

# Foam-Based Mass Emergency Depopulation of Floor-Reared Meat-Type Poultry Operations

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**ABSTRACT** Current control strategies for avian influenza and other highly contagious poultry diseases often include quarantine, depopulation, and disposal of infected birds. For biosecurity reasons, on-farm depopulation and disposal methods are preferred. The options for mass depopulation are limited, as reported by the “2000 Report of the AVMA Panel on Euthanasia.” Current depopulation techniques may have excessive labor requirements, are not appropriate for all house types, and may not be suitable for large-scale emergency implementation. A procedure has been developed that uses foam to rapidly form a blanket over the birds. The procedure requires relatively few people, can be performed in a variety of house types, and is compatible with in-house composting. Results from 2 experiments using foam for depopulation

are presented in this paper. These studies have shown that foams are comparable to the CO<sub>2</sub> polyethylene tent procedure in time to death in small groups and that the foam is faster as group size increases. Adding CO<sub>2</sub> to the foam does not enhance its efficacy. Based on corticosterone levels, the study also showed that the foams are no more stressful than the CO<sub>2</sub> depopulation method. Necropsy and histological examination of birds indicated that blood was present to some degree in the trachea, syrinx, and bronchial tree in broilers subjected to foam with CO<sub>2</sub>, foam without CO<sub>2</sub>, and CO<sub>2</sub> polyethylene tent methods of depopulation. Foam caused a rapid onset of airway occlusion. In both foam- and CO<sub>2</sub>-euthanized broilers, lesions are consistent with anoxia or hypoxia. This suggests that foam acts by physically induced hypoxia, whereas CO<sub>2</sub> causes chemically induced hypoxia.

**Key words:** broiler chicken, mass euthanasia, depopulation, avian influenza, foam

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## INTRODUCTION

The possibility of a highly pathogenic avian disease outbreak is an ongoing concern for the poultry industry. Avian influenza (AI) is a significant threat to the poultry industry. Specifically, the Asian bird flu or H5N1 highly pathogenic AI has been confirmed in Asia, Europe, and Africa and continues to spread to other countries at an alarming rate. Within the United States, one of the worst AI events occurred in Pennsylvania in 1983, in which an outbreak of highly pathogenic AI H5N2 resulted in the destruction of 17 million birds at a cost of \$61 million (Panigrahy et al., 2002; Goldblatt et al., 2004; Lu et al., 2004). More recently, a low pathogenic AI H7N2 outbreak in Virginia in 2002 resulted in the destruction of 4.7 million birds at a cost of \$160 million (Capua and Alexander, 2004). The detection of a H7N2 low pathogenic AI strain in the Delmarva region in 2004 resulted in the depopulation of 328,000 chickens on 3 farms in Maryland and

Delaware (Capua and Alexander, 2004; Luladey et al., 2004; Smeltzer, 2004).

Control of AI in meat birds (i.e., broilers) often consists of surveillance, rapid detection, confinement, and depopulation. Quarantine of the area is an immediate step in control of an infectious poultry disease and can significantly reduce the spread of the disease. Birds that are infected or suspected of infection are depopulated using the most expedient and humane methods available. Vaccination has been used in control of several outbreaks; however, the usefulness of conventional vaccination as an eradication tool is unclear (Capua and Alexander, 2004). Evidence indicates that the virus can continue to replicate in clinically healthy vaccinated birds, which reduces the effectiveness of vaccination programs. For this reason, depopulation remains one of the primary control measures for avian influenza.

Emergency depopulation can be required under other conditions as well. Exotic Newcastle disease is an example of another avian disease in which depopulation of infected birds and birds suspected of infection is a primary control measure (Kingston et al., 2005). Natural disasters such as hurricanes, wind, snow, and fire can damage poultry housing, leading to a need to quickly and effi-

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ciently depopulate poultry houses that may be structurally unsafe for human entry.

Depopulation of infected birds must balance human health, animal health, and other risk factors. The American Veterinary Medical Association (AVMA) describes euthanasia as "rapid loss of consciousness followed by cardiac or respiratory arrest and the ultimate loss of brain function" (AVMA, 2001, p. 672). The AVMA guidelines indicate that the euthanasia techniques should minimize distress and anxiety experienced by the animal before loss of consciousness. The guidelines also indicate the absence of pain and distress cannot always be achieved. For poultry, acceptable methods include inhalant anesthetics, barbiturates, CO<sub>2</sub>, and CO. Conditionally acceptable euthanasia agents for poultry include N<sub>2</sub>, Ar, cervical dislocation, and decapitation. The methods cause death by 1) direct or indirect hypoxia, 2) direct depression of neurons, or 3) physical disruption of brain activity. However, during emergencies such as a disease outbreak, there are fewer options. The "2000 Report of the AVMA Panel on Euthanasia" (AVMA, 2001, p. 688) devotes only 1 paragraph on mass euthanasia, which states "Under unusual conditions, such as disease eradication and natural disasters, euthanasia options may be limited. In these situations, the most appropriate technique that minimizes human and animal health concerns must be used."

Gassing is one of the accepted methods for euthanizing poultry. A number of researchers have studied the effects of various gas mixtures for euthanizing poultry (Poole and Fletcher, 1998; Lambooij et al., 1999; Gerritzen et al., 2000). The findings from this research can also be applied to the development of contingency depopulation procedures for disease control. The gases most often used in this research were mixtures including Ar or N<sub>2</sub> and CO<sub>2</sub>. Argon and N<sub>2</sub> displace O<sub>2</sub> in the air, whereas CO<sub>2</sub> directly affects the central nervous system as well as displaces O<sub>2</sub>. Carbon dioxide is a well-known anesthetic gas that can induce rapid loss of consciousness, but at high concentrations (>65%) it is known to be an irritant to humans (Lambooij et al., 1999; Gerritzen et al., 2000). Webster and Fletcher (2001) have suggested that moderate CO<sub>2</sub> concentrations are more practical, particularly in on-farm applications. Emergency depopulation in the case of an AI outbreak would need to be conducted on-site at the poultry farm to comply with quarantine regulations. Under field conditions, it is difficult to maintain a tightly governed range of gas concentrations.

Alternate gassing mixtures involving Ar or N<sub>2</sub> rely on dilution of the O<sub>2</sub> in the surrounding atmosphere. To maintain the necessary concentrations of these gas mixtures, the birds would need to be in a closed environment (i.e., depopulation chambers). The use of depopulation chambers is more practical for layer operations and turkeys, but is less practical for broilers, which would require continued handling of many birds. Poultry houses are not airtight environments, and the practice of segregating small groups of birds in a disease control scenario is not only time-consuming but also labor-intensive and increases the potential biohazards.

The most widely used procedures for large-scale emergency depopulation of meat-type birds consist of exposing poultry to CO<sub>2</sub> gas. These procedures include, but not limited to, whole or partial house gassing and various types of enclosures in which birds are gassed in mass numbers. Whole and partial house gassing requires sealing the house to prevent gas leakage, the use of specialized equipment, and the quick introduction of large volumes of gas evenly over the birds. It has been a challenge to rapidly achieve the required gas concentration. In the turkey industry, a portable panel enclosure system has been used. This consists of driving groups of birds into a temporary enclosure, covering with a tarp, and introducing CO<sub>2</sub> gas. The panel enclosure method could be used for batches of up to 5,500 turkeys at a time and required an average of 6:20 (m:s) for audible signs of activity to cease (Kingston et al., 2005). The procedure required 7 workers approximately 2 h to construct and implement the procedure in a typical turkey house. Another procedure used for broilers in Virginia used a metal container placed over a live-haul cage and the container was injected with CO<sub>2</sub> (Kingston et al., 2005). This method was also performed in a batch mode, with 375 chickens per batch, and required an average of 1:28 (m:s) until audible signs of activity ceased. A third procedure called the polyethylene tent method, which has been used for broilers, utilized overlapping layers of polyethylene sheeting to cover birds, forming a tent over the birds and filling the environment under the sheet with CO<sub>2</sub>. These procedures can be very labor-intensive and create a biosecurity risk, with a large number of personnel required to euthanize a typical farm with 75,000 to 100,000 market-age broilers. Depopulating a typical farm with the polyethylene tent method in 1 d would require a labor and support staff estimated in excess of 40 people (Ritter, 2004). Furthermore, the process requires a tractor and additional personnel to remove the polyethylene from the houses. When this material is removed from the house, it may be contaminated with litter and carcasses, which creates another biosecurity risk. Disposal of the contaminated polyethylene sheet via sanitary landfill or on-site burning creates yet another challenge for the procedure. Although these procedures were effectively used to destroy flocks in the recent AI outbreaks, they can be very labor-intensive and include biosecurity hazards when carcasses or contaminated supplies are removed from houses.

There are considerable region-to-region differences in emergency depopulation techniques. Because of the difference in bird type and management, housing, and stocking density, it is difficult to propose a depopulation technique that will be suitable for all circumstances. Kingston et al. (2005) identified complicating factors for mass emergency depopulation, including a lack of personnel with emergency depopulation experience, emotional and physical fatigue, finding and training temporary employees, communicating with employees who were not native English speakers, maintaining employee morale, employee safety, biosecurity, time pressure, and a lack of

proven humane and efficient emergency depopulation methods. Recognizing these and other challenges during the response to the 2004 Delmarva AI event, an alternative emergency depopulation method for meat-type poultry was developed. This new process uses foam generation equipment to create a blanket of fire-fighting foam distributed over the top of the birds. The immersion in the foam causes rapid occlusion of the airway, resulting in cessation of heart activity. With increasing concern over human health risk during an AI depopulation event, this proposed procedure offers the potential to greatly reduce the number of responders and limit exposure to the virus. Fire-fighting foam equipment and 2 types of foam generation equipment were used in this research; however, other foam and foam generation strategies are possible.

## MATERIALS AND METHODS

### Experiment 1

This paper presents the results from 2 experiments that tested the application of fire-fighting foam for mass depopulation of poultry. Experiment 1 was a controlled laboratory experiment that was designed to evaluate the humaneness and cause mode of action of 3 treatments (foam with CO<sub>2</sub>, foam without CO<sub>2</sub>, and CO<sub>2</sub> polyethylene tent method) with 10 replications and 1 bird per replication. Experiment 1 included histology and necropsy examination to determine the cause of death. A total of 10 broilers, including seven to eight 5-wk-old and two to three 10- to 11-wk-old broilers, were used for each treatment.

Each bird was instrumented with electrocardiogram (ECG) sensors. BioPac Student Laboratory edition ECG monitoring pads (Biopac Systems Inc., Goleta, CA) were secured onto the left leg, right wing, and right leg. The ECG equipment was used to determine heart cessation of activity (time of death) of the bird. Blood samples were drawn from 10 birds per treatment pretest and postmortem. Corticosterone assays were conducted using 25 µL of serum diluted in 75 µL of steroid diluent; samples were run in duplicate following the methodologies outlined in the ImmChem Double Antibody I<sup>125</sup> Corticosterone DA Kit (MP Biomedicals, Irvine, CA). Precipitates were counted in a *T* counter for 1 min. The assay, validated for precision (CV = 0.2%) and parallelism (*P* > 0.05), was sensitive to 0.125 ng/mL. Intraassay variability was 2.0%, and interassay variability was 0.5%. Corticosterone steroid levels were analyzed using SAS PROC MIXED (SAS Institute Inc., Cary, NC) with Tukey-Kramer analysis.

Foam was created using 160 mL of Ansul Jet-X high expansion foam concentrate (Ansul Inc., Marinette, WI), 6 L of tap water and 1 Fort Dodge Animal Health Hi-Light blue dye tablet (Fort Dodge Animal Health, IA). Dye was added to assist in the postmortem gross and microscopic observations of the trachea, syrinx, and lungs. The foam was aerated and mixed using compressed air (foam without CO<sub>2</sub>) or compressed CO<sub>2</sub> gas (foam with CO<sub>2</sub>). The dyed foam mixture was reagentated

before use. For the foam trials, the birds were prepared individually and placed inside a 113-L chamber prefilled with foam. For foam, cessation time was recorded as the difference between the time from head coverage to cessation of activity, as measured by the ECG.

The CO<sub>2</sub> polyethylene tent method was used as a control for the experiment. For the polyethylene tent method, the broilers were placed inside a 79 × 65 × 79 cm clear polycarbonate chamber equipped with a CO<sub>2</sub> discharge system and covered with an opaque tarp. No dye was added to the CO<sub>2</sub> gas used for the polyethylene tent method. A polyethylene sheet was placed over the birds, and the edges were weighed down to minimize escaping gases. Carbon dioxide was applied for approximately 60 s, and the CO<sub>2</sub> concentration (61%) was calculated from the flow rate measurements. Cessation time was the difference between start of gas application to cessation of activity, as measured by the ECG.

At 5 min posttreatment, all birds were tested to confirm that there was no heart beat. Cervical dislocation was used to euthanize any bird that was unconscious but not clinically dead. All testing was performed under the approval and guidelines of the University of Delaware Agricultural Animal Care and Use Committee.

Posttreatment, the broiler was removed from the CO<sub>2</sub> chamber or foam chamber, and postmortem blood samples were removed using cardiac puncture. Blood samples were drawn within 2 min after treatment. Pretest and postmortem blood samples were chilled on ice, centrifuged to recover plasma, and stored at -28°C and analyzed for corticosterone steroid levels. Necropsies were performed immediately after treatment. Tissue samples of the trachea and lung were removed from each bird for histology.

### Experiment 2

Experiment 2 was a simulated field trial utilizing a typical compressed air fire apparatus in a small poultry house (11.58 × 14.63 m) with 2 treatments (foam without CO<sub>2</sub>, CO<sub>2</sub> polyethylene tent), 3 replications per treatment, and approximately 208 five-week-old broilers per replication. The treatments were performed on consecutive days.

For each replication, 1 bird per replication was instrumented with Student Laboratory edition ECG monitoring equipment (Biopac Systems Inc.). Blood samples were drawn from 10 birds per pen pretest and postmortem and treated as previously described.

For the polyethylene tent treatment, a gas injection nozzle was attached to a heavy metal object and in the approximate center of the pen. Opaque 6-mil polyethylene sheet material was placed over the birds and secured on all sides. After coverage of the birds, CO<sub>2</sub> gas was allowed to fill the polyethylene chamber. Carbon dioxide was applied for approximately 560 s, and the CO<sub>2</sub> concentration (60%) was calculated from the flow rate measurements. The gas was stopped and the tarp removed 2 min after all bird movement had ceased.

**Table 1.** Comparison of cessation time (s) between foam and the polyethylene tent methods

Treatment	Experiment 1		Experiment 2	
	Average	SD	Average	SD
Foam without CO <sub>2</sub>	64	32	274	94
Foam with CO <sub>2</sub>	73	35	NA <sup>1</sup>	NA <sup>1</sup>
Polyethylene tent	139	26	538	12

<sup>1</sup>NA = not applicable; treatment was not performed in experiment 2.

For the foam treatment, a commercially available Hale Products Inc. (Conshohocken, PA) compressed air foam system fire-fighting apparatus was used to generate the compressed air foam. At the discharge of the pump, a gas injection nozzle was installed in the hose fitting, allowing CO<sub>2</sub> to be injected into the created foam. No modifications were made to the actual pumping equipment. A standard fire-fighting hose and nozzle was used to apply the foam to the birds. National Foam KNOCK-DOWN Class A foam (National Foam, Exton, PA) was used. To avoid the direct stream of the high-pressure foam being directed onto birds, the foam was sprayed against the wall or ceiling of the house and allowed to flow around and onto the birds. A black polyethylene sheet was used to separate the pens to prevent the foam from flowing from pen to pen. Portable poultry pens were used to confine the birds to a smaller portion of the pen (2.13 × 3.45 m). Posttest samples of the foam were drawn from each trial for CO<sub>2</sub> concentration.

## RESULTS

### Experiment 1

In experiment 1, one bird in the foam without CO<sub>2</sub> depopulation group survived a 5-min exposure to the foam and was euthanized using cervical dislocation. Foam characteristics including cohesion, flowability, and moisture content are determined by the foam concentrate, proportion of foam concentrate used, and generation method. Changes in the formulation of the foam mixture can alter the characteristics of the foam. For example, when the foam is too dry, the foam does not flow into areas consistently and can be easily broken down by movement. For the 1 bird that survived foam depopulation, it was believed that the bird found an air pocket in the foam created when the foam was added to the chamber and was able to continue to breathe.

In experiment 1, birds died faster using the foam procedures than with the CO<sub>2</sub> polyethylene tent method, as shown in Table 1. The cessation of activity times were extracted from the resulting ECG data files for each study. Foam with CO<sub>2</sub> required 73 s for cessation, foam without CO<sub>2</sub> required 64 s, and CO<sub>2</sub> polyethylene tent required 139 s. Foam with and without CO<sub>2</sub> were 47 and 54% faster, respectively, than the CO<sub>2</sub> polyethylene tent method. The differences in cessation time were analyzed using Microsoft Excel ANOVA (Microsoft Corp., Redmond, WA), and

the differences between the foam methods and the polyethylene tent method were statistically significant at the 5% level. The differences between the foam treatments were not significant, indicating that the presence of CO<sub>2</sub> in the foam did not change the cessation time.

Postmortem examination through necropsy and histology observed blood in the lumen of the trachea, syrinx, and primary bronchi of a few chickens, as shown in Table 2. The finding of blood is common in chickens undergoing CO<sub>2</sub> euthanasia and results from mucosal capillary leakage. One bird had consolidation in the left lung, and histology results revealed bronchopneumonia as the cause. The bronchopneumonia was not related to the testing performed during the experiment. In the 10 birds in the CO<sub>2</sub> polyethylene tent treatment, no blue dye was present in the tissue, as no dye was used in this treatment group.

In the 10 birds in the compressed air foam without CO<sub>2</sub> treatment, blue dye was consistently present in the trachea. The reason that the dye was not present in the lung relates to the foam dye mixture being blocked by the narrow lumen of the syrinx, blocked by accumulating in the lumen of the trachea, or both. No significant amount of dye entered the lung, because dye did not color the formalin in which the lungs were placed for fixation.

In the 10 birds in the compressed air foam with CO<sub>2</sub> treatment, dye was present in the trachea of 9 out of the 10 broilers. Dye was present in the lung of at least 3 out of the 10 birds. The reason the dye entered some of the lungs in this group, but not the group without CO<sub>2</sub>, is unclear. It may be that CO<sub>2</sub> and dye were inspired before the foam could accumulate to a significant degree in the trachea or syrinx.

Blood, represented by red blood cells and mostly lacking the fluid portion, was present to some degree in the trachea, syrinx, and bronchial tree in all 3 treatments. The presence of blood in the respiratory lobules probably relates to anoxic changes in the endothelial cells of the thin air-blood barrier permitting leakage of blood into the air capillaries with progression to the lumen of the parabronchi (tertiary bronchi) and larger branches of the respiratory tree. The AVMA-recommended methods of depopulation by CO<sub>2</sub> administration results in blood accumulation in the respiratory tree to include the parabronchi. It appears the chickens died of hypoxia in all treatments. There was no evidence of drowning in any of the treated birds.

There was no statistically significant difference in pre-treatment and postmortem corticosterone steroid levels, as shown in Figure 1. Corticosterone levels were higher ( $P < 0.05$ ) postmortem than pretreatment. The differences among all 3 treatments were not statistically significant. There is no apparent difference in stress as measured by corticosterone level among the treatments. The corticosterone levels were similar to studies in which birds were subjected to catching stress.

### Experiment 2

The commercial compressed air foam system used for this experiment was effective and able to successfully

**Table 2.** Histology results, showing the postmortem presence of blood and dye in the respiratory system, experiment 1

Treatment	Dye		Blood			
	Trachea	Lungs	Trachea	Syrinx	Bronchi	Lungs
	no. of birds/total					
Foam with CO <sub>2</sub>	9/10	3/10	0/10	2/10	2/10	10/10
Foam without CO <sub>2</sub>	10/10	0/10	0/10	0/10	5/10	10/10
Polyethylene tent	NA <sup>1</sup>	NA <sup>1</sup>	3/10	2/10	6/10	10/10

<sup>1</sup>NA = not applicable; dye was added to the foam but not to the polyethylene tent treatment.

depopulate birds. Both the foam and CO<sub>2</sub> polyethylene tent methods were able to successfully depopulate the birds in larger groups. The foam method was faster (average 274 s) than the conventional polyethylene tent method (538 s), as shown in Table 1. The times indicated represent the time from when the process was started to when all measurable heart activity as measured by the ECG ceased. The difference in cessation times between methods was statistically significant ( $P < 0.05$ ).

In the CO<sub>2</sub> polyethylene tent method, 1 bird survived past the approximately 600-s treatment period. Three birds that were inside the foam treatment were able to stand high enough for their heads to clear the foam.

The commercial fire-fighting foam equipment delivered the foam at a high pressure (approximately 689 kPa) and flow rate. To avoid the direct force of foam at the birds, the foam was applied against a side wall or ceiling and allowed to flow over the birds. This allowed coverage of the birds without direct application onto the birds.

The achieved CO<sub>2</sub> concentration in the foam was low (1% or less). Because there were no significant differences

in CO<sub>2</sub> concentrations between foam with and without CO<sub>2</sub>, the data were combined.

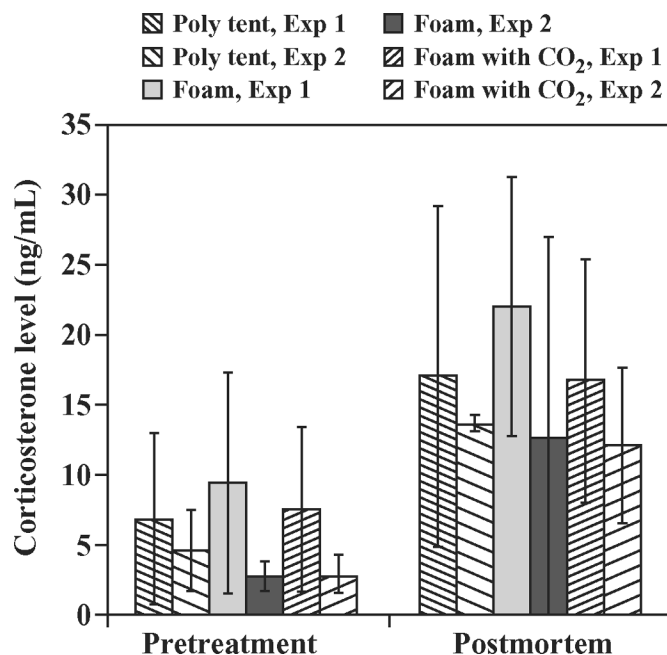
Corticosterone levels were higher postmortem than pretreatment. The difference in stress for a given treatment between the pretreatment and postmortem were statistically significant. The differences between treatments were not statistically significant. There is no apparent difference in stress as measured by corticosterone level between the foam and CO<sub>2</sub> treatment.

## DISCUSSION

The results from experiments 1 and 2 indicate that foam can be used to depopulate meat-type birds. In this research, 2 types of fire-fighting foam and 2 different methods of creating the foam were used. Both methods were equally successful and able to depopulate the birds; it is anticipated that other foam methods may also be suitable.

The foam procedure is relatively quick to implement. Both in individual bird experiments and in the larger field simulation, foam was faster than the CO<sub>2</sub> polyethylene tent procedure. The measured time differences between methods, however, only represent a portion of the time savings associated with foam. With the CO<sub>2</sub> polyethylene tent method, a significant amount of time is required to prepare for depopulation and clean up after completion of the process. Before depopulation, large CO<sub>2</sub> cylinders need to be distributed through the poultry house, the locations of the cylinders marked, overlapping sheets of polyethylene need to be distributed through the house, and the outside edges of the polyethylene sheet need to be buried in the litter for sealing. In contrast, the foam methods require relatively little set up before depopulation. The foam equipment is attached to water and foam sources and, when possible, the birds are herded to a smaller area. At the completion of the depopulation process, the hoses and equipment are disinfected and the equipment relocated to the next farm. The foam is allowed to collapse and degrade over time. Typically, the foam has degraded sufficiently within several hours to allow inspection and subsequent operations (in house composting, sampling, etc.) to proceed.

Foaming has potentially better biosecurity. The foam procedure is conducted entirely inside the house, and the foam acts as a protective blanket to help trap dust and particles inside the house. No contaminated material must be removed from the house and disinfected before



**Figure 1.** Corticosterone steroid levels pretreatment and postmortem, experiments 1 and 2 (Exp 1 and Exp 2, respectively). Error bars represent 1 SD. Poly = polyethylene.

disposal. The foam procedure is expected to be compatible with in-house composting. The foam process adds water to the litter, reducing the amount of additional water required for composting.

The foaming process reduces the labor required to depopulate a poultry house. The CO<sub>2</sub> polyethylene tent process requires an estimated 40 people to depopulate a typical farm in 1 d. The process and equipment used in experiment 2 required less active personnel (a pump operator and a single hose person) than the polyethylene tent procedure. During an outbreak, reducing the number of people involved in the process speeds the process. In addition, reducing the number of people involved reduces the human exposure to viral agents.

There has been some concern associated with chemical composition of the foams. The foam mixtures are proprietary and can include hydrocarbon surfactants, solvents and stabilizers, alcohols, propylene glycol, and corrosion inhibitors. The specific foams used in the process can cause both eye and mild skin irritation through removal of the oils. The birds, however, remain conscious in the foam for a relatively short time, typically <5 min. Several fire-fighting foams meet the biodegradability requirements for forestry use and may be suitable for foam depopulation.

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