A new humane method of stunning broilers using low atmospheric pressure¹

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Primary Audience: Veterinarians, Animal Welfare Auditors, Processing Plant Managers

SUMMARY

The use of toxic gas mixtures to stun broiler chickens before exsanguination continues to be controversial. This controversy concerns which gas mixture to use, the required structural modifications to the processing plant, and whether the methods are actually more humane than electrical stunning. In response to these criticisms, a system was designed and constructed to produce a controlled atmosphere by using reduced oxygen tension. In all these systems, the birds are rendered unconscious from a lack of oxygen. Low atmospheric pressure is achieved by using controlled slow decompression, which allows the body of the bird to adjust to changes in pressure and thus lose consciousness with minimal discomfort. Advantages include the absence of toxic gases, minimal plant modifications, and the ability to view the birds throughout the process via digital video feed, which can be recorded.

Key words: stunning, broiler, low atmospheric pressure, humane slaughter

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DESCRIPTION OF PROBLEM

In recent years, a considerable number of research studies have been performed on improving the slaughter process for broilers. Coenen et al. [1] noted that "slaughter methods for animals are acceptable when they result in minimal signs of agitation and distress during the period that animals have some degree of perception and consciousness" (p. 10). However, the general agreement is that for a method to be humane, it must produce insensibility as rapidly and painlessly as possible. Both electrical stunning and controlled-atmosphere stunning (CAS), using various gas mixtures, have been investigated and are used worldwide [2–8]. However, each has its own disadvantages and critics. Another method of controlling the atmosphere is through anoxia, by which a vacuum pump is used to reduce oxygen tension in the atmosphere. The European Union allows the use of a vacuum chamber for slaughter of farmed game species [9].

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In the United States, decompression has not been considered for slaughter because the American Veterinary Medical Association (AVMA) has ruled that it is unacceptable as a method of euthanasia [10]. The decision of the AVMA was based on several factors, including chamber design. According to the AVMA, "Many chambers are designed to produce decompression at a rate 15 to 60 times faster than that recommended as optimum for animals, resulting in pain and distress attributable to expanding gases trapped in body cavities" (p. 35). Other concerns are the potential for prolonged discomfort in immature animals, accidental recompression, and aesthetically unpleasant physiological reactions in unconscious animals.

The primary objections for using vacuum are the result of rapid decompression, which has been demonstrated to cause "marked abdominal distention immediately" because of the rapid expansion of gases present in the gastrointestinal tract [11]. However, when done slowly with proper controls, it can be humane. Smith [12] reported that, based on electroencephalogram recordings, hypoxia via high-altitude simulation in decompression chambers induces rapid unconsciousness. He noted that in humans, when decompression is "slow enough to allow the body cavities to adjust, the loss of consciousness is almost a pleasant experience and is certainly a painless one" (p. 178). The AVMA monograph on euthanasia notes that proper operation and maintenance by skilled and knowledgeable personnel is necessary when using decompression for euthanasia. However, all this research has been conducted on mammals of varying sizes and ages.

In the case of birds, specifically chickens, gases are not likely to be trapped in the abdomen because of the structure of the respiratory system. This system consists of a pair of lungs attached to the ribs, which are open on both ends. They do not change size during respiration. Attached to these lungs are 9 air sacs that fill all spaces in the thoracic and abdominal cavities and have tubules that extend into several long bones. Because birds do not have a diaphragm, they have to move air in and out by using the intercostal muscles. Avian respiration has no passive or relaxed period. The movement of air in and out is simultaneous and constant. Thus, without blocking the trachea, it is unlikely that significant amounts of gas would be trapped in the abdomen [13].

Early work with a low atmospheric pressure system (LAPS) showed that blood oxygen concentrations decreased from the normal value of 80 mmHg to 23 mmHg immediately after the birds were removed from the chamber [14]. This represents a drastic reduction in blood oxygen concentration, of approximately 85%. Broilers exhibited a 90% reduction in electrical activity of the brain (determined by direct electroencephalography) within 32 s in an atmosphere having a 20% vacuum [14]. Raj et al. [5], in addition to citing Dell et al. [15] and Ernsting [16], reported that loss of consciousness of broilers, as indicated by at least a 90% reduction in electrical activity of the brain, occurred when blood oxygen levels were drastically reduced by exposure to atmospheres of rare gases.

In humans, exposure to low oxygen tension is reported to have an initial stage of euphoria [17]. However, the use of a vacuum for euthanasia of mammals has been tested in the United States and has been determined as unacceptable because of various factors associated with rapid decompression [10]. In the case of domestic hens, in a study using nitrogen to slowly replace oxygen, the birds "slowly became unconscious without showing any signs of distress until respiratory failure supervened" (p. 377) [8]. The AVMA [10] lists objections to the use of decompression for euthanasia based on 4 primary considerations. These include the use of very rapid systems that result in pain and distress caused by expansion of gas trapped in body cavities; tolerance of hypoxia by immature animals, which require longer periods of decompression for respiration to cease; various aesthetically unpleasant reactions by the animals, such as bleeding, vomiting, convulsions, and defecation; and the potential for accidental recompression. It would follow that if there were a method for controlling decompression so that all these factors could be eliminated, then decompression through LAPS could be a humane method of euthanasia and could potentially be useful for commercial slaughter. Testing with a single-bird unit demonstrated that controlled atmospheric pressure reduction could be an effective and humane process [14]. Precision in the controls prevented problems that occurred with previous equipment and thus reduced any potential discomfort for the birds. This work was done using a system for a single bird and had a simple on-and-off switch to control the pressure. The speed of pressure reduction is critical to reducing discomfort reactions from the birds. This finding was in accordance with earlier work on the use of vacuum as a means of euthanasia of mammals [12]. Only limited data can be obtained from a single-bird unit, so it was necessary to develop a research model that could be used to refine the process further. This unit was constructed to hold one commercial cage unit and was put into operation. A series of test runs were completed at both a university and a commercial processing plant setting, and these provided data that allowed for the construction of a 2-cage system with automatic controls. This work set the parameters to be used to develop a research prototype for study.

Work with the prototype led to the development of a commercial unit. For testing the commercial prototype, a protocol was submitted to the USDA office of New Technology Testing Approval so that the unit could be tested under commercial conditions. This phase of research was undertaken to determine the operating parameters to ensure the system could be constructed and operated to provide a humane method of stunning broilers without creating processing problems.

MATERIALS AND METHODS

Commercial Unit

The unit is cylindrical, which provides sidewall strength, and measures 20 to 20.5 ft (6.1 to 6.25 m) in length and 7 ft (2.13 m) in diameter, with a capacity for 2 commercial broiler transport cages of the type typically used in the United States. The atmosphere is manipulated via a variable airflow withdrawal process. The time of pressure reduction and any hold time are controlled via a computer that is programmed to a precise sequence so that human error and climate cannot cause a change that would be stressful for the birds. A video camera with an external monitor is installed so that the activity inside the chamber can be monitored at all times. In the case of a power failure, the unit has a fail-safe mechanism that will immediately open the doors so that the chickens are not held in an atmosphere that would produce discomfort over time. A patent is pending on the system.

Cages move into and out of the unit via a powered transfer conveyor. Each chamber is equipped with a hydraulic door on each end, which opens for cage transfer, and when closed. seals the chamber. Chambers are installed to become an integral part of the plant conveyor system before the dump station. Vacuum pumps with a capability of removing 400 ft³/min of atmospheric air each are connected to the chamber via pipes, and vacuum is applied via pneumatically actuated valves. Evaluations using this system were performed for 6 mo to allow fine tuning of the atmospheric settings for maximizing humane conditions. For confirmation of the final settings, a series of runs were performed over 1 wk, with 10 cycles randomly selected for final evaluation

Experimental Procedures

Recovery. Each time the LAPS unit was in use, recovery of birds was observed continually in the shackling area as well as in any cages removed from the system for testing before processing. Recovery was determined to be any sign of movement by a bird, including eye movement as well as movement of any limb or any sign of respiration.

Variables Examined. Throughout the 6-mo test period, more than 10,000 birds were examined for wing damage. Before placing birds in the unit and after removal, each bird was examined for 5 d for damage to the wings (8 replications/d). Birds were also examined at the picker exit. Damaged wings were counted for 3 min at a line speed of 180 birds/min. This was repeated 8 times each on 5 consecutive days. Thus, 540 birds were examined for broken wings for each period, equaling 4,320 birds/d. In addition, during operation, the birds were observed and measurements of movements such as head shaking, mandibulation, deep breathing, and wing flapping were noted. Once the cycle was complete, the cage was removed from the unit so that the wings could be inspected for dislocated joints, broken bones, and bruising. Along with the wings, the lungs, liver, and intestinal tract were visually examined to verify whether any damage had occurred. Birds were randomly selected for necropsy over a period of 4 d. Ten samples of liver, lung, and breast muscle were taken from randomly selected birds for 4 consecutive days, and samples were placed in formalin for histopathological examination. From the 40 samples collected, 10 were submitted to a board-certified histopathologist for examination. Samples were identified by code.

Blood Flow. To evaluate whether an adequate bleed could be achieved with LAPS and to determine if a delay between stun and exsanguination would produce an adequate bleed, birds were stunned using LAPS and removed for testing before entering the processing plant. To evaluate the bleed, 8 replications with 6 birds/ replication were placed in poultry kill cones immediately after stunning and the carotid arteries were cut. Blood was collected until the flow had been reduced to an intermittent drip. Another group of 4 replications with 6 birds/replication was processed in the same manner but was held for 10 min before having the carotid arteries cut, to evaluate the length of time between stun and exsanguination that would yield adequate blood flow. Ten minutes was selected as the amount of time that might lapse between irreversible stun and bleed under normal operating conditions and before the early signs of rigor.

Dead on Arrival Evaluation. On 4 consecutive days, 1,000 birds were placed in the unit for stunning, dumped onto the shackle line, and shackled for processing. Birds were selected for shackling by the regular crew and the presence of birds dead on arrival (**DOA**) was verified in the shackling area before the birds were shackled.

Behavioral Evaluation. Videotapes were digitized and transferred to a CD for evaluation of movements associated with discomfort or signs of insensibility, such as head shaking, deep breathing, mandibulation, wing flapping, and loss of posture. A series of 10 stunning cycles were reviewed and behavioral actions were timed. Factors included were the time to the first movement, first signs of head shaking, number of bouts of wing flapping, and total wing flapping time. Total wing flapping time was determined by combining the times of the individual bouts and calculating averages.

Corticosterone Assay. Immediately after LAPS or electrical stunning, 4 replications, with 10 birds/replication, were randomly selected and blood samples were taken from the heart for corticosterone assay [18]. The birds were picked up after stunning, and the samples were taken within 45 s of contact. Blood was transferred into EDTA tubes, and plasma was separated by centrifugation ($800 \times g$ for 10 min at 4°C). Plasma corticosterone concentrations in blood samples were determined using an immunoassay [19].

Histopathology and Statistics. A sample $(2 \times 1 \text{ cm})$ was taken from the pectoralis major, lung, and liver of 10 randomly selected birds after LAPS and electrical stunning. The sample was placed in 10% formalin and shipped to a board-certified histopathologist for evaluation. The histopathologist was asked to examine the tissues for any signs of damage or pathology. He was not informed of the treatment. All experimentation was done in compliance with the appropriate Animal Care and Use policies of Mississippi State University. This field study was designed as a completely randomized design for statistical analyses using Statistix 9 Analytical Software [20].

RESULTS AND DISCUSSION

Preliminary work optimized test parameters so that the absence of recovery was expected. Nevertheless, the absence of recovery was carefully monitored and verified before exsanguination. This is the result of developing precise controls without human intervention. This lack of recovery has been noted previously, as required when using other methods of CAS, such as carbon dioxide [3]

Wing Damage

Wing damage caused by treatment was variable (Table 1). Once the operating parameters were optimized to minimize recovery and undesirable behaviors, wing damage was still present. However, we concluded that because of the absence of blood, the damage was postmortem.

| Table 1. Wing damage (%; mean ± SEM) with | low |
|--|-----|
| atmospheric pressure system (LAPS) stunning | VS |
| electrical stunning (ES), as counted after picking | |

| Day | Replications | LAPS | ES |
|------|--------------|-------------------|-------------------|
| 1 | 8 | 8.39 ± 0.46 | 4.97 ± 0.30 |
| 2 | 8 | 6.79 ± 0.53 | 3.76 ± 0.16 |
| 3 | 8 | 6.99 ± 0.49 | 3.46 ± 0.25 |
| 4 | 8 | 7.34 ± 0.36 | 3.55 ± 0.45 |
| 5 | 8 | 7.41 ± 0.92 | 5.06 ± 0.38 |
| Mean | | 7.38 ^a | 4.16 ^b |

^{a,b}Means within a row with different superscripts differ ($P \le 0.0001$).

The percentage of wing damage was greater in birds stunned by LAPS than in electrically stunned birds. This was, at least in part, due to the birds subjected to LAPS being stunned in a crate because even minimal amounts of wing flapping against the walls resulted in damage. The controls have been optimized to obtain minimal movement while ensuring an adequate stun.

Blood Flow

Blood flow from LAPS-stunned carcasses was different from blood flow from electrically stunned and exsanguinated carcasses but was within acceptable ranges for processing. Abram and Goodwin [21] reported that "chickens lose from 35 to 50% of their total blood volume during bleeding operations" (p. 69). Total blood volume is equal to 8.8 to 10% of bird weight, so the expected blood loss was calculated to represent between 3.5 and 5.0% of the live weight of the bird. The actual amounts collected from 36 broilers removed from the line for blood collection after LAPS stun are presented in Table 2. The length of time between stunning and exsanguination that yielded adequate blood flow was evaluated as 10 min after stunning. Blood loss after LAPS stunning was highly variable (Figure 1), although minimal information is available in the literature concerning this issue.

Hoen and Lankhaar [2] performed an evaluation comparing electrical stunning with gas stunning, in which blood loss was calculated by adding the weight of blood collected to the final bird weight to determine the amount of blood loss. Although this method demonstrated similar results for both treatments, it does not adequately reflect the amount of blood remaining in the carcass. To address this deficiency, we measured blood volume collected from each treatment and weighed carcasses before and after exsanguination. The weight difference was reported as blood loss (Figure 1). Neither of these methods is absolute but serve to provide a comparison between the methods of stunning. The only certain result is that no problems in processing occurred in either group of birds caused by an inadequate bleed.

Recovery and DOA

Recovery was verified for each run. The final control settings ensured that recovery would not occur before exsanguination (data not shown). The ability to detect birds DOA at the plant in the presence of dozens of stunned birds was an unknown factor that had to be evaluated. The characteristics that distinguished these 2 groups most effectively were temperature and stiffening, especially of the feet. During the 4-d testing period, no confirmed DOA were presented to USDA for inspection. Thus, LAPS-processed birds did not present a problem with DOA greater than was expected with electrical stunning because they were easily detected at the time of shackling.

Behavioral Evaluation

Perhaps the most important data were obtained from the video recordings, which showed the times and lengths of various behaviors before complete loss of consciousness. Table 3 contains

Table 2. Blood flow 10 min after low atmospheric pressure system stunning

| Day | Avg. bird weight, ¹ g | Calculated blood loss, mL | Actual blood collected, mL | Collected, % |
|----------------|---|------------------------------------|-------------------------------------|-----------------|
| 1 | 3,720 | 117 | 103 | 88 |
| 2 | 4,146 | 130 | 72 | 55 |
| 3 | 2,790 | 85 | 55 | 65 |
| 4 | 2,849 | 89 | 17 | 19 |
| 5 | 3,207 | 101 | 65 | 64 |
| 6 | 3,515 | 110 | 67 | 61 |
| $Mean \pm SEM$ | | | | 59 ± 9.17 |

¹Bird weight is average of 6 birds/d.



Figure 1. Blood loss after low atmospheric pressure system (LAPS) and electrical (ES) stunning based on differences in carcass weight. Carcasses were weighed before and after bleeding and the weight difference was assumed to be blood loss.

observable behavioral data from 10 repetitions of the LAPS process. These data were based on the birds visible to the video camera during each time that LAPS stunning was conducted. Of the total time of evaluation (280 s), only 6% exhibited wing flapping. Wing flapping typically occurs in 3 bursts of approximately 5 s each. The first movement is associated with an awareness of a change in atmosphere that occurs approximately 60 s after pressure is reduced. A period of head movement begins approximately 70 s later. This is followed by wing flapping. Neither mandibulation nor deep open-bill breathing was observed in the birds. Bill breathing and mandibulation are commonly reported in research reports concerning CAS stunning with various

| Replication | Time to first movement, ¹ s | Light-headed, ² s | Bouts of flapping, no. | Total flap time, s | Time from first movement to LOP, ³ s |
|----------------|--|------------------------------|------------------------|-----------------------|---|
| 1 | 51.0 | 96.0 | 3.3 | 17.8 | 84.5 |
| 2 | 53.3 | 96.5 | 2.3 | 15.3 | 99.0 |
| 3 | 70.7 | 38.7 | 2.3 | 14.7 | 38.7 |
| 4 | 64.0 | 90.0 | 2.0 | 13.3 | 72.0 |
| 5 | 66.0 | 83.7 | 2.7 | 13.7 | 63.7 |
| 6 | 71.3 | 65.0 | 2.3 | 12.8 | 64.5 |
| 7 | 56.3 | 29.7 | 4.3 | 16.3 | 29.7 |
| 8 | 42.0 | 66.0 | 3.3 | 15.3 | 66.0 |
| 9 | 71.7 | 80.7 | 2.0 | 14.7 | 80.7 |
| 10 | 41.0 | 47.0 | 3.3 | 17.3 | 17.5 |
| Mean \pm SEM | 58.7 ± 3.02 | 69.3 ± 6.37 | 2.5 ± 0.19 | 15.1 ± 1.12 | 64.9 ± 6.09 |

Table 3. Observable behaviors of broiler birds during low atmospheric pressure system stunning

¹Time to first coordinated movement (stand up or sit down).

²Time from first head movement to first wing flap.

 $^{3}LOP = loss of posture.$



Figure 2. Corticosterone (pg/mL) analysis for electrical versus low atmospheric pressure system (LAPS) stunning. Twelve samples were taken at 4 different times for each treatment; n = 48. Means with different letters (a, b) are significantly different ($P \le 0.05$). Error bars represent the SEM.

gas mixtures [1]. Bill breathing indicates an attempt to obtain oxygen or occurs in the presence of excess CO_2 (deep breathing) or mandibulation (irritation of mucus tissues from CO_2 causing irritation). Coenen et al. [1] observed bird behaviors using electroencephalograms to compare 3 different gas mixtures, and they reported wing flapping events and durations similar to those observed in the present study. They also reported that mandibulation and deep breathing were observed for all 3 gas mixtures [1].

Corticosterone Analysis

Corticosterone concentration was greater (P < 0.05) in electrically stunned birds than in LAPS-stunned birds (Figure 2). It is likely that the lower stress level for the LAPS system was the result, at least in part, of not inverting the live birds for shackling [22]. This is also important because decreased stress levels can minimize meat quality problems, such as pale, soft, and exudative meat, under stressful conditions, such as summertime temperatures in the South. In addition, Battula et al. [23] reported that breast meat quality was excellent from both LAPS- and electrically stunned broilers that

were deboned 2 h postmortem. Other blood parameters are shown in Table 4. These parameters differed only in the CO_2 , O_2 , Na, and Cl^- levels, which are consistent with the change in atmospheric conditions.

Histopathology

Minor inflammation of foci were observed in the histopathology samples taken from LAPSprocessed birds (data not shown). We commonly observed this in processed broilers. This is a marked contrast to reports of hemorrhagic lesions found in the lungs, brain, and heart of animals undergoing rapid decompression [17]. No hemorrhagic lesions were observed through pathological examination of any of the submitted tissues from birds processed using the LAPS system. Overall, no discomfort or sensible damage was observed from stunning with the tested system using low atmospheric pressure to create a controlled atmosphere. Furthermore, based on previously published work with the system used in this study and the results observed in this study, using LAPS results in carcass quality that is at least equal to that of electrically stunned birds [23]. In addition, this system stuns the birds before they are dumped onto the shackling belt, thus eliminating the stress of removing live birds from the crate as well as inverting live birds for shackling. This is the most likely source of the significant reduction in corticosterone levels compared with electrical stunning.

 Table 4. Blood analysis: electrical stunning (ES) vs.

 low atmospheric pressure system (LAPS) stunning¹

| Parameter ² | LAPS | ES |
|---------------------------------------|------------------|--------------------|
| pCO ₂ | 45.3 ± 2.7^{a} | 56.4 ± 2.6^{b} |
| pO ₂ | 23.1 ± 3.7^{a} | 78.6 ± 3.5^{b} |
| HCO ₃ ⁻ , mEq/L | 20.5 ± 0.6 | 24.9 ± 0.6 |
| Na ⁺ , mEq/L | 140 ± 1^{a} | 134 ± 1^{b} |
| K ⁺ , mEq/L | 5.6 ± 0.1 | 5.6 ± 0.1 |
| Ca ²⁺ , mEq/L | 2.6 ± 0.04 | 2.6 ± 0.04 |
| Cl⁻, mEq/L | 128 ± 3^{a} | 148 ± 2^{b} |
| Hct, % | 22.1 ± 0.7 | 20.5 ± 0.7 |
| Hgb, g/dL | 7.05 ± 0.22 | 6.54 ± 0.22 |
| pН | 7.31 ± 0.02 | 7.27 ± 0.02 |

^{a,b}Means in a row with different superscripts are significantly different ($P \le 0.05$).

 $^1 12$ samples were taken at 4 different times for each treatment; n=48.

 ${}^{2}\text{pCO}_{2}$ = partial pressure of CO₂; pO₂ = partial pressure of O₂; Hct = hematocrit; Hgb = hemoglobin.

CONCLUSIONS AND APPLICATIONS

- 1. Low atmospheric pressure system stunning is a new, humane, controlled-atmosphere method of poultry stunning.
- 2. Low atmospheric pressure system stunning eliminates shackling of sentient birds and therefore makes the operation cleaner and easier.
- Low atmospheric pressure system stunning does not use gas mixtures that are stored under pressure and is therefore safer for humans in the area than other CAS methods.
- Low atmospheric pressure system stunning does not use greenhouse gases, reducing its carbon footprint when compared with other CAS systems.

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