Pathogen Reduction and Environmental Impacts Associated with Composting of Bovine Mortalities

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***Abstract*.** Composting of animal mortalities is prohibited in California. Rendering is the only legal option for carcass disposal. During emergencies, alternate disposal methods are necessary. Regulatory agencies are seeking direction as they consider new policy for mortality management. The main objective of our research was to investigate impacts of time and temperature during carcass composting on pathogen reduction. In addition, environmental impacts were assessed from compost piles with or without carcasses. Static piles were constructed on a lined base. Variables

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included amendment (screened dairy manure solids or dry lot manure scrapings); season (summer or winter); and carcass (piles constructed with or without four adult bovines). Data loggers and pathogens (fecal coliforms, *Bacillus cereus* spores, and *Salmonella enterica* serotype *Senftenberg*) were placed in and around carcasses before covering with amendment. Pathogens were retrieved at 0, 2, 6, 10, and 16 weeks. Bacteria were enumerated by the Most Probable Number method using appropriate selective media and approved protocols for each targeted pathogen. Bacterial loss or log killing over time was quantified. Liquid run off was quantified and analyzed for salt and nutrients. Air emissions were studied using US EPA surface emission flux chamber technology. Total hydrocarbons and ammonia were quantified. Preliminary results indicate that heating was rapid and prolonged, there was a 6 $\log_{10}$ reduction in microbial pathogens by week 6, and decomposition was extensive by week 16. Decision makers will consider results from this and other related research in development of policies for emergency animal disposal.

Keywords. Mortality management, carcass disposal, carcass composting

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Introduction

California – for many the name conjures up images of beautiful beaches, Disneyland, and movie stars. It is the land of the Golden Gate Bridge, Yosemite, and sprawling urban landscapes like the San Francisco Bay Area and Los Angeles. Less commonly known is the fact that California leads all other states in the US for agricultural production. The value of agriculture in California was $36.6 billion USD in 2007 (CDFA, 2009). Animal agriculture contributed $10.7 billion, highlighting the importance of animal agriculture, especially dairy production in this uniquely diverse state.

The dairy industry in California produces nearly 25% of the milk produced in the US. In 2008, there were 1.9 million dairy cows on 1852 dairy farms in the state (CDFA Dairy Marketing Branch, 2009). The average herd size was 1017 cows/herd, ranging from about 100 cows to 9000 cows per herd. The most densely populated dairy area is the southern end of the San Joaquin Valley, which includes Tulare County, home to 493,383 cows and 329 dairies. Other species including poultry, beef, small ruminants and swine add significantly to the total animal agriculture population in California. Table 1 shows numbers of animals by species/class for 2008.

Table 1. Farm Animal Population by Species/Class, 2008

Animal Species/Class	Total Number of Animals
Dairy, cows	1.9 million (CDFA, 2009)
Poultry, layers	19.6 million (USDA, 2009)
Cattle and calves, all	5.5 million (USDA, 2009)
Sheep, all	620 thousand (USDA, 2009)
Goats, all	129 thousand (USDA, 2009)
Hogs, all	80 thousand (USDA, 2009)

Background

Routine carcass disposal for dairy and livestock is by commercial rendering. There are currently four rendering companies servicing livestock and dairy operations. With yearly average “routine” mortality rates for mature cows ranging from 2 to 8% (Stull et. al., 2008), and 5 to 10% for calves, there is a colossal amount of tissue to process under normal conditions. Capacity is currently adequate to render this routine volume of mortalities, although a disease outbreak or extreme weather event could perilously overwhelm existing rendering capacity. Furthermore, a recent US Food and Drug Administration enhanced ruminant feed ban (FDA, 2008) will likely alter rendering profitability and ability to serve dairy and livestock mortality disposal needs.

In July 2006, an extended heat wave created a nightmarish carcass disposal emergency throughout the San Joaquin Valley. Daytime high temperatures exceeded 100˚ F for nearly a month. The worst period was 14 consecutive days with high temperatures ranging from 103˚ F to 113˚ F, and night time low temperatures above 90˚ F. All dairies experienced heat related animal losses despite the common use of shades, fans, and misters for cow cooling. The

extreme weather coincided with equipment failures at one of only two large rendering plants serving the Tulare area. Tens of thousands of carcasses throughout the area became too badly decomposed for rendering. Alternative means of disposal (burial, landfill, composting) were employed in the wake of emergency declarations. These options are normally restricted by local, state or regional agencies because of concerns for public health, or environmental impacts to air and water resources. Timely response in some cases was hampered by a morass of ambiguous regulatory jurisdictions.

California's Emergency Animal Disposal Working Group

This event clearly underscored the need for strategic planning and coordination of emergency response at all levels of government and among industry stake holders. A statewide emergency animal disposal working group (EADW) was convened to develop solutions for emergency mortality management and to explore alternative carcass disposal methods (Payne et. al., 2009). Regulations constrain most alternate disposal options in California. Restrictive waste discharge requirements imposed by the Regional Water Control Board on most dairies prohibit on-farm burial (CRWQCB, 2007). Open burning of deceased animals is not allowed by the San Joaquin Valley Air Pollution Control District (SJVAPCD, 2007). Although some states allow carcass composting, the California Integrated Waste Management Board prohibits "composting of mammalian flesh" (CIWMB, 2008), because of concerns about persistence of pathogens. Among alternate disposal options, composting was selected as the most promising for research by the EADW. The microbial risks associated with composting of animals were recently reviewed (Berge et. al., 2009), and it appears that this method of disposal can be safe and efficient when done correctly. Well developed guidelines and instructions for carcass composting are available on numerous institutional websites. Further studies to determine the effects of composting on microbial pathogens including spore forming bacteria would provide valuable information for agencies that are developing mortality management policies.

Carcass Compost Research

In 2008, the California Code of Regulations section that prohibits composting animal carcasses was amended to allow: 1) research designed to obtain data on pathogen reduction and 2) temporary carcass composting during declared emergencies. Funds were obtained and a research protocol was developed. The primary objective was to quantify microbial pathogen reduction, including the effect on spore-forming bacteria, during composting of adult dairy cows. Secondary objectives were to assess potential environmental impacts of composting bovine mortalities in manure on air and water resources. Another important consideration was to simulate conditions that would be common on local dairies in an emergency situation. The composting amendment chosen was dairy manure solids - an abundant, readily available resource on all dairies. Our desire was not to optimize a composting protocol, but to study the process under "real world" conditions.

Materials and methods

Pile construction: Initially, a single "pilot" compost pile containing four Holstein cows (approx. 1400 lbs. each) was constructed in an empty manure storage pit at the University of California Veterinary Medicine Teaching and Research Center in Tulare, California. The pile footprint measured 16' by 24' and it was lined with plastic (6 mil) secured to wooden posts outlining the perimeter. The plastic was overlain by three lengths of perforated 2" PVC pipe sloped to a vessel to facilitate collection of leachate (liquid run-off). The perforated pipe was covered with a single layer of landscape cloth before adding base material to prevent clogging of pipe with manure solids. Base material consisted of 0.5' of wood shavings overlain by 2.5'of screened

dairy manure solids (SDMS – 70% moisture). SDMS are fibrous solids remaining after manure is flushed from cow lanes and pumped over an inclined manure separator screen.

Figure 1. Pile footprint showing liner and leachate collection set-up.

Freshly dead (<12 hours) cows were placed back to back as recommended by Glanville, et. al., 2006. Prior to covering, three data loggers set to record temperature every 30 min. for 16 weeks were placed near the head, abdomen and hindquarters of each cow. Plastic balls containing pathogens (see description below) were also placed in these general areas. Finally, 3' of SDMS cover material was added. All cows and manure came from local dairies. No turning, addition of water or further manipulation was made to the pile after construction.

Figure 2. Compost pile showing placement of four cows. Metal stakes were used for measuring depth of base and cover material as it was added.

Pathogens: Sentinel pathogens were introduced to the carcass area of the pile for retrieval at 0, 2, 6, 10 and 16 weeks of composting. Selected pathogens included organisms (or surrogates for these) that could potentially be encountered in animal mortalities and manure. Fecal coliforms and E. coli were derived from the rectum of cows received for composting. *Salmonella enterica* serotype Senftenberg (ATCC 43845) and *Bacillus cereus* (ATCC 4342) were purchased. *B. cereus*, a spore forming, gram positive bacterium was chosen as a surrogate for anthrax, because it exhibits similar characteristics. Sterile amendment was spiked with 1 X 10^7 cfu/gm of each organism and sealed in porous nylon bags. The nylon bags were secured in plastic, toy "wiffle" balls which were secured with nylon rope to enable retrieving them at appointed time intervals without disrupting the compost pile. The balls containing the

sentinel pathogens were placed in the head region, inside the abdominal cavity and near the hindquarters of each carcass prior to covering. Most Probable Number (MPN) of bacteria cultured from the retrieved pathogen balls was used to assess log killing over time.

Characterization of chemical /physical changes, manure amendment, and leachate: A manual probe was used to check oxygen levels at two and four feet during the first month of composting. Thermocouples buried in the pile recorded temperature every 30 minutes. Replicated samples of all manure amendments and leachate produced were analyzed using US Composting Council (TMECC) and EPA methods for moisture, pH, electrical conductivity, nutrients, minerals, biological oxygen demand, carbon to nitrogen ratio, carbon to phosphorus ratio, bulk density and water holding capacity. Manure amendments and leachate were also cultured for microbial pathogens.

Charcterizing extent of decomposition: Visual observations of carefully excavated remains were made at approximately 120 days. Selected samples were carried to the CDFA California Animal Health and Food Safety pathology lab on-site for closer inspections of tissue.

Subsequent piles: Armed with experience from the initial pile and supplementary funding, six additional piles were constructed in the same manner, using different manure sources, and a modified leachate collection system. Plastic liner material used for subsequent piles was a double layer of left over plastic (5.5 mil) silage cover material. For leachate collection, a 55 gallon plastic barrel with lid replaced the 5 gallon bucket used for the initial pile. Tubing was installed to aid in pumping leachate from the barrel if it became full. A protocol for evaluating spore forms of *B. cereus* in compost was developed and tested prior to initiation of the subsequent piles. On September 30, 2008, two piles were created with SDMS (50% moisture) and on October 1, two piles were built with dry lot scrapings (DLS – 25% moisture). DLS are manure solids scraped from open corrals and stacked prior to crop land application. For each manure type, one pile was built with cows, and one pile without cows. On January 13, 2009, the last two piles were created with SDMS (60% moisture), one containing cows and one without. These piles are being used to compare microbial pathogen reduction, including spore forms, as a function of time and temperature by season, and by manure source. We are also comparing leachate, and air emissions from piles with or without cows in each manure type.

Air emissions monitoring: In order to provide comparative data between piles containing cows and those without, surface isolation flux chambers were used to assess the emissions of reactive organic gases (ROG) and ammonia. EPA flux chamber technology protocols were used (USEPA, 1986), including methods of California's South Coast Air Quality Management District (SCQAMD 25.3 for ROG; and SCQAMD 207.1 for ammonia). Testing was from multiple locations on five separate days (day 2, 7, 14, 38 and 66) of the120 day compost cycle. Laboratory analyses were performed by Almega Environmental Laboratory in Huntington Beach, CA.

Figure 3. Air emissions monitoring and close up of flux chamber.

In total, seven compost piles have been constructed; six are currently still under investigation. Table 2 provides an overview of the compost piles included in this project.

Table 2. Description of Bovine Mortality Compost Piles

Date	# of Carcasses	Amendment	% Moisture	Analyses
4/2/08	4	SDMS	70	pathogens, leachate
9/30/08	4	SDMS	50	pathogens*, season, leachate, air emissions
9/30/08	0	SDMS	50	season, leachate, air emissions
10/1/08	4	DLS	25	pathogens*, season, leachate, air emissions
10/1/08	0	DLS	25	season, leachate, air emissions
1/13/09	4	SDMS	60	pathogens*, season, leachate,
1/13/09	0	SDMS	60	season, leachate,

* including spore forms

Preliminary results and discussion

Initial pile: The SDMS used for our first compost pile were too wet. Manure solids derived from a separator screen are very wet initially (>80%). After being stacked and windrowed, the manure heats up and the moisture declines. The local dairy where we obtained our manure was actively engaged in composting separated manure solids for bedding. We selected material that had been windrowed about a week. It was quite hot (130 – 150˚ F), and we hoped that it would be in the 60 to 65% moisture range. What we received at the research site was 70% moisture. Knowing that this could be too wet, we quickly improvised by adding a bottom layer (about 6 inches) of dry wood shavings, bedding material that had been left over from a previous calf research trial at the site. Despite the high moisture of the SDMS used, heating was rapid and prolonged. Pile temperatures exceeded 131˚ F for 100 days; and 140˚ F for 45 days. Oxygen levels in the pile at the 2 foot depth varied between 6 to 9%. At the four foot depth, oxygen levels varied between 1 to 3%, suggesting that the pile was not sufficiently aerobic throughout. *Salmonella enterica subtype Senftenburg*, fecal coliforms, E. coli and *Bacillus cereus* (vegetative forms) evaluated in the packets recovered from the pile and in the leachate were not detected after 6 to 10 weeks. This represents 6 $\log_{10}$ reduction based on the MPN analyses (spore forms of *B. cereus* were not evaluated in the initial pile). The total volume of leachate collected over 120 days was 300 gallons. Most leachate was produced during the first two weeks. Because there was no control pile, it is not possible to establish the contribution of fluid from carcasses to the total leachate volume. We believe the high moisture amendment was the biggest factor for leachate production. Decomposition of carcasses by 120 days was extensive – we estimated that 95% of the soft tissue was decomposed. The large bones remaining could easily fit into a 55 gallon barrel.

Subsequent piles: Analyses and data summarization from these six piles are on-going. Heating in the January pile was slower than in the other piles. We were able to obtain drier SDMS for these piles. Dry lot scrapings were included in our composting study because some dairies do not use manure solids separators. In an emergency, the source of manure for carcass composting on these dairies would be scraped from their corrals, so we wanted to learn more about composting in those conditions. We were hoping to find DLS containing about 30%

moisture. After a long, hot summer, the only DLS available to us was quite dry (25% moisture). No leachate was produced by the DLS piles, and the volume of leachate for the SDMS piles, (50 and 60% moisture) was much less than for the initial pile containing SDMS with 70% moisture. Despite conditions that might be considered too dry, carcass decomposition in DLS was extensive after only 120 days.

Conclusion

Field tests of composting bovine mortalities in dairy manure indicate that this method may be a practical and effective means of carcass disposal during emergencies. All materials used are readily available on dairies. Rapid, prolonged heating contributed to microbial pathogen destruction and extensive tissue degradation. Suggested guidelines for carcass compost are to use amendments that range between 30 to 60% moisture. Our experience using manure amendments ranging from 25% to 70% moisture suggests that pathogen reduction and carcass decomposition can occur even when conditions are not ideal. Our final report will provide complete data on microbial pathogen reduction including spore-forming bacteria, as well as air emissions and leachate analyses. These data will be helpful for policy makers as they weigh the risk vs. the benefit of composting bovine mortalities.

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Appendix

Abbreviations

ATCC – American Type Culture Collection

CDFA – California Department of Food and Agriculture

CIWMB – California Integrated Waste Management Board

CRWQCB – California Regional Water Quality Control Board

DLS – Dry lot scrapings (manure scraped from open corrals)

EADW – Emergency Animal Disposal Working Group

MPN – Most probable number

ROG – Reactive organic gases

SDMS – Screened dairy manure solids (manure from an inclined solids separator screen)

SCAQMD – South Coast Air Quality Monitoring District

SJVAPCD – San Joaquin Valley Air Pollution Control District

TMECC - Test Method for the Examination of Compost and Composting

UC VMTRC – University of California Veterinary Medicine Teaching and Research Center

US EPA – United States Environmental Protection Agency

US FDA – United States Food and Drug Administration

USD – US Dollars ($)

USDA – United States Department of Agriculture