

Effect of Temperature on Bacterial Species Diversity in Thermophilic Solid-Waste Composting†

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Received 8 March 1985/Accepted 22 July 1985

Continuously thermophilic composting was examined with a 4.5-liter reactor placed in an incubator maintained at representative temperatures. Feed was a mixture of dried table scraps and shredded newspaper wetted to 55% moisture. One run at 49°C (run A) employed a 1:4 feed-to-compost ratio, while the other runs used a 10:1 ratio and were incubated at 50, 55, 60, or 65°C. Due to self-heating, internal temperatures of the composting mass were 0 to 7°C hotter than the incubator. Two full-scale composting plants (at Altoona, Pa., and Leicester, England) were also examined. Plate counts per gram (dry weight) on Trypticase soy broth (BBL Microbiology Systems) with 2% agar ranged from 0.7×10^9 to 5.3×10^9 for laboratory composting and 0.02×10^9 to 7.4×10^9 for field composting. Fifteen taxa were isolated, including 10 of genus *Bacillus*, which dominated all samples except that from run A. Species diversity decreased markedly in laboratory composting at 60°C and above, but was similar for the three runs incubated at 49, 50, and 55°C. The maximum desirable composting temperature based on species diversity is thus 60°C, the same as that previously recommended based on measures of the rate of decomposition.

The management of various solid wastes is increasingly troublesome and costly, especially in densely populated metropolitan areas. As a result, composting has come into widespread use as a treatment process for one type of solid waste, i.e., sewage sludge. By May 1983, over 60 full-scale sludge-composting plants were in operation in the United States, with more than 20 others in the design stage or already under construction (42). Composting also can be used for the treatment of the organic fraction of municipal refuse, including household garbage and most yard wastes, and for various organic industrial wastes.

Deliberate control of the composting process at the microbial level involves four interrelated factors, i.e., metabolic heat generation, temperature, ventilation, and moisture content (15, 23). A critical element of control is maintenance of temperature in a desirable range, since temperature both reflects prior microbial activity and powerfully determines the current rate of activity. In particular, the composting ecosystem tends to limit itself due to inhibitive high temperatures produced by the excessive accumulation of heat. This key relationship has previously been elucidated by assessing the response of this ecosystem to temperature by physical and chemical measurements, such as heat output (29), drying (a manifestation of heat output; 15), carbon dioxide production (3, 24, 29, 36, 40, 41), oxygen uptake (22, 40, 41), labeled-carbon substrate incorporation (24), and dry-weight loss (36, 40, 41). According to such studies, the threshold to substantial self-limitation is approximately 60°C (3, 15, 24, 36).

The effect of temperature on the microbial community structure of the composting ecosystem is now reported. The assessment is based on cultivable thermophiles and is summarized in the form of a species diversity index. This represents one of the few (2) published descriptions of a species diversity index for a microbial ecosystem.

MATERIALS AND METHODS

Laboratory composting unit. Bench-scale continuously thermophilic composting was carried out in the laboratory in a 4.5-liter reaction vessel (Fig. 1). Air flow was adjusted manually to maintain 10 to 18% O₂ in the outlet gas. Temperature in the center of the composting mass was recorded continuously. Samples for pH and moisture determinations were withdrawn periodically immediately before and after feeding. The total mass of the composting material was measured at both the start and the end of each feeding cycle. Microbiological samples were collected at the termination of each composting run.

Feed. Feed to the reactor consisted of an equal mixture by weight of dried table residue and shredded newspaper (8-mm-wide strips), with distilled water added to give a moisture content of approximately 55% (wet weight). This mixture consisted of approximately 98% (dry weight) volatile matter and had a pH of 5.6.

The dried table residue was prepared by collecting the table waste from 1 day of operation of a university dining facility serving 10,000 meals per day. Principal components of this residue were food wastes and paper (napkins, wrappers, etc.), with some plastic also present. Before collection, the waste had been mixed with water and processed in a Somat wet pulper. Approximately 0.4 m³ of pulp was dried for 4 days by forced air in a crop-drying oven to a final moisture content of 5%. It was then screened (13-mm-mesh screen) and stored at room temperature in plastic bags.

Establishment of the population. For the first laboratory run (run A), the incubator temperature, which established the minimum composting temperature of the material, was set at 49°C. The initial charge of feed was inoculated with small amounts of activated sludge, raw sewage, garden soil, and tap water. Every 3 to 4 days, the mass was removed from the reactor, a small portion was discarded, and the remainder (600 g [wet weight]) was thoroughly mixed with 150 g (wet weight) of feed (feed/compost ratio, 1:4) to recharge the reactor. This composting procedure, which corresponds to that of an intermittently charged, intermit-

† New Jersey Agricultural Experiment Station publication no. D-07526-1-85.

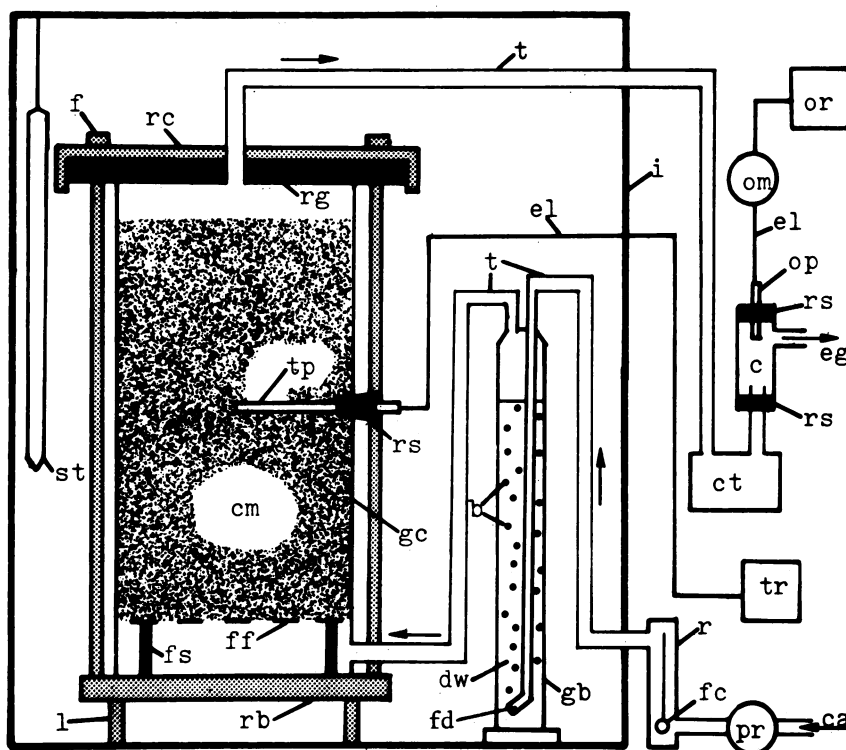


FIG. 1. Schematic of laboratory composting apparatus. Abbreviations: b, air bubbles; c, chamber, compressed air; cm, composting mass; ct, condensate trap; dw, distilled water; eg, exhaust gas; el, electrical lead; f, fastener; fc, flow controller; fd, fritted disc; ff, false floor (stainless steel); fs, false-floor support; gb, gas-washing bottle (one or two in series); gc, glass cylinder with bottom; i, incubator; l, legs; om, oxygen meter (Beckman Oxygen Analyzer model 777); op, oxygen probe; or, oxygen recorder (Sargent model SR); pr, pressure reducer; r, rotameter; rb, reactor base; rc, reactor cover; rg, rubber gasket; rs, rubber stopper; st, stem thermometer; t, tubing; tp, thermistor probe (Rustrak model 1332); tr, temperature recorder (Rustrak model 2133).

tently completely mixed reactor, was continued for a period of several months prior to microbiological analysis.

All subsequent laboratory composting was performed with a feeding pattern corresponding to a nominal plug-flow reactor with moderate dispersion. Each initial charge of feed was inoculated with small amounts of activated sludge, raw sewage, soil, and water collected from several laboratory baths (44.5, 50, and 65°C) and mixed in a 10:1 ratio with frozen compost saved from previous runs. After 3 days of incubation, the mass was removed, and a small amount was mixed with additional feed, again in a feed/compost ratio of 10:1. This mixture (about 700 g [wet weight]) was incubated for 5 more days, during which time it was removed once for mixing without refeeding. It was then fed again in the same way. During 5 additional days of incubation, the mass was removed twice for mixing without feeding. Thus the total composting time for each run was 13 days, which included two feedings and three additional mixings after initial start-up. Composting in this fashion was performed at incubator temperatures of 55, 65, 50, and 60°C (runs B through E).

For run C (65°C), a slight modification was necessary (see Results). The feed was brought up to 55% moisture by the addition of distilled water containing 1 mg of $\text{Ca}(\text{OH})_2$ per ml, thus raising the initial pH of the mixture to 6.0 to 6.3. Run G was identical to run C except that the pH was not adjusted.

Field composting. Microbiological analysis was also performed on samples from two full-scale solid-waste-composting operations.

The Fairfield-Hardy unit at Altoona, Pa., was described in detail by Schulze (33) but has since undergone some modi-

fication. At the time of sampling, this plant was receiving the curbside collection from a city of 70,000 people and composting approximately 125 tons of waste per week. Detention time in the reactor was estimated to be 4 days with daily feeding and a spiral plug-flow pattern which involved considerable dispersion. This treatment was followed by curing in windrows if a stabilized compost was desired. Temperatures in the reactor were routinely 65 to 70°C, with 16% interstitial oxygen and a 50 to 55% moisture content. Sampling (run F) was performed prior to the daily feeding. Temperatures were taken by inserting a 3-ft (0.9-m)-long Tel-Tru 3-in. (76-mm) dial thermometer (Brooklyn Thermometer Co., Inc.). A 0.7-m-deep hole was dug in the mass, and a large sample (about 4 kg) was removed and placed in a plastic bag. Four such samples were taken, two from a short distance behind the row of augers at about 1.2 and 3.7 m from the central overflow standpipe and two on the opposite side of the reactor at the same distances from the center. The samples taken 1.2 m from the overflow pipe were estimated to represent a 4-day residence in the reactor, and the samples taken 3.7 m from the pipe represented a 2-day residence. The samples were placed in insulated containers for transport (6 h) to the laboratory.

The composting plant in Leicester, England, employed six Dano Bio-Stabilizers (drums) which processed 1,200 tons of refuse and 500 tons of wet, digested sewage sludge (2% solids) per week. Material exiting the drums was screened and then cured in windrows for 6 weeks. This plant was described by Hughes (21). Mass transport in the drums was nominally plug-flow, but considerable dispersion occurred. Two samples were collected from stabilizers, and two were

TABLE 1. Results of laboratory composting

Run ^a	Feeding pattern ^b	Temp (°C)			Maximum O ₂ uptake (mg of O ₂ /g [volatile matter] per h) ^c	pH		Moisture (%)		Dry wt loss (%)	Plate counts ^c		No. of colonies picked	
		Incubator ^d	Probe maximum ^e	Isolation ^f		Start	Finish	Start	Finish		×10 ⁶ /g (dry wt)	% Increase, day 2	Randomly	Total
A	cm	49	55	48	1.3	8.0	8.2	62	67	15	53	203	59	127
D	pf	50	57	45	1.8	6.1	8.7	57	62	21	36	100	57	107
B	pf	55	61	48	1.5		8.6	55	57	15	7	19	65	92
E	pf	60	65	53	1.7	6.2	8.5	62	63	14	17	14	62	91
G	pf	65	65		0.04	5.1	4.8	58	52	-5	ND	ND		
C ^h	pf	65	69	55	0.9	6.0	8.3	59	55	3	37	10	46	83

^a Arranged in order of increasing temperature. Values refer to last 3.5-day (run A) or 5-day (runs B through E and G) feeding cycle.

^b Intermittently completely mixed reactor with intermittent feed (cm) or plug-flow reactor with moderate dispersion (pf).

^c Geometric mean of duplicate samples, each plated in triplicate, after 20 h of incubation. ND, None detected (<0.00005 × 10⁶/g).

^d Incubator used to house reactors and for plate counts, measured via the stem thermometer (Fig. 1).

^e Measured via thermistor probe (Fig. 1).

^f Used for isolation and testing of cultures.

^g Based on airflow rate (1.4 to 4.2 liters/h) and minimum exhaust O₂.

^h Initial pH of feed adjusted with Ca(OH)₂.

collected from windrows. The stabilizer samples were of material which had exited the drums several minutes earlier and had been screened. A 0.6-m-long probe inserted through a port in each drum halfway along its length, with rotation stopped, provided a reading intermediate between ambient (15°C) and the actual temperature of the mass, since the probe was sensitive to temperature along its entire length. The 2-week-old windrow that was sampled had been turned once, but not within the previous week. The sample was collected at a height of about 1 m, and 35 cm from the surface. A mercury stem thermometer was used to determine temperature. The 6-week-old windrow, which had been turned several times but not within the previous week, was sampled in the same way. The samples were placed in plastic bags and processed initially in facilities at the county analyst's laboratory located on the same grounds.

Physical and chemical methods. Measurements for pH were made with a dilute slurry of 10 g (wet weight) of sample in 500 g of distilled water which was swirled for 5 min and then allowed to settle (6). Duplicate moisture determinations were made by drying 10 to 30-g subsamples overnight at 103°C, except in Leicester, where 30 to 50-g samples were dried at 120°C.

Enumeration. Samples (20 g) were placed into sterile stainless-steel blender cups, with 270 ml of phosphate-buffered dilution water (1) added, and blended for 30 s at maximum speed. Tenfold serial dilutions were made in phosphate-buffered water, and 0.1 ml of appropriate dilutions was surface plated in triplicate on Trypticase soy broth (BBL Microbiology Systems, Cockeysville, Md.) with 2% agar. These plates had been predried overnight at 45 to 65°C to minimize spreading of colonies. Duplicate samples were diluted and plated for laboratory composting runs. Plating was completed within 2 h of sampling for laboratory composting samples, 10 h for Altoona, and 6 h for Leicester. All plates were incubated for 20 h, and some were then reincubated for an additional 24 h. Where possible, dilutions giving 10 to 100 colonies per plate were used in calculating bacterial concentrations.

Isolation and identification. Selection of colonies for isolation was by two procedures. Random selection consisted of picking all colonies from a plate or sector. Deliberate selection consisted of picking some of the less common colony types from the spread plates. Cultures which were success-

fully transferred to fresh plates of Trypticase soy broth (BBL) with 2% agar were streaked again onto soil extract agar slants (18) for storage. Unsuccessful streakings were repeated from the same colony where possible. Isolated cultures were identified by standard techniques and references (4, 8, 9, 18, 28, 34; P. F. Strom, Ph.D. Thesis, Rutgers University, New Brunswick, N.J. 1978).

RESULTS

Table 1 summarizes the results from the laboratory composting runs. Active composting occurred in all runs except run G as indicated by the (i) 4 to 7°C temperature elevation within the mass relative to the incubator temperature (self-heating), (ii) rate of O₂ utilization, (iii) increase in pH, (iv) loss in dry weight, and (v) increase in plate count. Plating of the feed with incubation at 50°C yielded no colonies, corresponding to a plate count of <5,000/g (dry weight). Run G was inactive by all five of these criteria (the apparent gain in dry weight is attributed to experimental error).

Incubating the spread plates for an additional 24 h resulted in large increases in counts at the two lower composting temperatures (runs A and D), but small changes at higher temperatures. All random and some deliberate colony selections were performed after the shorter incubation period to minimize the problems of spreading and satellite colonies.

Comparable details for the field composting runs are given in Table 2. At Altoona, samples F1a and F1b were intended to be replicates, as were F2a and F2b, representing material of similar ages from opposite sides of the reactor. Except that F1a was 6°C cooler than F1b, the results were similar. Temperatures and pH increased slightly with composting time, while moisture content and plate counts decreased. The plate counts are almost 2 orders of magnitude lower than those for the laboratory composting samples. Only sample F2b showed a substantial increase in plate count with an additional 24 h of incubation.

At Leicester, samples I1 and I2 were intended to represent similar materials from two different drums. However, the plate counts differed dramatically until after day 2 of incubation. Plate counts for the windrow samples were 1 to 2 orders of magnitude lower.

Table 3 gives the identification of the isolated microorganisms. (For details of the identifications and a discussion of

TABLE 2. Results of field composting

Facility	Sample no.	Sampling site	Composting conditions				Plate count		No. of colonies picked	
			Time (days)	Temp (°C)	pH	Moisture (%)	×10 ⁸ /g (dry wt)	% Increase, day 2	Randomly	Total
Altoona ^a	F1a	3.7 ^b	2 ^c	59	5.5	60	0.52	4	48	67
	F1b	3.7 ^b	2 ^c	65	5.6	61	0.60	7	54	56
	F2a	1.2 ^b	4 ^c	66	5.8	58	0.16	13	15	17
	F2b	1.2 ^b	4 ^c	66	5.8	59	0.26	83	24	37
Leicester ^d	I1	Drum f	4 ^e	>42	8.6	65	74	6	56	60
	I2	Drum b	4 ^e	>35	8.9	69	3.9	870	55	58
	I3	Windrow	14 ^e	66	9.0	50	0.34	60	56	61
	I4	Windrow	42 ^e	55	8.9	56	0.89	39	55	60

^a Samples collected before daily feeding on 27 May 1977. Plate counts were incubated at 61 to 62°C, and isolation and testing of isolates were at 54°C.

^b Distance in meters from central overflow standpipe. Samples a were from just behind the row of augers, and samples b were from the opposite side of the reactor.

^c Estimated time in reactor based on distance from center.

^d Samples collected 5 September 1977. Plate counts, isolation, and testing of isolates were at 48 to 49°C.

^e Estimated detention time of drums or age of windrows.

the significance of the organisms observed, see the accompanying paper [34]; Strom, Ph.D. thesis). Except in run A, members of the genus *Bacillus* made up a large majority of the isolates. Runs C and E, involving composting at 60°C and above, yielded virtual monocultures of *Bacillus stearothermophilus*. Samples IA, IB, and ID yielded almost entirely *Bacillus circulans*. Isolation failures (i.e., colonies "lost" upon initial transfer from spread plates) were substantial only in run C.

The relative species diversities of the communities, based on isolates from the different laboratory composting runs, are given in Table 4. Diversity dropped markedly at composting temperatures above 60°C. Field samples were not included because of the differences between composting and plating temperatures.

DISCUSSION

An important consideration in this study is the suitability of the laboratory composting procedure. The explicit purpose of this study was to isolate temperature as a variable and examine its effect on community structure. This goal was accomplished, and the procedure is expected to be satisfactory based on several considerations. The feed was representative of the portion of residential wastes in the United States that can be composted, which consists largely of paper, food, and yard wastes (10), and it had a comparable pH (16, 27). Moisture content and oxygen levels were chosen to be in the optimal range (36) and were consistent with other reports (17, 27, 32) and field practice at Altoona and Leicester.

Operational results from the laboratory studies are also

TABLE 3. Distribution of isolates among taxa^a

Organism:	% Isolates per sample in run (°C) ^b :									
	A (49–55)	D (50–57)	B (55–61)	E (60–65)	C (65–69)	F1a and b (59–65)	F2a and b (66)	I1 and 2 (>35)	I3 (66)	I4 (55)
Fungus										
<i>Aspergillus fumigatus</i>	–	–	17	–	–	–	–	–	–	–
Actinomycete										
<i>Streptomyces</i> sp.	12	2	–	– ^c	–	– ^d	–	–	–	2
<i>Thermoactinomyces</i> sp.	–	–	+	– ^c	–	– ^d	+	–	–	–
<i>Bacillus</i> sp. (total)	(23)	(77)	(78)	(100)	(83)	(99)	(100)	(99)	(98)	(98)
<i>B. licheniformis</i>	5	+	+	–	–	2	–	+	+	+
<i>B. subtilis</i>	–	–	–	–	–	–	–	+	–	–
<i>B. coagulans</i> type A	–	5	42	2	–	–	–	–	–	–
<i>B. coagulans</i> type B	–	42	2	–	–	69	56	–	–	–
<i>B. circulans</i> complex	3	19	8	–	–	18	10	99	45	91
<i>B. stearothermophilus</i>	+	4	20	97	83	8	33	–	14	–
<i>B. brevis</i>	3	+	5	–	–	–	–	–	12	2
<i>B. sphaericus</i>	–	–	3	–	–	2	–	–	27	5
Unassigned <i>Bacillus</i> sp. type i	12	–	–	–	–	–	–	–	–	–
Unassigned <i>Bacillus</i> sp. type ii	–	7	–	2	–	1	–	–	–	–
Nonsporeformer										
Type a	17	21	–	–	–	–	–	–	–	–
Type b	47	+	+	–	–	–	–	–	–	–
Lost	2	+	5	+	17	1 ^d	–	1	2	–

^a Numbers refer to percentage of random isolates for sample. +, Found only among nonrandom isolates (present in low numbers); –, not found in sample.

^b Runs A through E were laboratory runs, runs F1a and b and F2a and b were from Altoona, Pa., and runs I1 through I4 were from Leicester, England.

^c Two actinomycetes were lost prior to isolation or identification.

^d One actinomycete was lost prior to isolation or identification.

TABLE 4. Diversity of isolates from laboratory composting

Run ^a	Temp (°C)	No. of isolates		No. of species among isolates:		No. of species among randomly picked isolates to account for		Species diversity index ^b
		Randomly picked	Total	Randomly picked	Total	>50%	>90%	
A	49–55	59	127	7	8	2	5	0.65
D	50–57	57	107	7	10	2	5	0.67
B	55–61	65	92	7	10	2	5	0.66
E	60–65	62	91	3	4 ^c	1	1	0.07
C	65–69	46	83	1 or 2 ^d	1 or 2 ^d	1	1 or 2 ^d	0

^a Arranged in order of increasing compost temperature. Values given are based on the assumption that lost cultures included no new taxa, except where noted.

^b Shannon index: $H = \sum P_i \log P_i$, where P_i is the number of individuals in i th species as a fraction of the total number of individuals in the sample (26). Values are based on random isolates, excluding those lost.

^c Includes unidentified lost actinomycete.

^d Depends on identity of lost colonies.

consistent with other work. The maximum oxygen utilization rates and thermophilic plate counts from Table 1 are compared with values reported in the literature in Table 5. The oxygen utilization values fit well into the lower part of the range given in the literature, as might be expected in

view of the abundance of relatively inert paper provided in our feed. (The 50% of dry weight accounted for by the newspaper was employed primarily as a bulking agent to provide porosity.) The plate counts, on the other hand, were near the maximum reported in the literature (Table 5). This

TABLE 5. Thermophilic plate counts and oxygen utilization rates in various composting materials

Material	Reactor		No. of trials	Temp (°C)		Count ($\times 10^8$ /g (dry wt))	Rate ^a		Reference
	Type	Size		Composting	Plating		mg of O ₂ /g per h	Basis	
Refuse ^b	Laboratory	4.5 liters	5	49–69	49–65	7–53	0.9–1.8 ^c	VM	This work
Refuse	Altoona	200 m ³	1	59–66	61–62	0.2–0.6			This work
Refuse-sludge	Leicester-Dano	250 m ³	2	>35	48–49	3.9–74			This work
Refuse-sludge	Leicester-windrow	2.5 m high	2	55–66	48–49	0.3–0.9			This work
Refuse	Laboratory	42 liters	7	50–70			2.5–6.3	VM	22
Refuse	Shaker flask	0.5 liters	13	50–67			3.1–9.3 ^{c,d}	VM	22
Refuse ^b	Laboratory	4.5 liters	15	55–61	55	4.3–33 ^c			Morris ^e
Refuse ^b plus G or A ^f	Laboratory	4.5 liters	2	55–61	55	8.7–9.9 ^c			Morris ^e
Refuse-sludge	Dewar flask	1 liter	2	74–76 ^c	55	2.1–13 ^c			25
Garbage	Laboratory	21 liters	3	50–62			3.3–5.0 ^c	VM	31
Garbage	Closed, 95% O ₂	Small	6	50–65			1.7–11 ^c	DW	31
Garbage-sludge	Laboratory	200 liters	1	50–68			4.1–7.3	VM	32
Straw	Dewar flask	1 liter	2	72–73 ^c	55	0.6–1.8 ^c			25
Straw	Adiabatic	1 liter	7	50–76			2.1–3.2 ^{c,d}	AD	5
Straw-manure	Pot	150 g (dry wt)	1	50	50	40 ^c			38
Straw-manure	Stack	24 m ³	1	70 ^c	53	4.3 ^{c,h}			20
Straw-manure plus S ^f	Stack	24 m ³	1	70 ^c	53	110 ^{c,h}			20
Straw plus NH ₄ NO ₃	Bin	1 m ³	1	79 ^c	45	20 ^c			7
Straw plus NH ₄ SO ₄	Incubated jars	30 g (dry wt)	3	60 ^c		1.5–6.5 ^c			37
Hay	Stack	50 m ³	1	63 ^c	60	1.8 ^c			19
Hay	Stack	50 m ³	1	63 ^c	60	110 ⁱ			19
Hay	Dewar flask	4 liters	1	63 ^c	60	13 ^c			13
Hay	Adiabatic	2 liters	12	50–75+			0.5–7.4 ^{c,d}	DW	30
Grass	Adiabatic	2 liters	1	50–75+			3.1 ^{c,d}	DW	30
Wool	Adiabatic	2 liters	1	50–73	60	0.2–0.8			39
Wool	Adiabatic	2 liters	1	50–74	60	3.9–6.5			29
Wool	Glass flask	2 liters	4	50–74			0.3–0.5 ^{d,j}	DW	29
Wool	Adiabatic	2 liters	4	56–80 ^c	50	0.4–3.5 ^c			12
Wool	Adiabatic	2 liters	16	50–70			0.1–1.1 ^{c,d}	DW	12

^a Rate is measured per gram of: VM, volatile matter; DW, dry weight; or AD, air-dried material.

^b Mixture of dried table scraps, and shredded newspaper.

^c Maximum value over a period of time.

^d Based on reported rate of CO₂ evolution and respiratory quotient (31) of 0.85 ml of CO₂ evolved per ml of O₂ consumed.

^e Morris, M.S. thesis.

^f Glucose (G), alanine (A), or sucrose (S) supplement.

^g Maximum.

^h Wet weight basis.

ⁱ Maximum count for cross-section of stack, 186 days after assembly.

^j Measured after 24 h at each temperature.

result may be due in part to the favorable conditions of aeration and moisture provided and to the good mixing possible with the small composting mass. The plating procedure and medium were selected based on previous work indicating high recoveries (11; M. L. Morris, M.S. thesis, Rutgers University, New Brunswick, N.J., 1975). Additionally, the use of an incubation temperature for the plate counts which corresponded to the composting temperature helped ensure maximal recoveries. The plate counts for Altoona and the Leicester windrows were lower than those for the laboratory composting samples, while counts for the Dano drums were comparable to those of laboratory samples.

A final validation of the laboratory composting method was that all of the species recovered from samples at Altoona and Leicester, except one, were also found in the laboratory runs. The exception, *B. subtilis*, was present in the Dano drums (at unknown temperatures) in low numbers. The *Streptomyces* sp. isolated from the windrow at Leicester may also have been a different species than that found during laboratory composting (runs A and D).

It should be noted, however, that this procedure did not simulate either the self-heating or the heat removal mechanisms of field-scale composting. For this reason, the laboratory apparatus would be unsuitable for certain types of composting research, such as an examination of the relationship between heat removal and drying by water vaporization (14, 15, 35). Also, it more closely approximated continuously thermophilic composting conditions, rather than batch-type composting such as occurs in windrows or static piles.

Species diversity generally refers to two components of community (or trophic-level) structure, i.e., the number of different species present (species richness) and the relative "evenness" of the number of individuals (or amount of biomass) in each species. The Shannon index used here combines both of these factors and has the additional advantage of being relatively independent of sample size (26).

Species diversity is usually not correlated with biological productivity in any direct way (26). This lack of correlation appears to be the case in the laboratory composting samples, in which the diversity index seems unrelated to the plate counts. However, species diversity is generally considered to be correlated with community stability. This correlation is one reason that species diversity is desirable in composting. Another reason is that to achieve the degradation of a maximum number of the many diverse organic compounds in refuse and sludge, a metabolically versatile population is required. In practice, this means a diverse community.

In communities which are limited by extreme physical conditions, such as high temperature, species diversity is usually low. Among the laboratory composting runs, diversity dropped off sharply at incubation temperatures of 60 and 65°C. The species diversity indexes for the three runs at 49 to 55°C (including the completely mixed reactor simulation) were virtually identical.

The ultimate example of physical factors limiting community diversity would be an environment in which no species could grow. Such a situation can more easily arise with lesser extremes of two inhibitive factors, such as temperature and pH, in combination. Such a combination may describe the case in the laboratory trial at 65°C without initial pH adjustment (run G). In this inactive run, a temperature of 65°C was rapidly imposed, denying the system a come-up stage during which it might have adjusted its own pH. Under field conditions, pH would necessarily increase as a result of bacterial activity before such activity could raise the tem-

perature to this level. For example, at Altoona a similar limitation may have been approached, possibly explaining the low plate counts, but this built-in self-regulating mechanism prevented a complete cessation of activity.

This study sought to explore some aspects of the microbiology of the composting ecosystem. The results leave little doubt that temperatures above 60°C have a marked detrimental effect on species diversity. Therefore, to the extent that a high species diversity is desired for population stability and metabolic versatility, the maximum desirable temperature for continuously thermophilic composting appears to be 60°C.

ACKNOWLEDGMENTS

I deeply thank Melvin S. Finstein for his extensive and invaluable collaboration and Ruth E. Gordon for her generous assistance in my efforts to identify the *Bacillus* isolates. I also thank E. Glynn Hughes of the Leicester composting plant, the staff of the county analyst's office in Leicestershire, England, the personnel at the plant in Altoona, Pa., and Sue Strom.

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