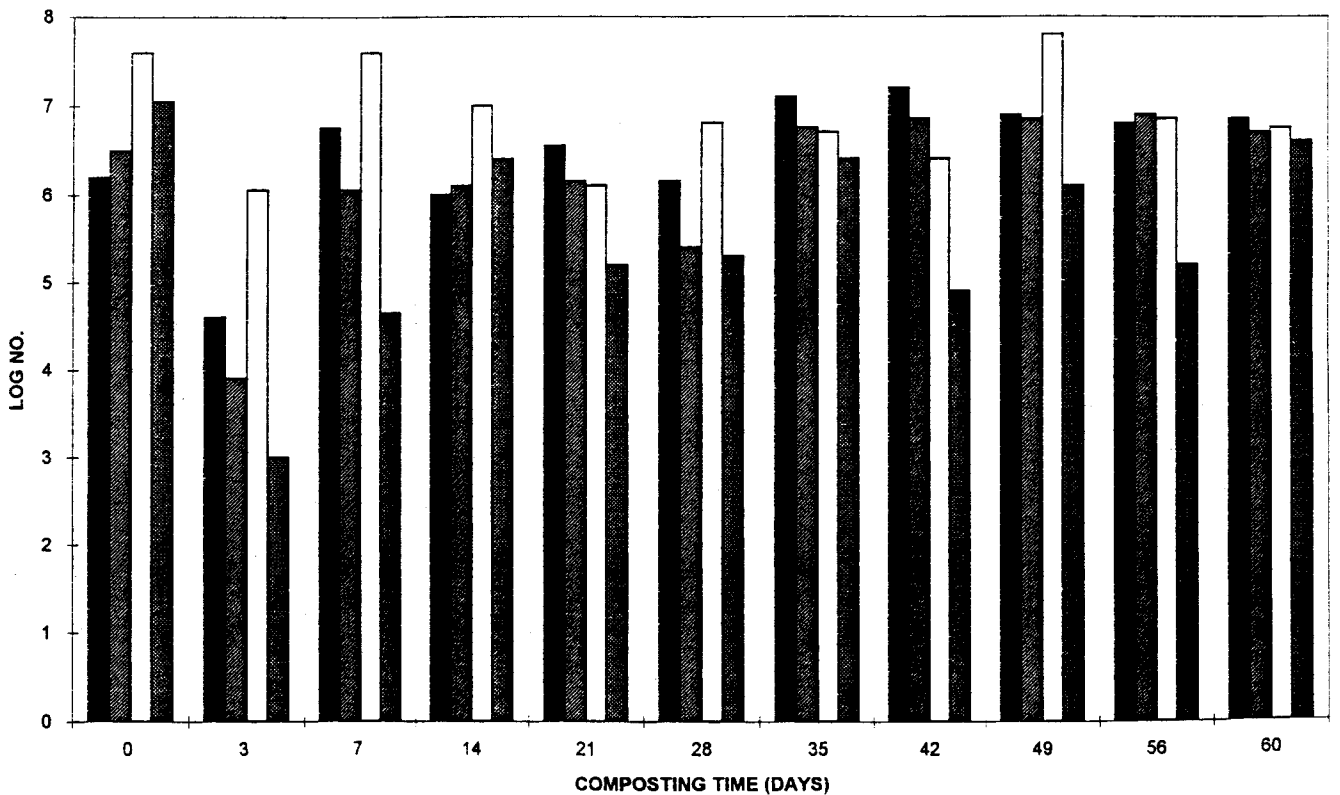


Fig. 10. Log numbers of mesophilic actinomycetes during the composting process. ■, Heap 1; ▨, Heap 2; □, Heap 3; ▩, Heap 4.

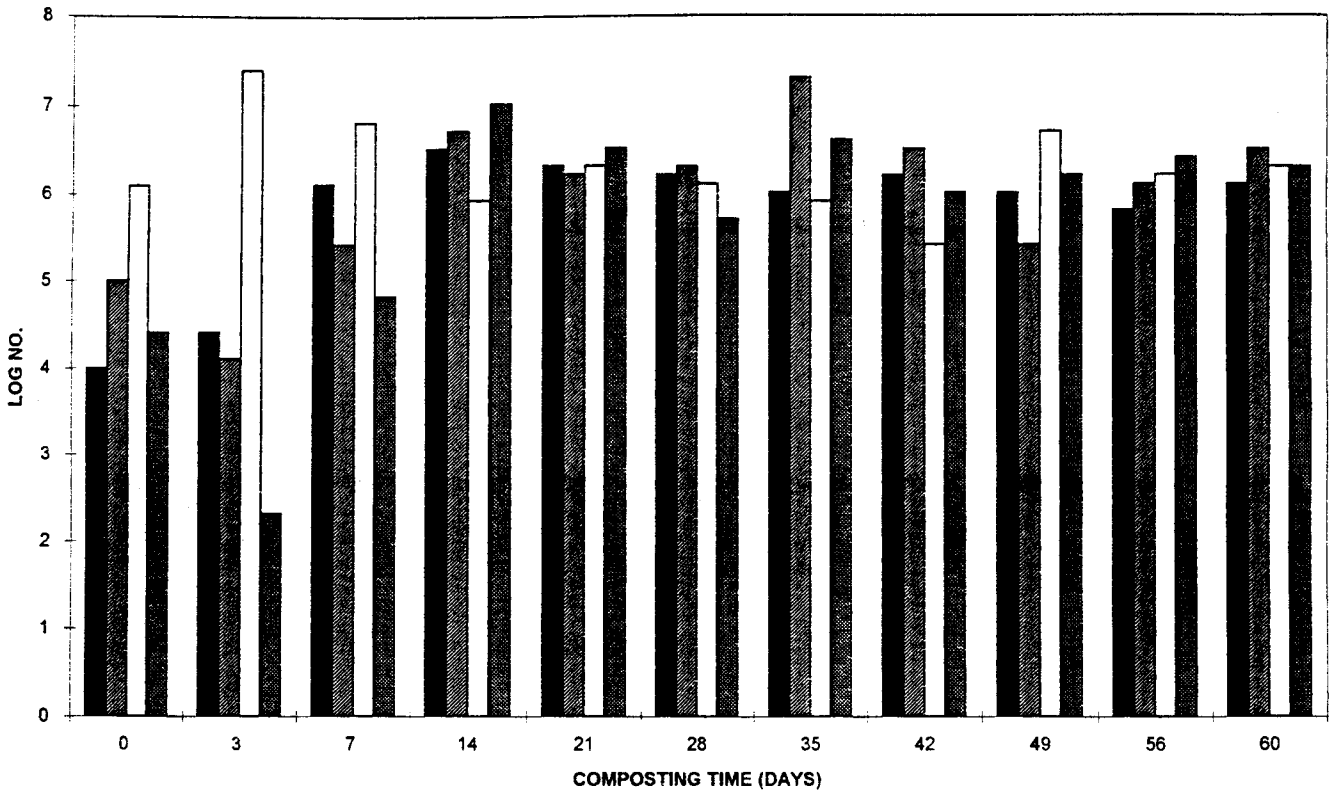


Fig. 11. Log numbers of thermophilic actinomycetes during the composting process. ■, Heap 1; ▨, Heap 2; □, Heap 3; ▩, Heap 4.

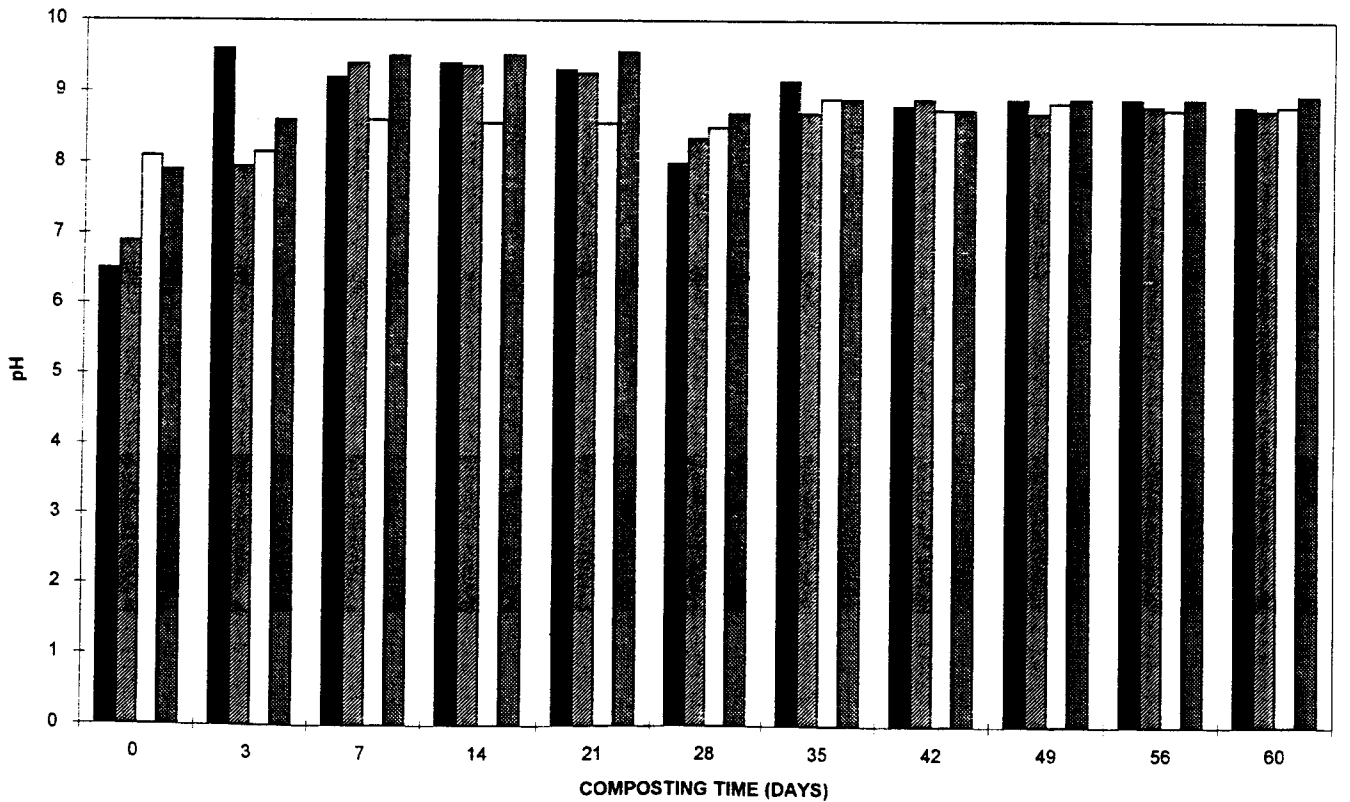


Fig. 12. The pH of the substrate during the composting process. ■, Heap 1; ▨, Heap 2; □, Heap 3; ▩, Heap 4.

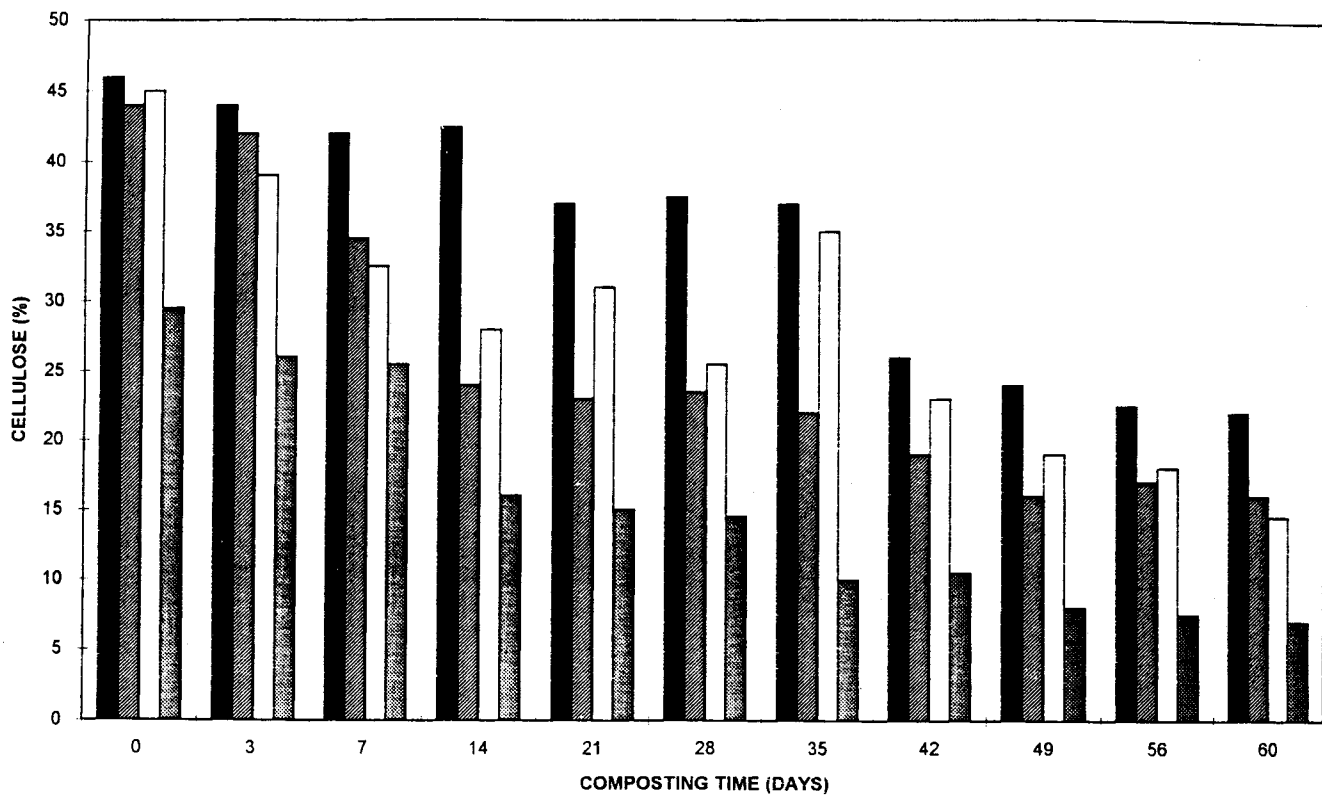


Fig. 13. Cellulose concentration during the composting process. ■, Heap 1; ▨, Heap 2; □, Heap 3; ▩, Heap 4.

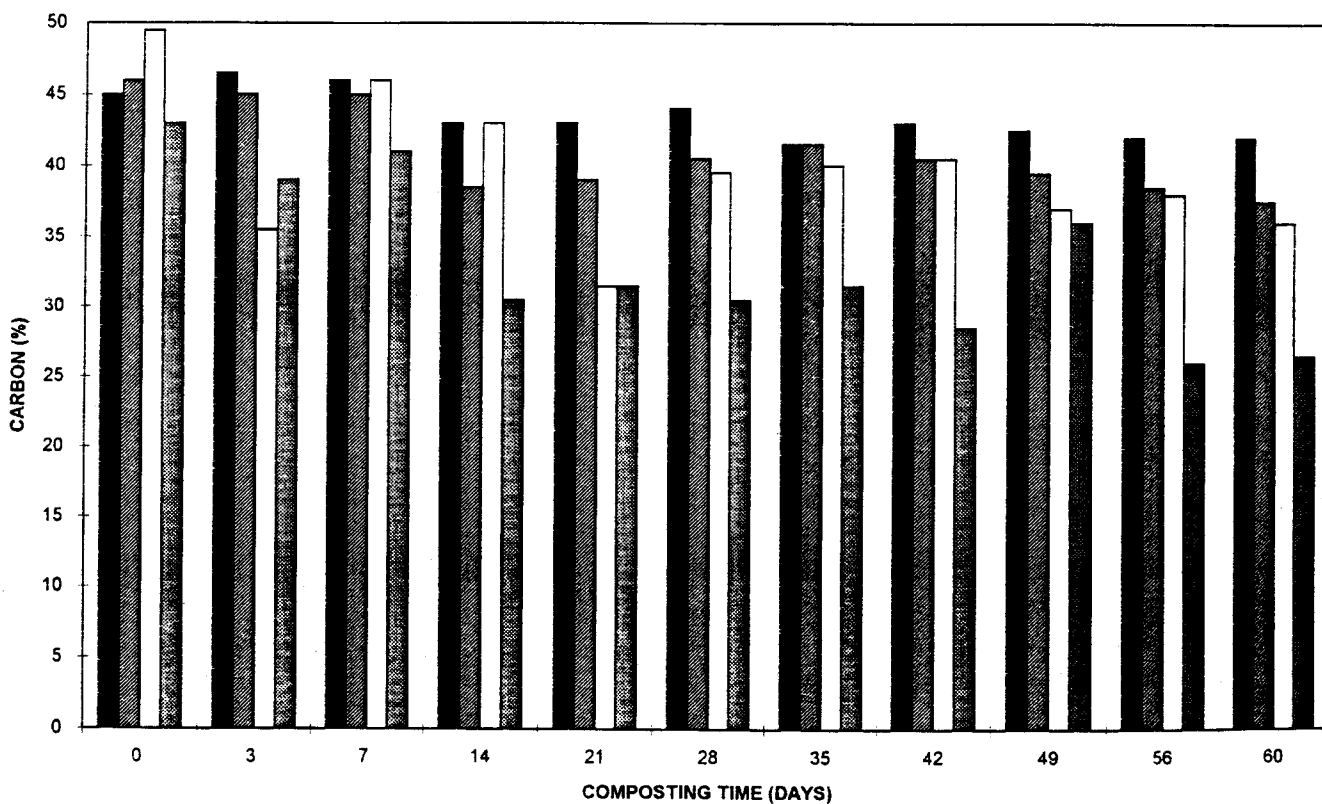


Fig. 14. Carbon concentration during the composting process. ■, Heap 1; ▨, Heap 2; □, Heap 3; ▩, Heap 4.

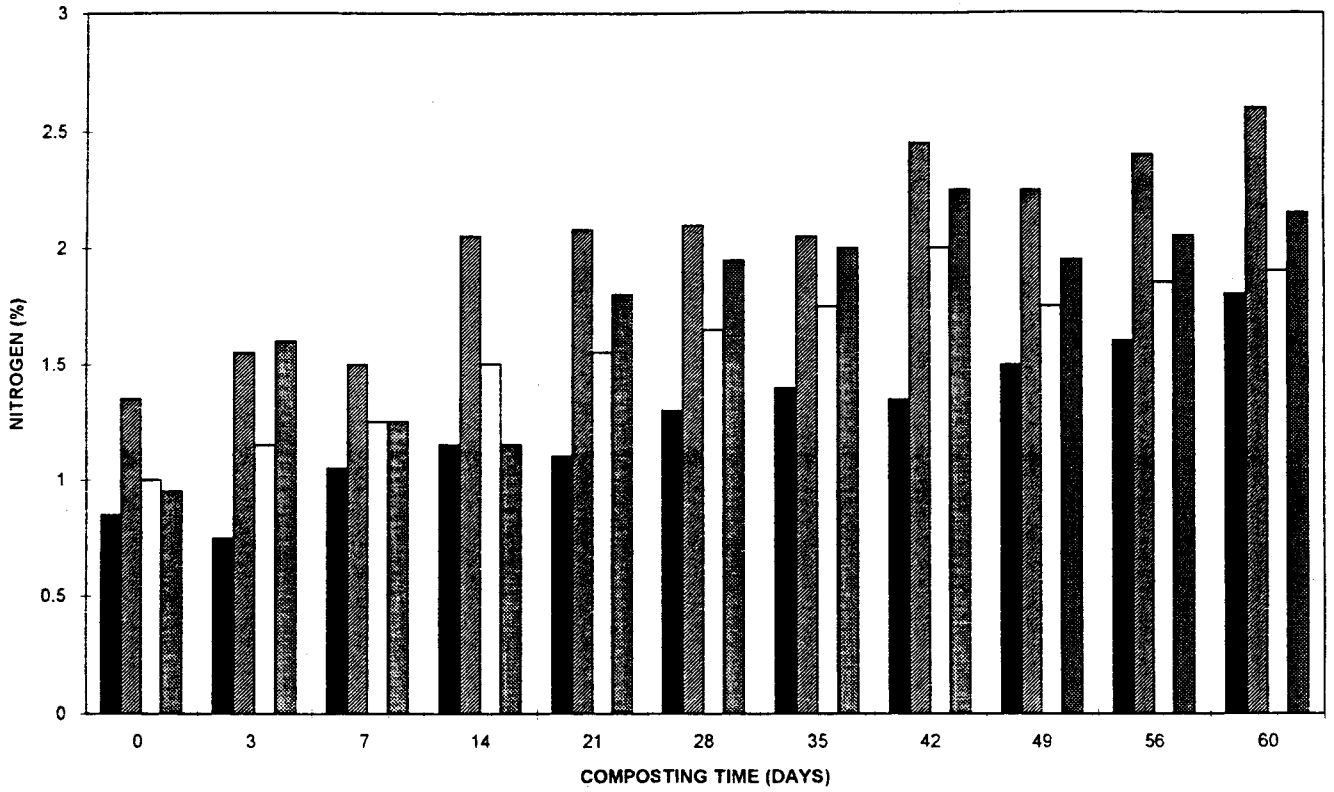


Fig. 15. Nitrogen concentration during the composting process. ■, Heap 1; ▨, Heap 2; □, Heap 3; ▩, Heap 4.

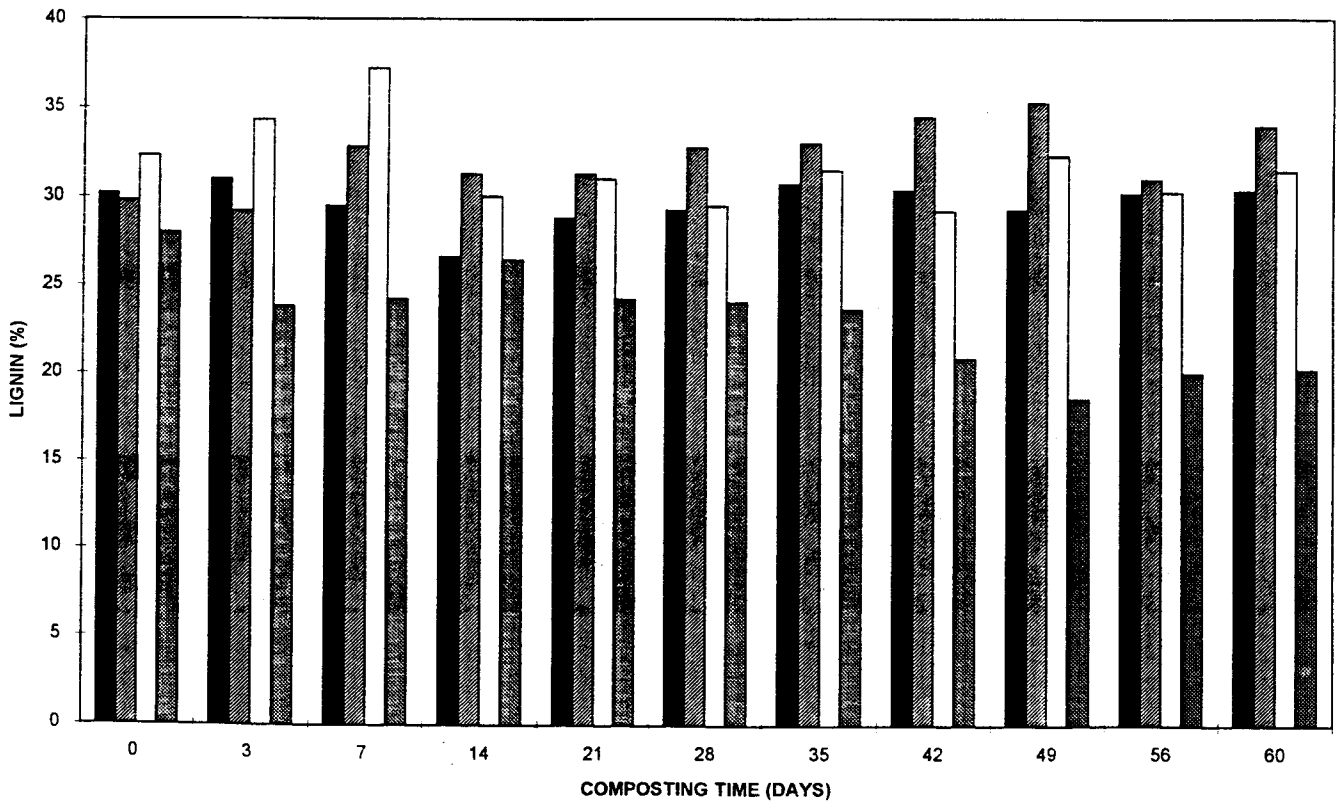


Fig. 16. Lignin concentration during the composting process. ■, Heap 1; ▨, Heap 2; □, Heap 3; ▩, Heap 4.

Table 1. ANOVA of C:N ratios, cellulose, lignin and pH with respect to heap and composting time

Source of variation	C:N ratio				Cellulose conc.				Lignin				pH			
	I	II	III	IV	I	II	III	IV	I	II	III	IV	I	II	III	IV
Co-variants (day)	2802	1	96	0.00	2928	1	273	0.00	4	1	1	0.31	1	1	2	0.09
Main effects (heap)	1757	3	20	0.00	2010	3	61	0.00	569	3	46	0.00	0.83	3	0.73	0.53
Residual	1102	38			425	39			157	39			14	39		
Total (corr.)	5662	42			5418	43			731	43			16	43		

I — Sum of squares.

II — d.f.

III — F-ratio.

IV — Sig. level.

between 5.4 and 8.2, and an average of 9.5 was achieved at the end of the process (Fig. 12). Nyns (1986) observed losses in ammonia in compost heaps which had low carbon:nitrogen ratios. However, nitrogen is generally stabilized in the process by being transformed into microbial protein.

Microbial decomposition of plant material resulted in an overall loss of cellulose and carbon (Figs 13 and 14) and an increase in nitrogen content (Fig. 15). These changes, which resulted from microbial activity on the cellulosic substrate and nitrogen from the dungs, increased microbial protein and humic substances. Although lignolytic microorganisms occur in compost heaps (Fergus, 1964), the breakdown of lignin was not noted (Fig. 16), concurring with an earlier report on palm press fibre (Thambirajah & Kuthubutheen, 1989).

Statistical analyses showed highly significant differences for changes in C:N ratio and cellulose in the four heaps during the composting process (Table 1). No significant difference was observed for changes in lignin and pH.

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REFERENCES

- Allen, S. E., Grimshaw, H. M., Parkinson, J. A. & Quarmby, C. (1974). *Chemical Analysis of Ecological Materials*, ed. S. E. Allen. Blackwell Scientific, Oxford, UK.
- AOAC (1975). *Official Methods of Analysis*, 12th edn. Association of Official Agricultural Chemists, Washington, DC, USA.
- Cappuccino, J. G. & Sherman, N. (1983). *Microbiology: A Laboratory Manual*. Addison-Wesley, Reading, Massachusetts, pp. 81–4.
- Chang, Y. & Hudson, H. J. (1967). The fungi of wheat straw compost. I: Ecological studies. *Trans. Brit. Mycol. Soc.*, **50**, 649–66.
- Chanyasak, V. & Kubota, H. (1981). Carbon/organic nitrogen ratio in water extract as measure of compost degradation. *J. Ferment. Technol.*, 215–19.
- Fergus, C. L. (1964). The thermophilic and thermotolerant molds and actinomycetes of mushroom compost during peak heating. *Mycologia*, **56**, 267–84.
- Frobisher, M., Hindsill, R. D., Crabtree, K. T. & Goodheart, C. R. (1974). *Fundamentals of Microbiology*. W. B. Saunders, Philadelphia; Toppan, Tokyo, Japan, p. 108.
- Garg, S. K. & Neelakantan, S. (1982). Bioconversion of sugar cane bagasse for cellulase enzyme and microbial protein production. *J. Food Technol.*, 271–9.
- Gomez, R. J. H. C. & Park, Y. K. (1983). Conversion of cane bagasse to compost and its chemical characteristics. *J. Ferment. Technol.*, 329–32.
- Gray, K. R., Sherman, K. & Biddlestone, A. J. (1971). A review of composting. Part I. Microbiology and biochemistry. *Process Biochem.*, **6**, 32–6.
- Husin, M., Zakaria, Z. Z. & Hassan, A. H. (1985). Potentials of oil palm by-products as raw materials for agro-based industries. *Proc. Natl. Symp. Oil Palm By-products for Agro-based Industries*, 5–6 November 1985, Kuala Lumpur, Malaysia.
- Jimenez, E. I. & Garcia, V. P. (1989). Evaluation of city refuse compost maturity: a review. *Biol. Wastes*, **27**, 115–42.
- MacGregor, S. T., Miller, F. C., Psarianos, K. M. & Finstein, M. S. (1981). Composting process control based on the interaction between microbial heat output and temperature. *Appl. Environ. Microbiol.*, 1321–30.
- Ministry of Primary Industries (1991). Planted area under oil palm: Malaysia. *Statistics on Commodities*, May 1991, pp. 33–4.
- Nyns, E. J. (1986). Can biomethanation be included in the processing of compost-like materials? In *Compost: Production, Quality and Use*, ed. M. De Bertoldi, M. P. Ferranti, P. L. Hermite & F. Zucconi. Elsevier, London, pp. 97–9.
- Poincelot, R. P. (1974). A scientific examination of the principles and practice of composting. *Compost. Sci.*, **15**, 24–31.
- Spaggiari, G. C. & Spigoni, G. L. (1986). Transformation of urban sludges mixed with grape stalks into organic fertilizers. In *Compost: Production, Quality and Use*, ed. M. De Bertoldi, M. P. Ferranti, P. L. Hermite & F. Zucconi. Elsevier, London, pp. 100–7.
- Statistical Graphics Corporation (1987). Statgraphics statistical graphic system, Version 2.6, 2115 East Jefferson Street, Rockville, MD 20852, USA. See *Stratgraphic User's Guide*, Sections 14.8–14.11.
- Taiganides, E. P. (1977). Composting of feedlot wastes. In

Animal Wastes, ed. E. P. Taiganides. Applied Science Publishers, London, pp. 241-51.

Thambirajah, J. J. & Kuthubutheen, A. J. (1989). Composting of palm press fibre. *Biol. Wastes*, **27**, 257-69.

Updegraff, D. (1969). Semimicro determination of cel-

lulose in biological materials. *Anal. Biochem.*, **32**, 420-4.

Zadrazil, R. F. & Brunnert, H. (1980). The influence of ammonium nitrate supplementation on degradation and *in vitro* digestibility of straw colonized by higher fungi. *Eur. J. Appl. Microbiol. Biotechnol.*, **9**, 37-44.