

Comparison of the Persistence of Bacterial Pathogens during Composting, Liquid-Storage and Pack Storage of Dairy and Swine Manures

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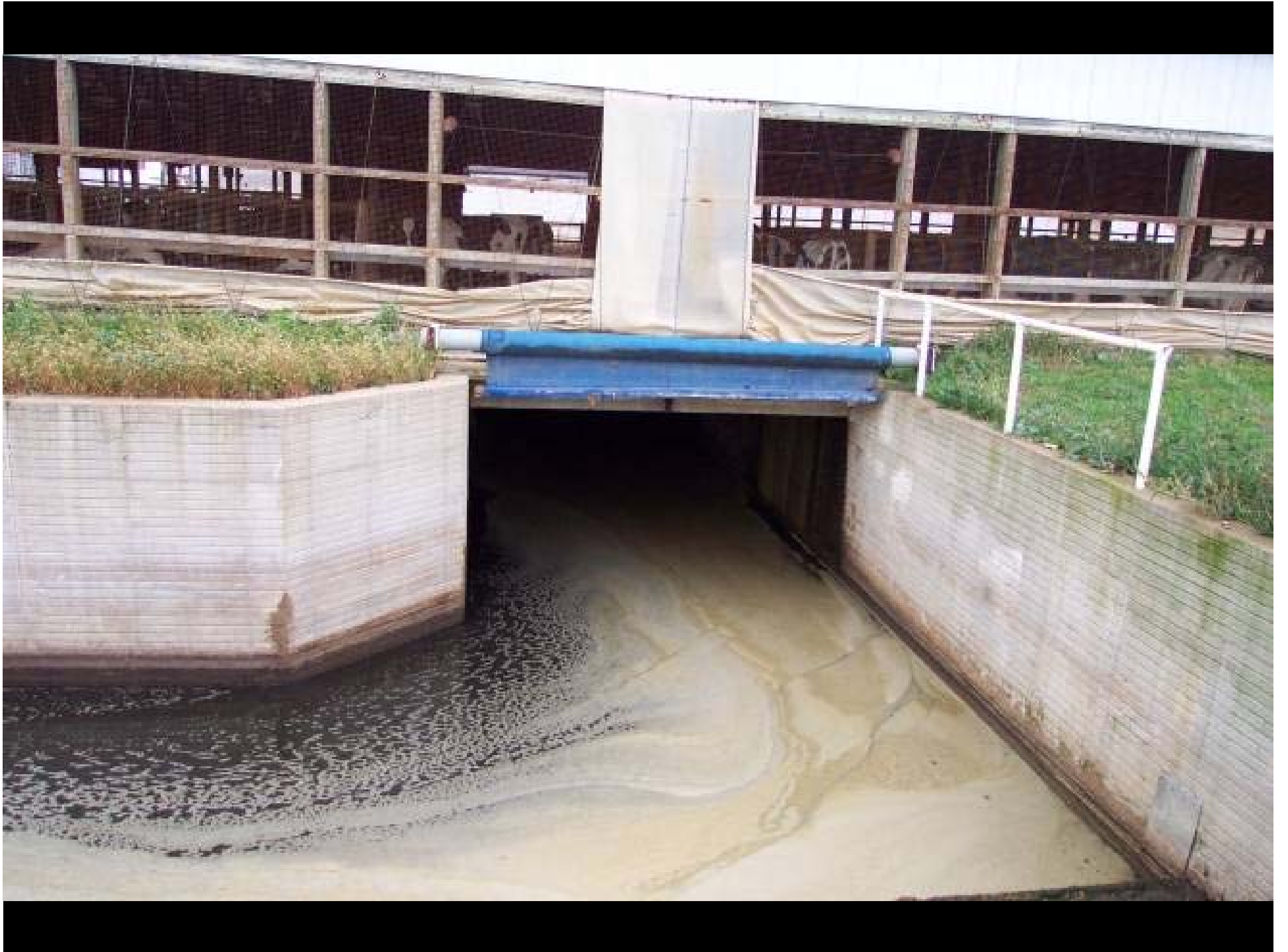
Introduction

- Animal manures often contain pathogens such as *Escherichia coli* O157:H7, *Listeria monocytogenes*, *Salmonella* spp., and *Mycobacterium avium* subsp. *paratuberculosis* which infect humans and/or animals.
- Farms use many different manure treatment and storage systems.
- Little is known about the relative persistence of these pathogens in different treatment systems.



Liquid Based Manure Management Systems







Lagoon Storage



Land application

Manure pack storage



MANURE TREATMENT VIA COMPOSTING

**Compost amendments
(sawdust, hay, straw, horse bedding, recycled compost, etc.)**



**Unseparated
Manure**



**Value
Added
Markets

(Residential,
Organic,
Potting
Media)**

**Free-stall barn
Manual
or Automatic
Scrape system,**

**Sand,
Straw or
Sawdust
bedding**

**Windrow
or
Agitated Bed
Composting**

COMPOSTING



Gradients of Temperature
Oxygen, Moisture

Swine Manure Management Deep Pit or High Rise Composting Systems



Priority Pathogenic Bacteria

USDA

- High Priority

- Enterohemorrhagic and related *E. coli* (DAIRY)
- *Salmonella* spp. (DAIRY AND SWINE)
- *Campylobacter*
- *Listeria monocytogenes* (DAIRY AND SWINE)



- Medium Priority

- *Yersinia enterocolitica*
- *Clostridium perfringens*
- other pathogenic *E. coli*



- Low Priority

- *Mycobacterium av. Paratuberculosis*
(Johne's disease, Crohn's disease, DAIRY)

Objective

Compare the persistence of *Salmonella spp.*, *Listeria monocytogenes*, *E.coli* and *Mycobacterium avium paratuberculosis* (Johne's disease), during the treatment of swine and dairy manure under conditions that simulate the most commonly used manure management methods



Methods

- Dairy Manure collected from a large farm previously shown to harbor *E.coli*, *Salmonella* and *Mycobacterium paratuberculosis*.
- Swine Manure collected from the OARDC swine facility
- Manure mixed with sawdust, straw or water and added to triplicate reactors.
- Dairy manure inoculated with 10^6 CFU/g *Mycobacterium avium* subsp. *paratuberculosis*.
- Swine manure inoculated with *Listeria monocytogenes* 10^6 CFU/g and *Salmonella typhimurium* 10^6 CFU/g.
- Samples tested for *Salmonella*, *Listeria*, *E.coli* and *Mycobacterium avium paratuberculosis* using standard culture and MPN methods.
- *Mycobacterium avium paratuberculosis* in dairy manure also assayed using an IS900 targeted PCR hybridization method.

Dairy Manure Treatment simulations

inoculated with *Mycobacterium avium subsp. paratuberculosis* (10^6 MPN/g)
Native Salmonella and E.coli already present

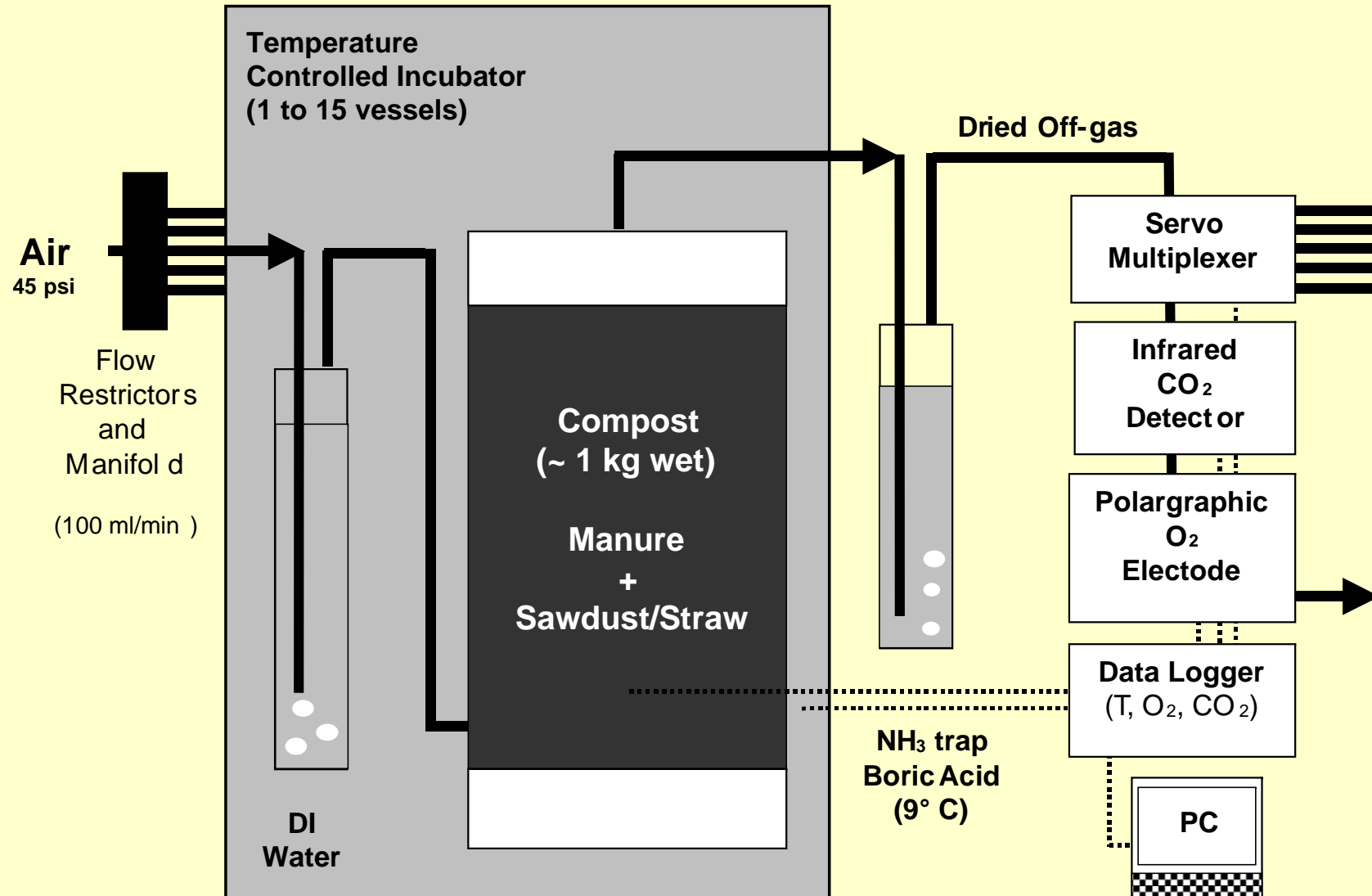
Amendment	Incubation Temperature	Initial moisture content (%)	Simulation
Sawdust	55°C	58	Composting
Sawdust	25°C	58	Manure pack
Straw	55°C	60	Composting
Straw	25°C	60	Manure pack
Water	20°-25°C	85	Liquid storage

Swine manure treatment simulations

inoculated with Salmonella and Listeria at 10^6 MPN/g

Amendment	Incubation Temperature	Initial moisture content (%)	Simulation
Sawdust	55°C	60	Composting
Sawdust	25°C	60	Manure pack
Water	25°C	85	Aerated liquid
Water	20°-25°C	85	Liquid storage

Compost Reactor System Used for Pathogen Fate Studies



Bioreactor System

CO₂, O₂ and temperature measurement, calibration and data acquisition.

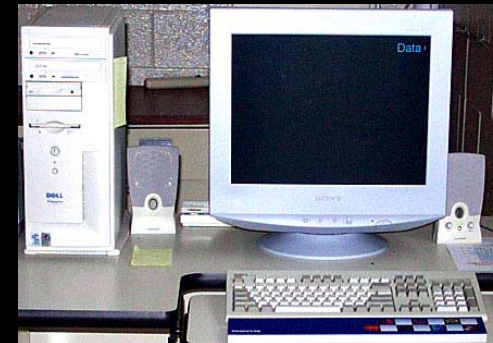
Bioreactor incubators



Flow restrictor system
(air 100 ± 3 ml/min)



Water condenser and
ammonia trap (9° C.)



Data collection computer



**Composting and
Pack Storage Simulation**



Aerated Liquid simulation



Liquid Storage simulation

PROPERTIES OF THE DAIRY AND SWINE MANURE

Sample	pH	Moisture Content (g/g wet)	Volatile Solids (g/g dry)	Total N% (% dwb)	Total C% (% dwb)	C:N ratio
Dairy Manure	8.0 \pm 0.1	72.8 \pm 1.4	25.9 \pm 1.4	1.4 \pm 0.2	14.6 \pm 1.1	10.5 \pm 0.4
Swine Manure	6.5 \pm 0.1	75.1 \pm 0.1	76.2 \pm 0.2	3.0 \pm 0.1	37.8 \pm 1.3	12.5 \pm 0.3

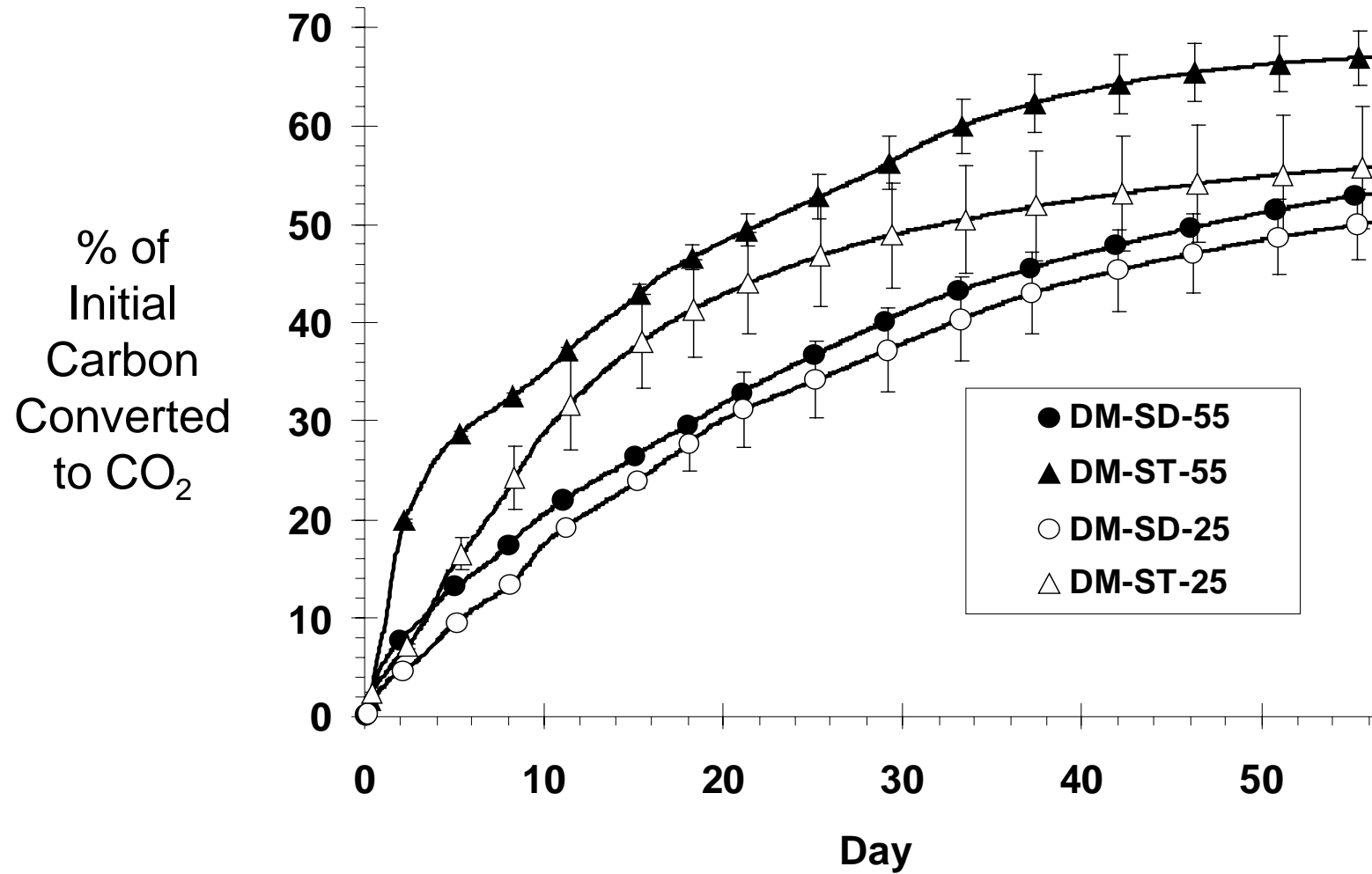


PROPERTIES OF THE INITIAL TREATMENTS

Sample	pH	Moisture Content (g/g wet)	Volatile Solids (g/g dry)	Total N% (% dwb)	Total C% (% dwb)	C:N ratio
DMSD	8.7 ±0.3	58.3 ±0.6	58.6 ±0.8	0.7 ±0.1	28.7 ±0.4	42.3 ±5.1
DMST	8.6 ±0.1	60.0 ±0.9	51.2 ±3.6	0.9 ±0.1	25.7 ±1.3	30.4 ±1.7
DML	8.6 ±0.2	85.1 ±0.2	31.8 ±0.2	1.5 ±0.1	16.9 ±0.5	11.7 ±0.4
SMSD	6.7 ±0.1	59.7 ±0.9	89.3 ± 0.7	1.4 ±0.2	44.3 ±3.3	32.3 ±3.2
SML	6.5 ±0.1	84.9 ±0.2	76.1 ±0.4	3.2 ±0.1	36.2 ±0.8	11.4 ±0.4



CO₂ EVOLUTION DURING DAIRY MANURE TREATMENTS



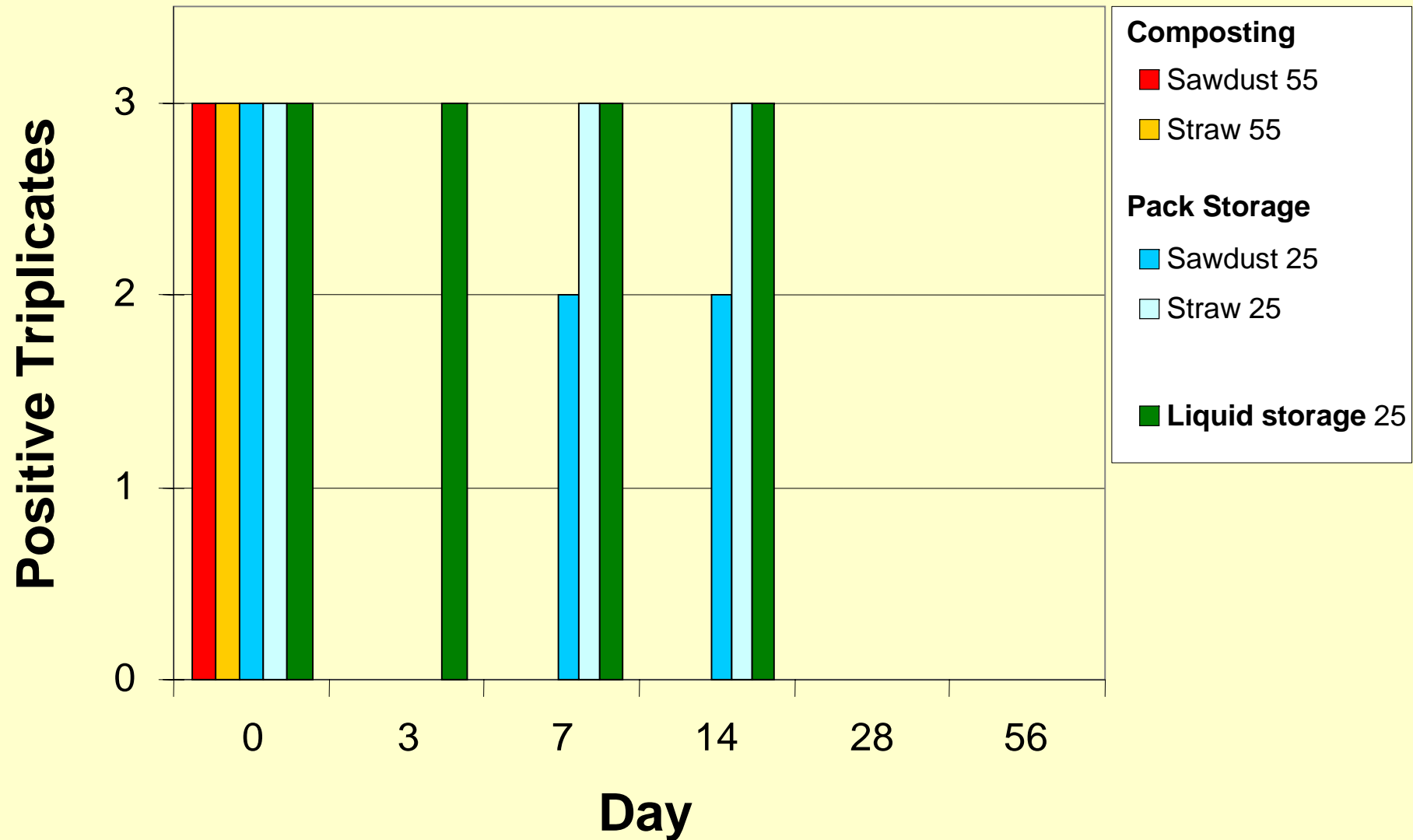
PROPERTIES OF THE FINAL TREATMENTS

Sample	pH	Moisture Content (g/g wet)	Volatile Solids (g/g dry)	Total N% (% dwb)	Total C% (% dwb)	C:N ratio
DM-SD-55	7.8±0.0	63.4 ±0.8	33.7 ±1.9	1.1 ±0.1	17.2 ±0.7	15.4 ±0.8
DM-ST-55	8.0±0.1	62.5 ±1.7	19.7 ±0.7	1.2 ±0.1	10.2 ±0.1	8.5 ±0.3
DM-SD-25	8.3±0.1	67.5 ±0.3	37.1 ±1.2	1.2 ±0.1	20.1 ±0.9	17.5 ±1.2
DM-ST-25	7.7±0.1	68.1 ±0.3	31.2 ±0.9	1.4 ±0.1	15.5 ±0.8	11.0 ±0.2
DM-L-RT	7.9±0.4	83.9 ±0.6	29.7 ±1.5	1.7 ±0.1	16.1 ±1.0	9.6 ±0.2
SM-SD-55	7.2 ±0.1	64.1 ±5.6	77.0 ±1.6	2.8 ±0.1	38.3 ±1.3	13.6 ±0.9
SM-SD-25	7.3 ±0.1	75.4 ±0.2	78.9 ±1.1	2.7 ±0.1	40.4 ±0.9	14.8 ±0.2
SM-AL-25	6.8 ±0.1	86.4 ±0.4	71.2 ±0.3	3.5 ±0.1	38.5 ±0.1	11.1 ±0.2
SM-L-RT	6.8 ±0.1	85.1 ±0.2	73.5 ±0.3	3.4 ±0.1	39.5 ±0.4	11.5 ±0.2



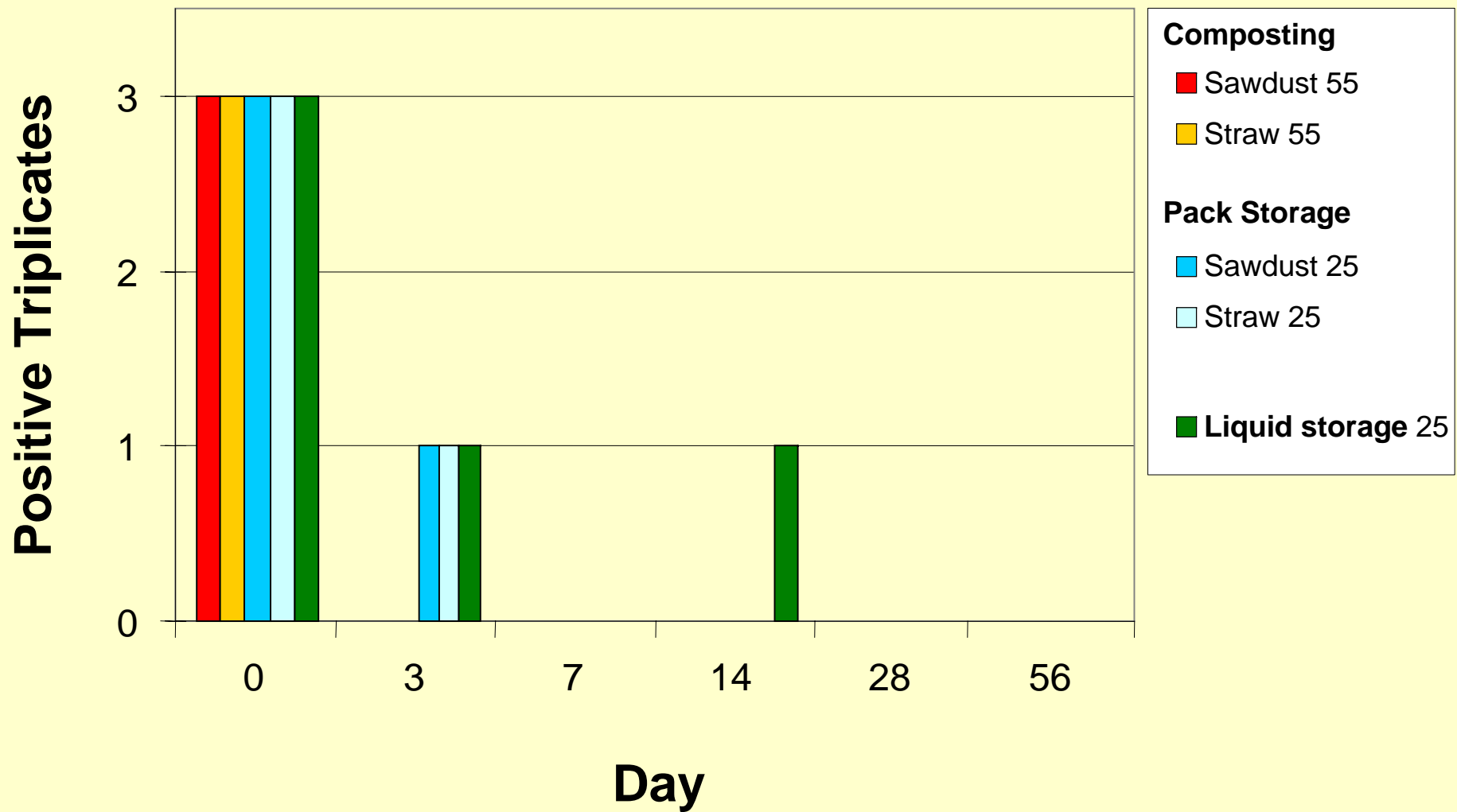
E.coli Persistence in Dairy Manure

detection limit ~11 MPN/gm



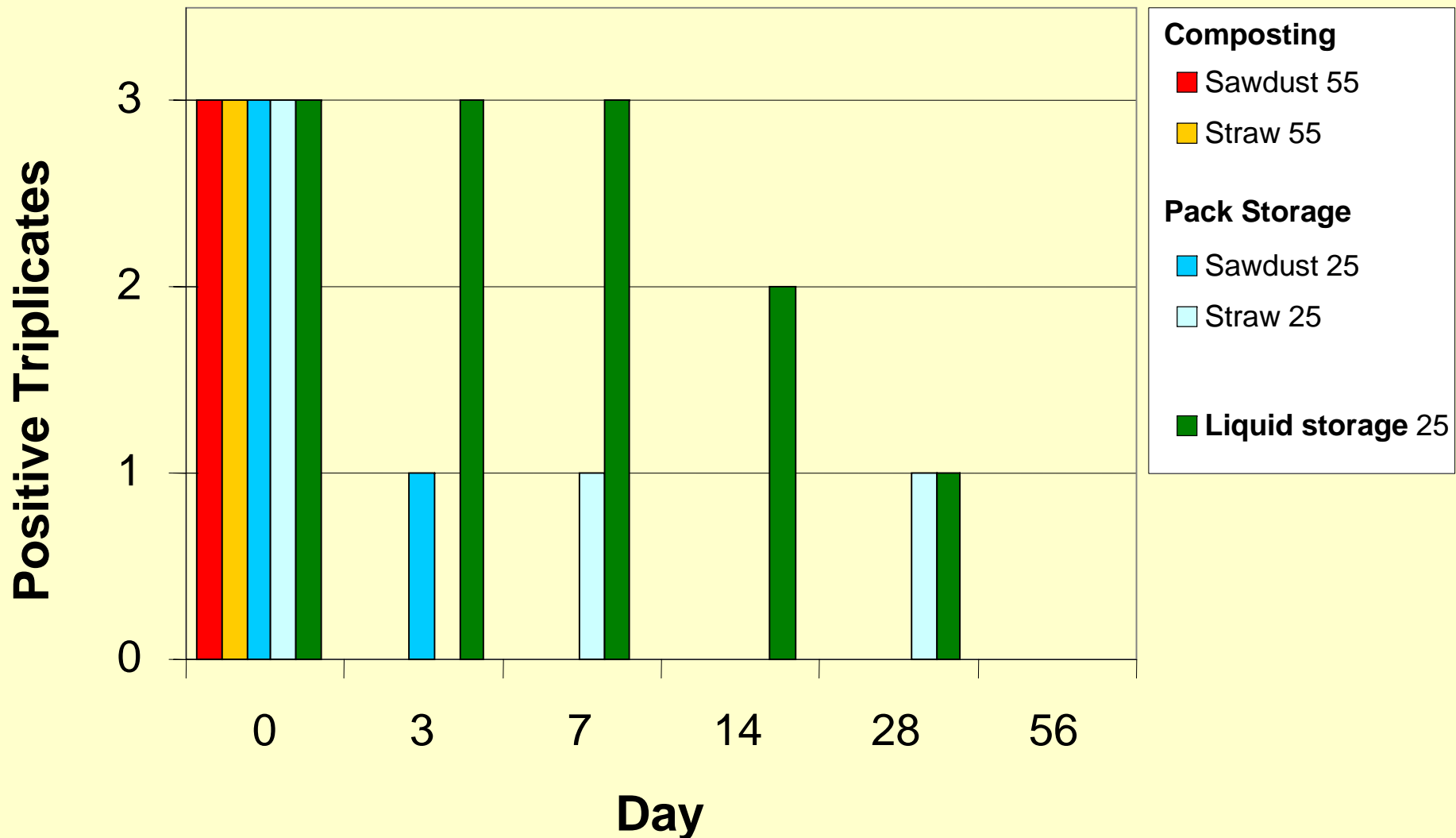
Listeria Persistence in Dairy Manure

detection limit ~11 MPN/gm

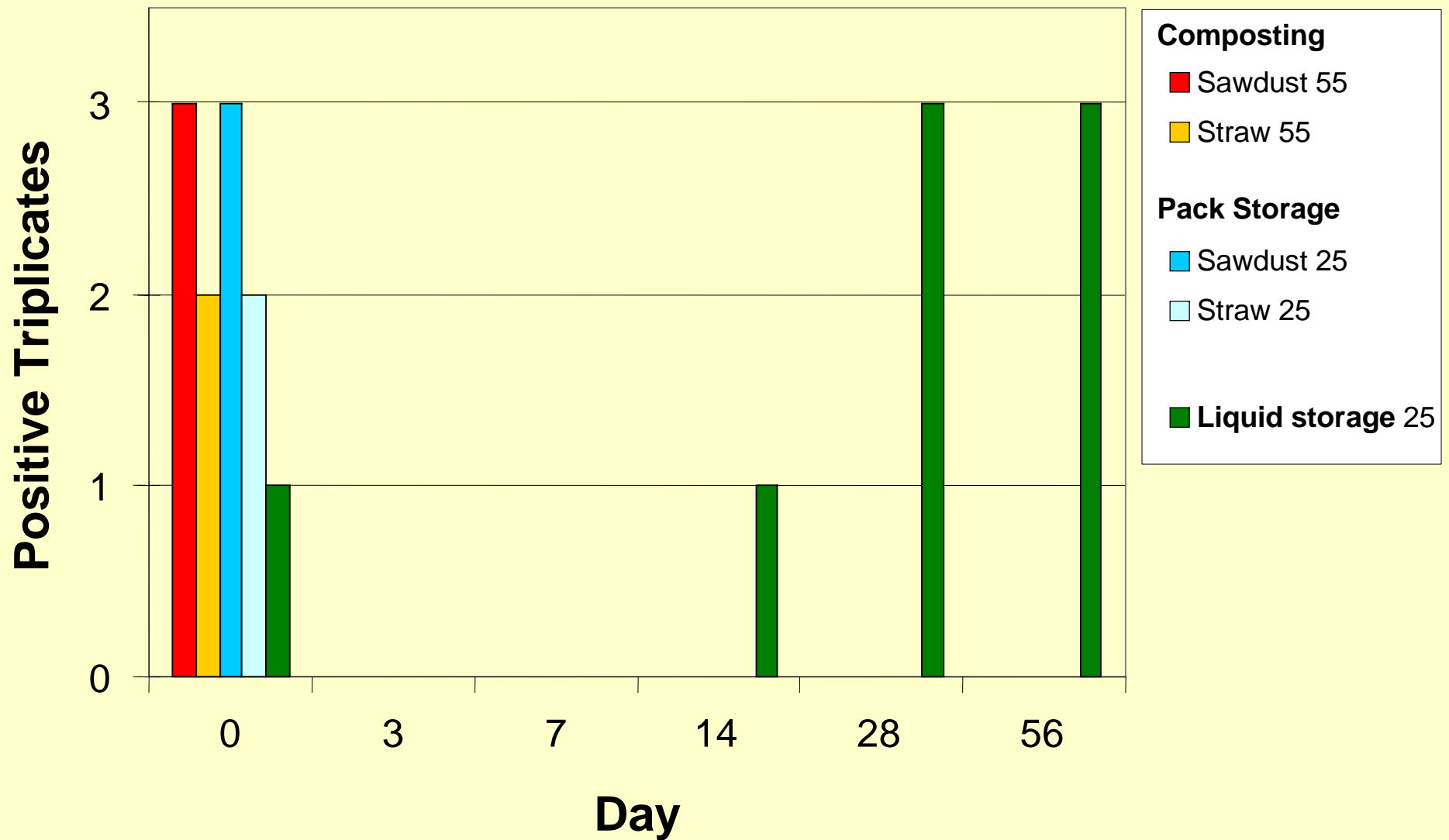


Salmonella Persistence in Dairy manure

detection limit ~11 MPN/gm

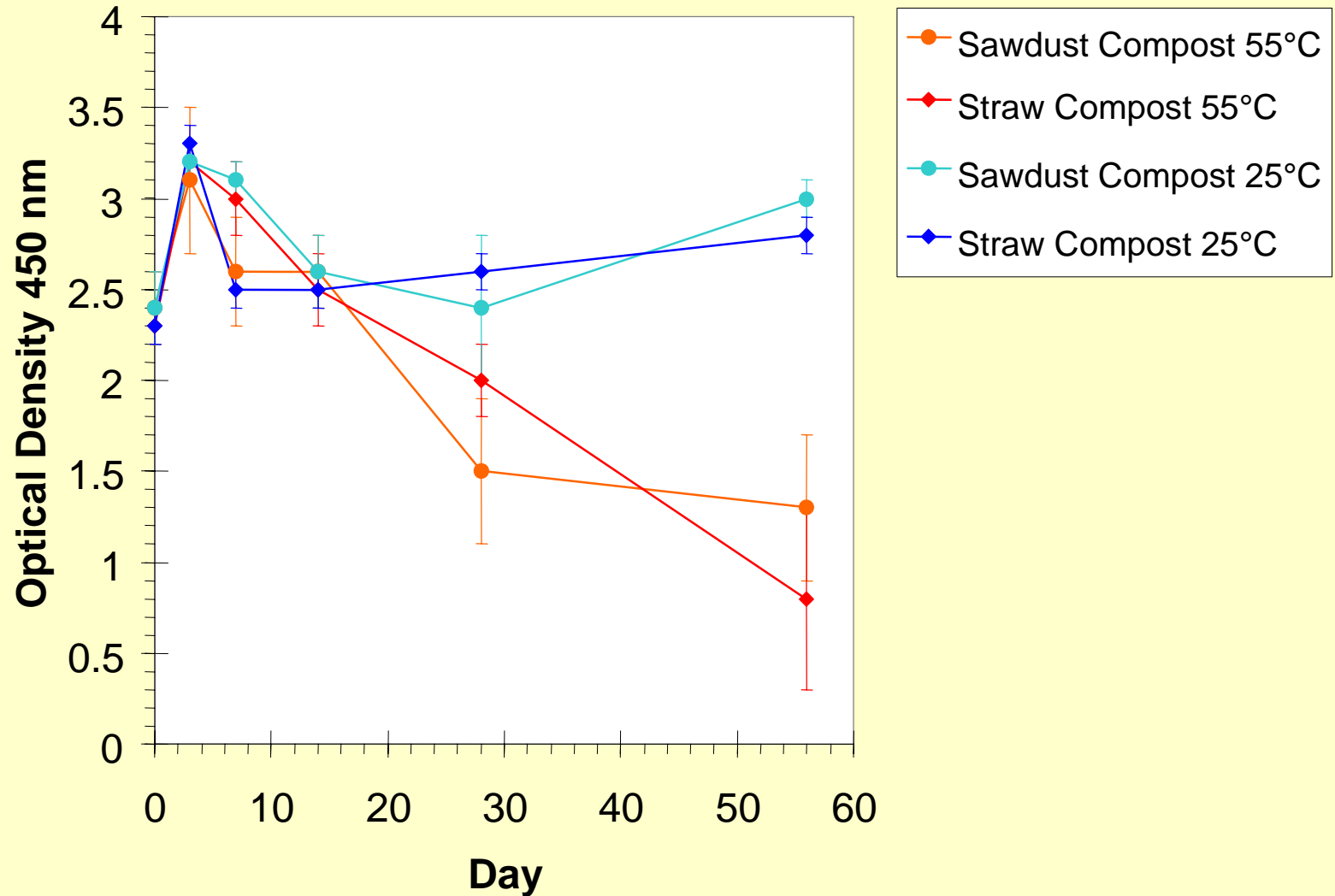


Mycobacterium avium paratuberculosis in Dairy Manure

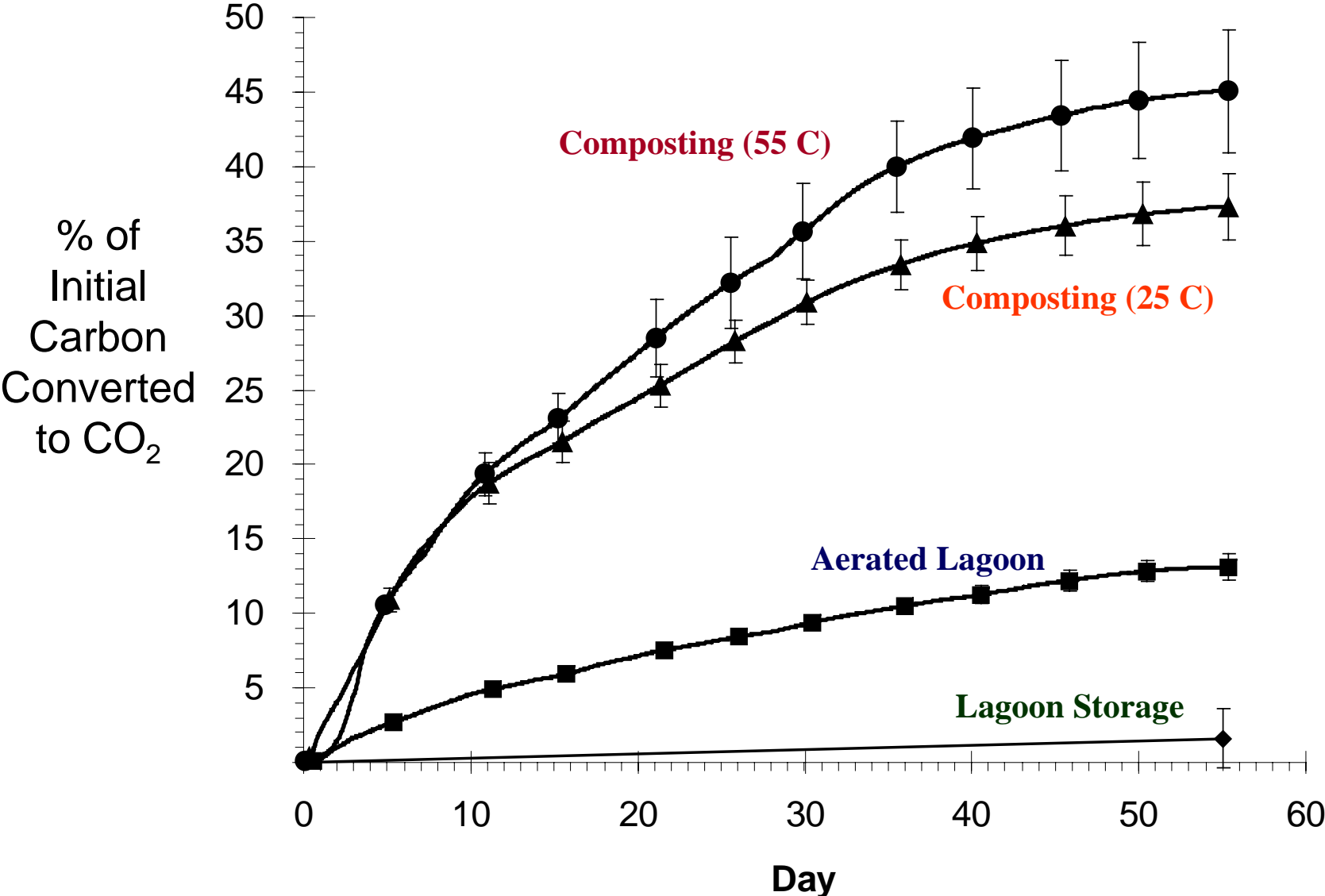


Mycobacterium avium paratuberculosis persistence during composting

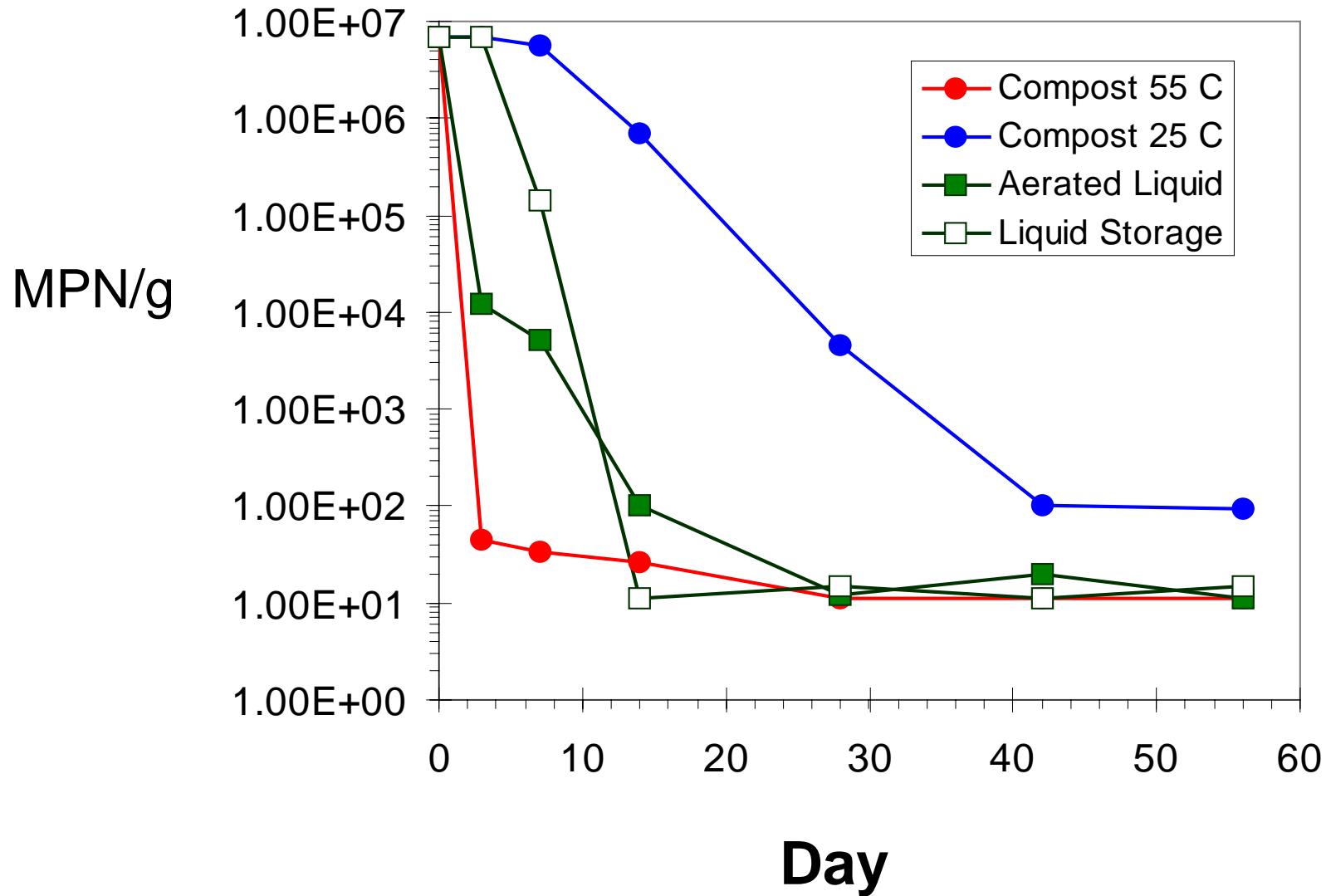
PCR product hybridization assay with MAP specific *IS 900* integration site



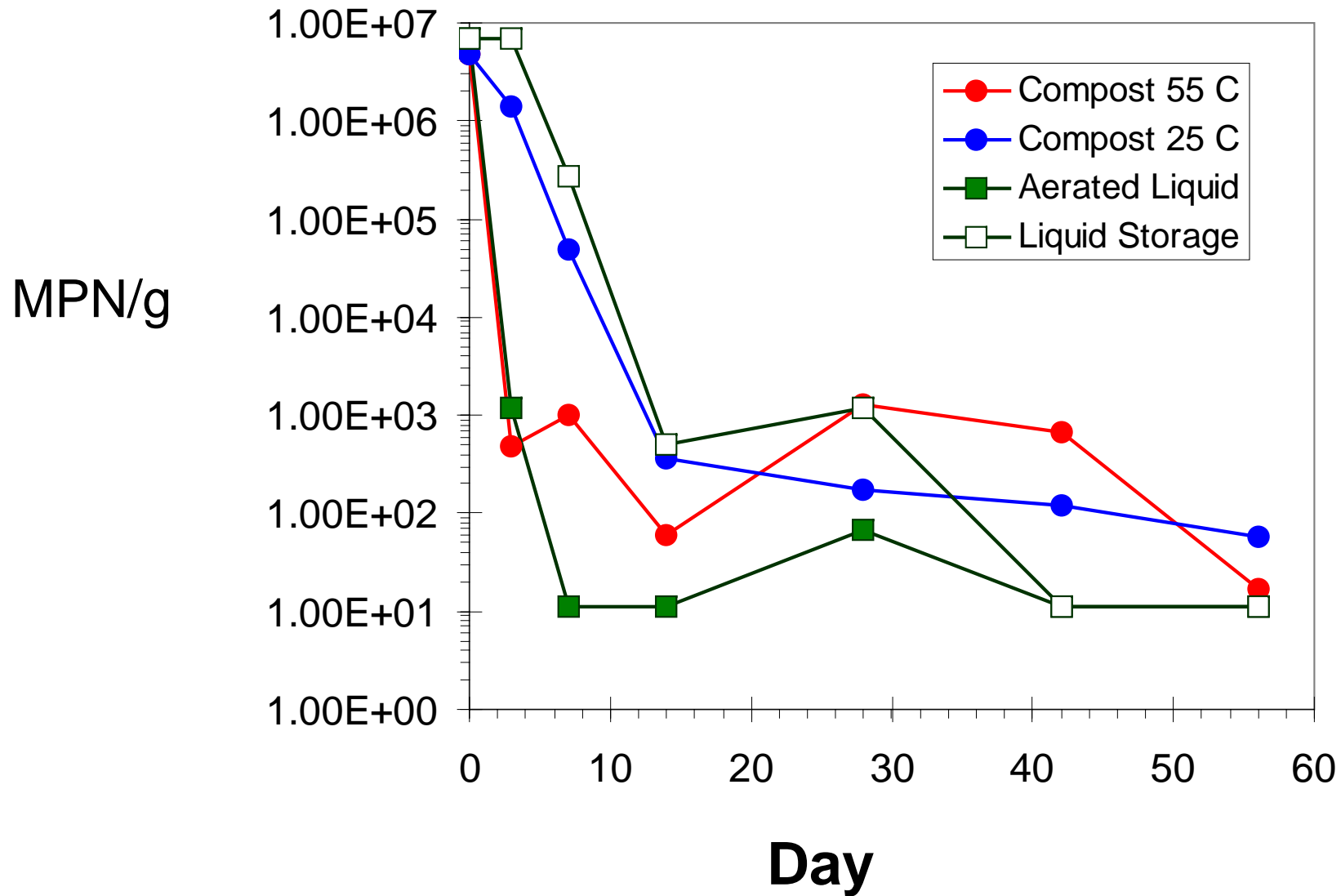
CO₂ EVOLUTION DURING SWINE MANURE TREATMENTS



Salmonella Persistence in Swine manure



Listeria Persistence in Swine manure



Conclusions

- **High temperature (55 C) composting of both dairy and swine manure was most effective in reducing priority pathogens.**
 - **After 3 days of dairy manure composting (55°C), naturally occurring *Escherichia coli*, *Salmonella*, and *Listeria* were not detectable.**
 - **In swine manure *Salmonella*, and *Listeria* were reduced more than 3 logs after 3 days of composting (55°C), or aerated liquid treatment.**
 - ***Salmonella* and *Listeria* persisted for from 14 to 56 days in both swine and dairy manure during liquid storage.**
 - ***E.coli* and *M. avium paratuberculosis* persisted in dairy manure for 14 to 56 days during simulated lagoon treatment.**
- **Under all treatment conditions *Salmonella*, *E.coli* and *Listeria monocytogenes* reduced to low numbers after 56 days.**
- **Thermophilic composting is recommended for the treatment of manure destined for pathogen sensitive uses such as fruit or vegetable production, gardening or application to well drained fields.**

Acknowledgements

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- *Jerome Rigot, Tom Wilkinson, Dave Elwell, Mary Wicks*



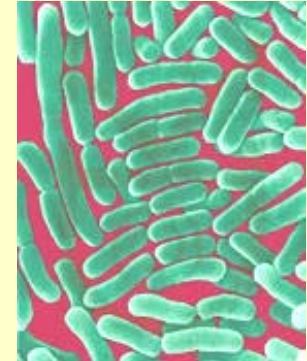
Questions?

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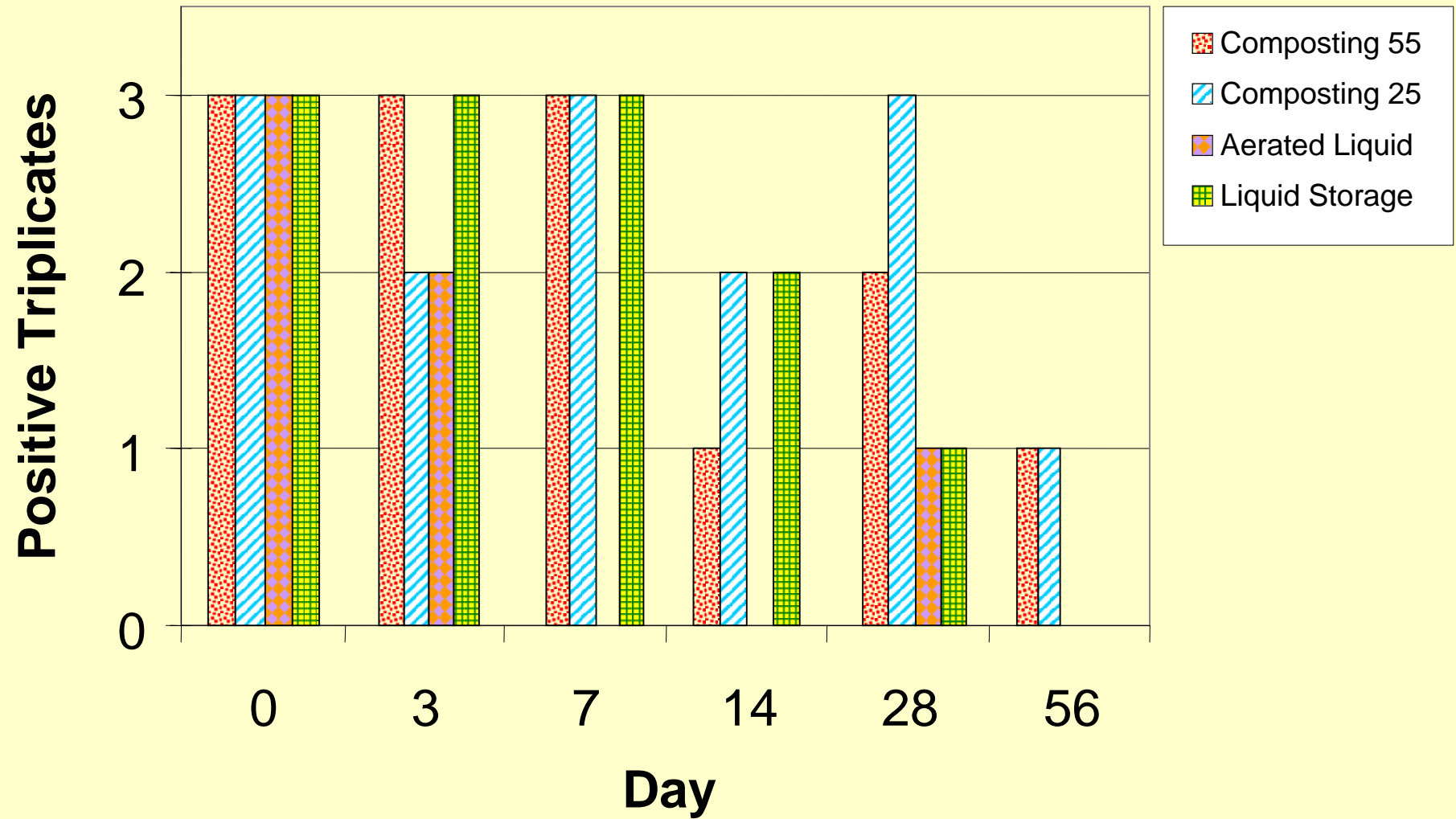


Factors Affecting Pathogen Survival during Manure Treatment and Storage

- Manure type
- Temperature
- Moisture content
- Aeration
- Microbial competition
- Amendments
- Treatment system
- Animal feed
- Antibiotic use
- Many others



Listeria Persistence in Swine Manure



Salmonella Persistence in Swine manure

