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A systematic evaluation of laying hen housing for improved hen welfare

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A systematic evaluation of laying hen housing for improved hen welfare

by

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Chapter 1

General Introduction

Husbandry practices for egg production vary throughout the world. Current production systems are typically classified as caged, cage-free, or free range in terms of housing style. In the US, there is approximately one laying hen per capita, which equates to approximately 350 million laying hens (NASS, 2007), of which approximately 280 million produce table eggs, with a similar number in Europe. The predominant hen housing systems in the US are high-rise and manure-belt battery cages. European nations have a much higher percentage of cage-free and free range systems than the US, as well as enriched cage systems, as a result of increasing concerns or consumer pressure over animal welfare. Some European countries, such as Switzerland and Sweden, have no caged-bird egg production, and conventional cage production will be phased out in the EU entirely by 2012 (Europa, 2006). Economic and management considerations and consumers' choice are the prime factors driving the dominant systems in the US, while legislation and consumer demands strongly influence the European systems.

Laying Hen Housing Systems

Comparative descriptions of characteristics of the different housing systems will be provided in detail in Chapter 2 "Literature Review". The following sections describe some highlights of each system.

Traditional Cages

Traditional cage systems, also referred to as conventional cages or “battery” cages, are the most common housing system in the US (98% of production) and the most prevalent system throughout the world (IEC, 2005). The traditional cage system consists of enclosures constructed of wire or plastic mesh arranged in rows and stacked three to five tiers within a barn. The mesh floor allows droppings to fall into a manure collection area beneath the house or onto a belt which then transports the manure to a collection area for removal. Each cage has 1 or 2 nipple drinkers supplied by a pipe spanning the length of the house. An automated feed line passes along the front of each cage. The mesh floor is sloped so that eggs roll out the front of the cage onto a conveyor belt that transports them to a collection and processing area.

Enriched Cages

Enriched cage systems, also referred to as modified or furnished cages, provide facilities in each cage for roosting, scratching, pecking, and egg-laying behaviors. One example of an enriched cage, The Edinburgh Modified Cage, consists of a cage 600mm wide, 450mm deep, and 450mm high, with a perch, nest box with litter or artificial turf, and a dust bath (Appleby and Hughes, 1995).

Cage-free Systems

Cage-free systems, also called barn systems or perchery systems, house birds indoors without cages. Different types of barn systems may house birds on the floor or provide different levels, and flocks may range in size from a few thousands for floor-raised houses to

over 100,000 in larger commercial aviaries. Typically, the birds are provided nest boxes for laying eggs, areas for perching and roosting, and an area of litter. Egg collection is typically automated and floors may be partially slatted or mesh with manure collection and removal beneath.

Free range

Free range housing systems allow hens to access outdoors part of each day, as little as a few hours. For the remainder of the day, the hens remain in the barn, similar to the cage-free floor-raised situation.

Regulations Concerning Laying Hen Housing

The debate over laying hen housing is nothing new. The discussion intensified in 1964 with the release of a book, entitled Animal Machines (Harrison, 1964). Shortly after the book release, a committee was formed in the United Kingdom to discuss treatment of animals in farm production, and the result of these discussions was the original set of Animal Freedoms (Brambell, 1965). Neither of these publications resulted in recommendation of a ban on traditional cages or confinement, but made suggestions for improving the welfare of confined farm animals. These suggestions went largely ignored, paving the way for legislative action, as evidenced by current European Union (EU) farm regulations.

Regulations in European Union

Formal regulations for housing of laying hens began in various European countries many years ago. For example, formal legislation was initiated in 1963, and Swiss egg

production has not allowed new cage facilities since 1988. Currently, member countries of the EU must abide by specific legislation regarding farm animal husbandry. For laying hens, the phase out of the traditional cage system will be implemented by the year 2012. The systems allowable under the current law are cages which meet specific criteria (average floor space of 750 cm^2 or 116 in^2 per bird, perch length of 15 cm or 6 inch per bird, unrestricted feed access with feed trough of 12 cm or 4.7 inch per bird, nest, and litter for pecking and scratching) or cage-free systems with nests (at least 1 per 7 hens), perches, and no more than 9 hens per m^2 usable area. Also, it is compulsory for eggs sold in the EU to be labeled according to the system by which they were produced: 'eggs from caged hens', 'barn eggs', or 'free range' (EFSA, 2005; Europa, 2006).

Each country is responsible for fulfilling the legislative requirements, including farm inspections and compliance assurance. In the UK, the Department for Environmental, Food, and Rural Affairs (DEFRA) addresses most of these obligations (DEFRA, 2006). In addition to the EU guidelines, each country may set additional regulations of its own. For example, the government of Switzerland imposed an effective ban on all cages 20 years ago. Several other EU countries (Germany, Austria, and Sweden) are scheduled to complete the phase out of conventional cages earlier than the 2012 deadline.

The consumer voice has also exerted great influence in egg production methods throughout Europe. ASDA (Wal-Mart's counterpart in the UK) converted all sales to free-range in an attempt to target a growing contingency of free-range egg consumers. One consequence of this transition was a situation of free-range egg prices temporarily dropping below cost of cage-produced eggs (and below cost for farmers). Prices soon recovered to

profitable margins, but the situation created a temporary situation of losses for farmers (BFREPA, 2006).

Regulations in the US

There is no formal legislation for producers within the US governing the housing of laying hens. The Animal Welfare Act exempts farm animals (USDA, 1990). Although most states exempt normal agricultural practice from prosecution for animal cruelty, some states have laws to prevent neglect, abandonment, and other abuses. For instance, under Pennsylvania law, gross neglect of laying hens has been prosecuted because the severity was not deemed normal agricultural practice (HSUS, 2006); the accused in this case was acquitted (Johnson, 2007), but similar cases are becoming more common in US state courts.

The animal agriculture industry in the US is largely self-regulated. Certain standards are frequently imposed by producer groups and commercial contractors. Association with these groups is voluntary; however, compliance with their recommendations is mandatory for a producer wishing to gain their endorsement. The United Egg Producers (UEP) has set animal husbandry guidelines that its members must adhere to for UEP certification (UEP, 2006). Some of these guidelines require specific criteria to be met, such as cage slope not to exceed 8 degrees and space allowance in the range of 432 to 555 cm^2/hen (67 to 86 in^2/hen). However, other recommendations are arbitrary and may have variations on their interpretation, such as housing that allows hens to stand comfortably and feeder space that allows all hens to eat at the same time. UEP guidelines also address topics including environmental control, cage arrangement, beak trimming and molting.

More recently, direct buyers of large quantities of eggs have set their own hen welfare standards, which must be met by their supplying farms. For example, contracting directly with producers, McDonald's has assembled a panel of experts to develop and evaluate a set of welfare standards for their suppliers of beef, pork, and poultry products (McDonald's, 2006). For laying hens, they require a space allowance of 465 cm^2/hen (72 in^2/hen) with a minimum of 10 cm (4 in) feeder space per bird, and a precisely controlled environment, including uniform lighting, as well as additional guidelines regarding molting and beak trimming.

Additionally, independent auditors offer humane certification programs, such as Animal Welfare Approved by the Animal Welfare Institute (AWI, 2007), Certified Humane Raised and Handled by Humane Farm Animal Care (HFAC, 2007), and Canadian Martime Certified (Henry, 2002).

An emerging approach taken by consumers is to pressure suppliers and food preparers to purchase only eggs produced by a specific housing method. For example, the University of Iowa announced a pilot program in which it will only purchase cage-free eggs from local producers in nearby Kalona, Iowa (Poe, 2006). The trend for purchasing only cage-free shell eggs has also been observed at a few other universities, several upscale restaurants, and recently the US House of Representatives Dining Services (Compass Group, 2007).

Most recently, activist groups such as The Humane Society of the United States have begun aggressively targeting state governments to impose legislative restrictions on agricultural practices (HSUS, 2007). In California, an initiative has been proposed and is seeking voter petition signatures that would place legislation on California ballot in

November 2008. The proposed legislation, entitled the California Prevention of Farm Animal Cruelty Act, includes wording that would prevent housing hens in all types of cages.

Need for Systematic Approach to Evaluating and Assessing Hen Housing

Previous studies have considered individual aspects of hen housing environments, including feeder space, nutrition, cage floors, behavior patterns, etc. However, no studies found in the literature attempted to incorporate multiple measures simultaneously into a housing assessment. It is important to recognize and acknowledge that there is no perfect housing system, and adjusting the system for improvements in one area may inevitably result in undesirable consequences in another area. A systematic approach to quantify and predict these consequences would be valuable and necessary for sound decision-making toward a well-balanced housing system.

A literature review to assess the current situation in laying hen housing and compare systems, including significant research results from both Europe and North American has been conducted and will be presented in Chapter 2. This review revealed a number of information gaps. This dissertation aims to address three of the gaps with laboratory or field studies reported in the subsequent chapters.

Many unknowns arise in the considerations of alternative housing practices. The studies presented in the following chapters focus on interactions with hens and their environment under varying housing conditions. Specifically, considerations were given to quantify conditions, anticipate differences in controlling the environment, and assessing behavior choice responses of hens.

Limited information was found in the literature quantifying environmental conditions experienced by hens in different systems (with bird-level monitoring) under varying weather conditions. It would be expected that differences are present between macro-environments and micro-environments. It would also be expected that differences exist between housing systems. Cage-free houses may have more difficulties than caged houses maintaining comfortably warm temperatures during extreme cold without compromising air quality. On the other hand, caged houses would be expected to have more difficulty limiting temperature increases during hot weather. Additionally, effects of environment on bird health and prevalence of foodborn pathogens have presented conflicting reports in the literature, and weather has not been considered.

With the adoption of reduced stocking density by sectors of the industry, new challenges have been reported for controlling the environment during cold weather. Additionally, little is known about hen ability to cope with heat challenges when given different space allowance or groups of different size. A larger group size allows more range of motion by all birds, and potential for more movement may translate to greater heat and moisture production (HMP).

Most research regarding hen responses to environment has focused on physiological and production changes. Dawkins (1999) highlighted the importance of psychological health and discussed its assessment using birds' own choice behavior. One research group has previously studied active choice responses of hens to environmental conditions via preference testing (Kristensen et al., 2000). The results of their work showed great potential for assessing hen perceptions of environment and factoring these into husbandry decisions. They reported hens displaying a strong aversion to atmospheric ammonia at and above 25

ppm. Other studies have used preference and motivation testing to assess housing and behavioral needs (Dawkins, 1981; Lindberg and Nicol, 1996; Webster and Fletcher, 2004). No additional studies were found using similar methodology to verify the previous results for environmental conditions.

Statement of the Issue

As the debate for proper housing of laying hens grows within the US, so does the need for research-based information that will provide help with correcting misperceptions, filling literature gaps, and allow policy and husbandry decisions made based on science. It is based on this increasing need that a series of related studies were carried out in this dissertation research.

Objectives and Organization of the Dissertation

The following chapters supplement the existing knowledge base for laying-hen housing. Where possible, a systematic assessment approach was used in the comprehensive literature review, and combined field monitoring and controlled-environment laboratory studies. The literature review and experimental studies address the following specific objectives:

1) Review current understanding of advantages and disadvantages and identify knowledge gaps for traditional cage, enriched cage, cage-free, and free-range laying-hen housing systems (Chapter 2);
2) Advance understanding of on-farm housing conditions, by demonstrating advantages and disadvantages of traditional cage and cage-free houses (Chapter 3);

3) Explore unknowns regarding control of environment and hen responses to varying space allowances and group sizes in traditional cage houses with respect to:
    a. metabolic heat and moisture production (Chapter 4)
    b. short-term condition scores and productivity (Chapter 4)
    c. thermoregulation (core body temperature, and mortality) under heat-challenge conditions (Chapter 5)
    d. micro and macro environmental conditions (Chapter 5)
4) Develop a system to assess active responses of laying hens to different environmental factors (Chapter 6), specifically,
    a. Design and build an environment preference test chamber (EPTC) for laying hens that features electronic controls and location monitoring; and
    b. Perform an introductory test on aversion responses of laying hen to atmospheric ammonia using the newly developed EPTC.

Expected Outcomes and Practical Implications

Deliverables from the dissertation efforts provide the available science-based data regarding the impacts of different housing systems and practicing reduced stocking density and group sizes with caged layers on housing environment and hen responses. A preference testing chamber system tool developed in this research endeavor will be used for more studies assessing hen perceptions of the environment. These results are expected to assist the egg industry and regulatory agencies in making more informed, science-based decisions toward modifying production practices. They also contribute to clarification of uncertainties that arise in engineering design for environmental control of laying-hen houses when

conditions deviate from those under which the design data had been collected for the current handbooks (i.e., change in stocking density).

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Chapter 2

Current and Emerging Housing Systems for Laying Hens – A Literature Review

A manuscript to be submitted to Poultry Science

A.R. Green and H. Xin

Introduction

Laying-hen housing may have different schemes in modern production agriculture, including traditional cages, enriched cages, cage-free floor-raised house or aviary, or free-range system. Each housing scheme has come into practice for various reasons as the scale of production has increased from the family farm to commercial-scale operations. Each system offers benefits to the producer, the bird, the consumer, or a combination. Unfortunately, none of these systems is perfect because of certain inherent limitations or negative aspects associated with each.

Morrow-Tesch (1997) stated that the outcome of any animal production unit should have the following goals: 1) a system of raising farm animals that enhances well-being, 2) a safe and pleasant environment for farm workers, 3) being ecologically sound, and 4) producing a safe food product that consumers can afford. In the past 40 years, egg production practices have changed and diversified to meet requirements of various entities in the production process. In a review of Swiss egg production, Studer (2001) stated that no commercial system provides what a hen really wants: a small free-roaming group, of approximately 30 birds, with a cock and chicks. Instead, we must strive for a balance

between what is desirable for the animals and what is viable to produce safe and affordable food. Armstrong and Pajor (2001) noted the need to find ways to enhance animal welfare in an economically and environmentally sustainable fashion. Fraser (2002) noted that as husbandry guidelines change, it is important to have sound research and expertise to ensure acceptable methods are accessible and well-tested; economic conditions are favorable for the changes; regulatory environment adequate to meet needs of producer and consumer; and organizational leadership for animal industries to anticipate and prepare for emerging issues.

The objective of this literature review is to comparatively review advantages and disadvantages, as reported in the literature and/or field experiences, and identify information gaps concerning four predominant types of modern laying-hen housing systems: traditional cage, enriched cage, cage-free barns, and free range. The comparative description and discussion of the systems are based on *management requirements, welfare of the hens, economics,* and *food safety*. Table 1 provides a side-by-side comparison of the systems, summarizing the discussion that follows here.

Management requirements

Management is the most important aspect of responsible farming. The perfect system in theory can be the worst in practice if the management is misaligned. Management can make the difference between welfare and cruelty, safe and hazardous, profit and deficit. Some of the most important facets of management are related to the decisions and oversight regarding animal housing and care, facility maintenance, worker training and supervision, and environmental stewardship. There are decisions regarding animal husbandry that must be made for the initial design and the ongoing operation of a facility, and it must be appropriate

for the entire life of the birds. Studer (2001) discussed two studies in Switzerland, looking at the effects of skilled and unskilled staff, which showed that given the exact same system, staff can make the difference between positive and disastrous results for animal welfare and farm profitability. Temple Grandin strongly stresses the importance of management for welfare (Grandin, 2006). She recommends that a manager set their farm involvement level such that he/she spends some time with the animals and the workers, but not enough time to become desensitized to the environment and the potential for occupants to suffer if conditions become bad. She also emphasizes the importance of good management for profitability.

A report on poultry welfare in North America states that good management can minimize welfare problems and that knowledge about improved methods must be communicated to managers and their staff (Mench and Duncan, 1998). Notable in that report is the lack of understanding of European animal welfare research amongst North American scientists and industry, and the potential to improve housing systems by considering all perspectives. This review includes both North American and European research results.

Hen Welfare

One approach for assessing welfare of animals is through application of the five animal freedoms. In 1965, the Brambell Committee met in England and developed the original set of Animal Freedoms (Brambell, 1965), the things which every captive animal should be afforded to maintain full welfare. These were re-evaluated and refined by the Farm Animal Welfare Council in 1993 (FAWC, 1993). The five freedoms are:

1) Freedom from thirst, hunger, and malnutrition by ready access to fresh water and a diet to maintain full health and vigor
2) Freedom from discomfort by providing a suitable environment, including shelter and a comfortable resting area
3) Freedom from pain, injury and disease by prevention or rapid diagnosis and treatment
4) Freedom to express normal behavior by providing sufficient space, proper facilities, and company of the animals of its own kind
5) Freedom from fear and distress by ensuring conditions that avoid mental suffering

These five freedoms are generally accepted as the standard for welfare assessment, and no argument against them was found in the literature whether they are too critical or too weak.

Dawkins (1999) noted three erroneous assumptions for welfare assessment: 1) There are general indicators of welfare that apply to all situations; 2) Indicators of good welfare and those of reduced welfare are distinct from one another; 3) Any change in welfare 'indicator' reflects a change in state of welfare. Instead, the focus should be on the purpose of the responses to determine suitability of the response. The most important parameter of welfare is physical health, and second, psychological health, which may be assessed using methods of choice behavior. Kirkden et al. (2003) considered the consumer demand theory for assessing animal motivation and needs.

Traditionally, the US perspective has considered production and performance characteristics as a benchmark for welfare assessment. Mench (1992) concluded that good

productivity and health are not necessarily indicators of good welfare, especially when viewed in isolation. As with Swiss example, a simple list of indicators may be used to judge animal welfare of a housing system, with importance placed on incorporating and understanding of behavior and function into the housing design (Wechsler et al., 1997). On the opposition to this approach, Curtis (2007) contended that performance should be considered more importantly than behavioral patterns in well-being assessment.

Economics

Economics are an important consideration for commercial animal production because farms must be profitable in order to sustain themselves. In addition to providing safe and affordable food to Americans, the US farm system is responsible for a significant contribution to our economy. According to the 2002 Agricultural Census, animal agriculture product sales totaled $105 billion, and the poultry and egg industry comprised 22.7% of that total (NASS, 2004).

Food safety

Health concerns impact not only the welfare of the birds, but also food safety of the consumer. Every effort should be made to reduce human infections of food origin and to maintain consumer confidence in food safety. The USDA has identified reduction in annual cases of foodborn illness a priority, with pathogens known to be carried by laying hens of the most concern for contamination of egg products. *Salmonella* causes nearly 1,343,000 cases of foodborne illness resulting in ~ 15,000 hospitalizations and ~ 500 deaths annually (Mead et al. 1999). *Campylobacter spp.* causes nearly 2 million cases of foodborne illness resulting

in ~ 10,000 hospitalizations and ~ 100 deaths annually (Mead et al. 1999). *Campylobacter jejuni* and *C. coli* are frequently reported in clinically healthy live birds and poultry meat, but infrequently in egg products (Kapperud et al., 2003; Neal et al. 1995; Stern et al. 2003).

Because human cases of foodborn *salmonellosis* are linked to consumption of *Salmonella* contaminated poultry and eggs, the USDA launched a *Salmonella* reduction initiative in 2006. The Center for Disease Control and Prevention (CDC) has targeted reduction of human *salmonellosis* from 18 cases per 100,000 population in 1987 to 6.8 cases per 100,000 by the year 2010 (CDC, 2007). Similarly, the CDC projects a decline in human *campylobacteriosis*, resulting from food contamination by *Campylobacter*, from the 1987 baseline (50 cases per 100,000) by the year 2010 (12.3 cases per 100,000). To reduce human foodborne illness, on-farm pathogen reduction strategies strive to deliver poultry, meat, and eggs to the American consumer that are free of *Salmonella* and *Campylobacter*.

Confinement Methods of Modern Commercial Laying-Hen Housing

Intensive farming (and subsequently the caging) of laying hens became widespread shortly after World War II, as a result of improving economies and an increasing number of families that could afford to purchase more meat and egg products. Prior to this time, most egg production occurred on small family farms. Intensive farming offered several benefits to housing large numbers of animals in small areas, including protection of the animals from negative influences such as weather and predators, year-round supply of optimal temperatures and fresh air, elimination or significant reduction of exposure to infectious diseases, and supply of clean fresh feed (Studer, 2001). Since that time, housing methods for intensive farming have evolved into highly elaborate, technologically advanced systems,

which makes possible and affordable the number of eggs we consume annually. The systems may easily be divided into two different types, those with cages and those without. The farms on which they are located may range in size from a few thousand to a few million hens.

Cage Systems

Invention of the original cage system has been attributed to German farmer Paul Collignon, with the intention of solving the problems being faced with egg production at the time (Studer, 2001). The 'battery' cage (meaning a collection of cages) system was effective at reducing hygienic problems including eggs being in contact with droppings, increasing hen performance, reducing the death rate from disease, and reducing feed requirement (intake + wastage) per egg produced. This cage system was reported to drastically improve welfare conditions. Modern cage systems vary in construction materials, space allowance, arrangement, and furnishings.

Traditional Cage Systems. Traditional cage systems, also referred to as conventional cages or 'battery' cages (Figure 1), are the most common housing system in the US and the most prevalent system throughout the world, with 90% or more farms using this system in the US, Canada, Mexico, Brazil, India, China, Russia, Japan, and many other countries (IEC, 2005). The traditional cage system consists of enclosures constructed of wire or plastic mesh arranged in rows and stacked three to five tiers within a barn. The mesh floor allows droppings to fall into a collection area beneath the house or onto a belt which then transports the manure to a collection area for removal. Each cage has 1 or 2 nipple drinkers supplied by a pipe spanning the length of the house. An automated feed line passes along the front of

each cage. The mesh floor is sloped so that eggs roll out the front of the cage onto a conveyor belt that transports them to an egg collection and processing area. Cage and group sizes may vary among producers, but a typical cage might be 51 by 61 cm (20 by 24 in), housing 6 to 8 birds, for 387 to 518 cm^2/bird (60 to 80 in^2/bird) (Chore-Time, 2007). Lighting provisions vary, and generally operate on timers to control photoperiod, typically 16L:8D, for adult laying hens (Hy-Line, 2007). Ventilation systems, cross or tunnel in style with fans and ceiling or perimeter inlets, provide fresh air to the houses. Negative-pressure ventilation system is most common, although positive-pressure ventilation system can be found in some cases. Typical barns may range in size from 15 m by 150 m (50 ft by 500 ft) to 27 m by 150 m (90 ft by 500 ft, i.e., double wide), and farms might have as many as 20 barns on the same site. Traditional cage houses are typically catergorized into two types that vary in manure collection method; conveyors are located beneath the cages for frequent manure removal in manure-belt houses and manure falls into a collection and storage area beneath the cage area in high-rise houses.

Under EU legislation, traditional cage systems will no longer be allowed after 2012 (Europa, 2006). The foundation for the EU ban on traditional cages was almost entirely due to welfare concerns (by the scientific community, animal rights activists, and the general public), specifically the concerns over lack of adequate space for performance of behaviors, lack of spatial enrichment, and increased risks of bone breakage and osteoporosis within traditional cages (Appleby and Hughes, 1991; Baxter, 1994). EFSA (2005) summarizes current regulations based on the most recent scientific studies.

Enriched Cage Systems. Enriched cage systems, also referred to as modified or furnished cages, are the allowed cage alternative for traditional cages under the most recent

legislation in the EU (Europa, 2006). The development of the systems occurred almost entirely in European countries, and the implementation of the systems remains almost entirely in EU countries. In 2005, Sweden reported only 3% traditional cage houses and 36% enriched cage houses, with the remaining production in cage-alternative housing; though not all reporting countries divided production within the cage category (IEC, 2005).

The design of the modified cage occurred over several years with a number of researchers attempting to address what 'needs' of the hen were not being met with the traditional cage system. Studies showed that hens were highly motivated to perch at night (Olsson and Keeling, 2002). The addition of perches also showed benefits for bone strength and reduced osteoporosis (Duncan et al., 1992). Other studies showed that laying their egg in a nest box was of importance to the hens, placing a high value on gaining access to a discrete nest site prior to oviposition (Cooper and Appleby, 1996a; Freire et al., 1996; Cooper and Appleby, 1997; Freire et al., 1997; Cooper and Appleby, 2003). In one instance, hens learned to reverse open a mechanical door intended to keep them out of an area in order to lay eggs in that area (Smith et al., 1990). Several studies attempted to assess the value of dustbathing to the hens. The studies showed that hens would dustbathe if given the opportunity, but hens were not willing to work as hard to gain access to a dustbath as for the perch or the nest box (Faure, 1991). The purpose of dustbathing is to control lipids on feathers (van Liere, 1992), and initiation of dustbathing behavior appears to be more complex than initiation of perching or nest-seeking. Widowski and Duncan (2000) concluded that dustbathing motivations better fit an 'opportunity' model than a 'needs' model. A needs model would indicate an essential behavior which would result in the potential for suffering

with deprivation, whereas an opportunity model indicates a desirable but not essential behavior.

It is not recommended to implement some of the enriched cage features without implementation of the others because the success of the modifications are interrelated, such as increased incidence of broken eggs when perches are supplied without nest boxes. The cage enrichments are used considerably by hens in enriched cages and show welfare benefits over traditional cages if properly designed, constructed, placed, and managed (Abrahamsson et al., 1996; Tauson, 1998). This was confirmed first with initial studies at laboratory scale, and further supported by research in commercial houses over an extended period of 3 or 10 years (Wegner, 1990; Appleby et al., 2003)

Based on these and many other studies, several generations of cage modifications were explored, including the getaway cage (Wegner, 1990) and the Hans Krer System (Norgaard-Nielsen, 1990). The Edinburgh Modified Cage was one of the most successful furnished cages concepts, based on the improved behavioral repertoire, with fewer negative consequences such as broken eggs and increased aggression. It consisted of a cage 600mm wide, 450mm deep, and 450mm high, with a perch, nest box with litter or artificial turf, and a dustbath for housing a group of 4 hens at 675 cm^2/bird (104 in^2/bird) plus 281 cm^2/bird (44 cm^2/bird) in nest box area. (Appleby and Hughes, 1995). During its development and research trials, the Edinburgh Modified system housed ISA Brown hens. Getaway cage includes similar features with a different arrangement (Wegner, 1990). A comparison of getaway and Edinburgh Modified cages revealed better production and lower mortality for Edinburgh Modified cages (Abrahamsson et al., 1995).

Cage-free Systems

Cage-free systems, also called barn systems or perchery systems, house birds indoors without cages. The different types of barn systems may house birds on the floor or provide different levels. Typically, the birds are provided nest boxes in which to lay eggs, areas for perching and roosting, and an area of litter. Egg collection is typically automated and floors may be partially slatted or mesh with a manure collection and removal system beneath. Cage-free systems may be further divided into floor-raised system and aviary system.

Floor-raised system. A floor-raised or deep litter system is characterized by a single level of birds, typically with a slatted floor over a manure collection area and an area of litter (Figure 4). Deep-litter systems are generally not practical for large scale production, though flock sizes may be up to 10,000 hens.

Aviary system. An aviary is a multi-level system with litter on the floor and manure removal on two or more levels, with tiers and perches at different levels, and separate areas for different behavioral functions (Figure 5). Aviary systems are typically found in large commercial facilities, where it is desirable to house a large number of birds in a small area.

Free-range Systems

Free-range housing systems provide a period of time each day when hens are allowed access to an outdoor area, as little as a few hours. For the remainder of the day, the hens remain in a barn typically like the cage-free floor-raised barns described in the previous section. In general, the barn has small doors along the sides called pop-holes that are opened mid-day (after egg-lay) and closed up near dusk when the hens have returned to roost for the evening. According to recommendations in the United Kingdom (UK), there should be no

more than 1000 birds/hectare (400 birds/acre) (RSPCA, 2006). Many free-range farms produce organic eggs, and the size of organic flocks in the EU is limited to 3000 hens (DEFRA, 2008). In the US, the FDA has no set standards for use of the term 'free range'.

Management Considerations for the Housing Systems

Responsible husbandry and labor requirements

In general, keeping hens in cages makes it simpler to provide proper care to and maintain the equipment, except regular observation of individual birds and birds housed on lower levels. It may be more difficult to identify mortalities in cages, although no documentation of this was found. It is easier to catch individual birds for removal from a cage, but the process is more tedious for placing or removing an entire flock. More labor is required for more extensive systems in terms of observing and caring for the birds as well as operating, managing and maintaining equipment and furnishings. Training of workers is important in all of the systems, especially regarding handling and interacting with the birds.

Egg collection, feeding, and watering can be and are typically automated in all the housing systems. Floor eggs are one of the biggest problems in cage-free systems. They are more labor intensive for collection and are frequently downgraded (VanHorne, 1996). The problem is greater when the flocks are young, as many floor-laying birds learn to use the nests for egg-laying over time. One study found that 80% of floor eggs were laid by the same hens (Cooper and Appleby, 1996b). This study speculated that the nests provided were somehow deemed unsuitable by these hens because they also exhibited greater nest-seeking behavior than non-floor-laying hens.

Manure Management

Different manure handling systems can be applied in the various housing methods, and it is important to consider collection, removal, storage, disposal, and emissions. Typical manure handling for cage systems is either manure-belt or high-rise storage collection. Cage-free housing systems commonly incorporate a combination of manure management schemes. The quantity of manure to be managed varies by farm type and size: the larger the farm, the more waste that must be handled.

Manure-belt collection and removal. For a manure-belt system, manure drops onto a belt beneath each row of cages. At a given interval, e.g., once a day, twice a week or once a week, manure is carried via the belt to one end of the house and removed to an on-farm or remote storage area. The initial investment for the manure-belt system is much greater than the high-rise system (about 50% higher); however, it has significant benefits. Manure removal from the manure belt house is less labor intensive than the other methods, but maintenance of the belt components is critical. The air quality (e.g., ammonia and dust concentrations) within the houses is generally much better than with other manure management systems. When the belt is in operation it passes from one end of the house to the other, and while the belts are moving the underside has potential to drop tiny particulates onto the birds below. Because the manure is removed from the house on a regular basis, the houses in which they are operated typically have significantly lower emissions. The manure removed from the house must subsequently be stored or immediately applied to the land, and emissions from the storage facility vary greatly by the storage conditions (ambient and manure temperature, manure moisture contents and manure stacking configuration) (Li, 2006). However, storage facilities may be ventilated at much lower rates because exposure

risk is reduced to humans and birds, and this can also reduce emissions of atmospheric ammonia.

High-rise collection and storage. For a high-rise manure collection and storage system, manure drops into a holding area beneath the cage level. Ideally, the ventilation is such that fresh air is brought in at the bird level and passes the manure storage just before exiting the house. However, in cold weather when ventilation rates are low, the ammonia concentrations may rise to and increase at the bird and worker level (the upper level of the high-rise structure). Manure is typically removed from the storage once a year (in the fall). Emissions from these houses are much greater than manure belt houses (Liang et al., 2005). Removal of manure is more labor intensive but occurs less frequently, and maintenance of manure handling equipment is less demanding.

Littered floor. For systems with hens reared on a littered floor or partially littered floor, manure collects on the floor, and is typically removed between flocks. The management of the littered floor has a significant effect on the ammonia concentrations within the barns. Regular additions of fresh sawdust or wood shavings can reduce the moisture content in the litter and thus the ammonia released into the air of the barn. Generally, littered floor houses have slatted areas where manure can fall into a collection area to be periodically removed. For free-range houses, some of the manure is excreted on pasture, and thus does not have to be collected and stored. However, this makes pasture management a critical issue for free-range systems, and results in greater environmental foot print.

Environmental Control

Control of the environment is a critical consideration for housing systems, for the welfare and optimal production of the birds, as well as the health and safety of the workers within the barns. Adequate ventilation is essential to provide comfortable temperature, relative humidity, ammonia, carbon dioxide, dust, and other potential air contaminants.

Heat and moisture production (HMP). Sizing of equipment and housing configuration to provide adequate ventilation is partially based on HMP of the birds and their housing system; therefore it is important to have current and relevant understanding of the effects of different housing systems on control of the environment. Most recent HMP data have shown appreciable differences for modern birds as compared to previous data. HMP of birds today is greater than that of bird strains 20 years ago (Chepete et al., 2004; Chepete and Xin, 2004). In addition to the effects of genetic strains, it is also likely that HMP is different for birds in different proximity to one another (as with varying stocking density), and it is possible that different housing systems and thus varying locomotion and feed intakes also affects HMP. HMP comparison for different cage stocking densities or different housing systems was not found in the literature.

Environmental temperature and relative humidity (RH). Comfortable air temperature and RH promote better bird health and improved production. Temperature should be maintained at thermoneutral conditions for the birds whenever possible. To control temperature and RH, higher ventilation rates are typical for warm seasons and lower ventilation during cold weather. Heat stress, one consequence of inadequate ventilation in hot weather, significantly reduces the performance of the birds. In addition, heat stress also inhibits immune function, reduces body weight and feed consumption, and negatively

impacts production (Mashaly et al., 2004). Methods for heat stress relief include evaporative cooling of inlet air and misting or fogging of airstreams. Partial surface wetting has shown potential for cooling caged laying hens (Chepete and Xin, 2000; Yanagi et al., 2002); however, it is not yet a standard practice.

Ammonia. Ammonia is a pollutant released from manure, and concentrations within some houses reach levels dangerous to health of birds and human occupants. Research has shown that concentrations of 25ppm are highly aversive to hens (Kristensen et al., 2000). Under UEP guidelines, atmospheric ammonia concentration should ideally be less than 25ppm, and should not exceed 50ppm except if temporarily unavoidable (UEP, 2006). Human exposure limits have been set at 25ppm by the US National Institute of Occupational Safety and Health (NIOSH) and 50 ppm by the US Occupational Safety and Health Administration (OSHA) by 8h time weighted average (OSHA, 2006). However, concentrations commonly exceed these limits in littered floor and high-rise pit houses and some naturally ventilated houses, especially during cold weather when ventilation rates are lower. Other methods for providing lower ammonia concentrations at bird level include the manure belt system, where manure is removed frequently, or periodic addition of fresh litter to littered floors. One study compared a traditional cage system vs a deep litter system (Appleby et al., 1988a), in which a low-cost conversion of a deep-pit cage house to a deep litter cage-free system with a slatted floor over the pit was completed. Total egg production was lower for the birds on litter, and dust and ammonia levels were high for the floor-raised system. Ammonia levels were also high for the deep-pit manure storage.

Wathes et al. (1983) highlighted the problem of ventilation solely for thermal comfort may result in an environment with poor air quality, which can result in another set of

problems. Wathes (1998) suggested that interactions between exposure to aerial pollutants and respiratory effects should be further explored in poultry. A summary of relevant literature at the time showed ammonia concentrations varied for the reporting countries from 1.6 to 11.9 ppm for caged layers and 8.3 to 29.6 ppm for cage-free houses.

Dust. Dust can be problematic and is the most difficult to control in confinement systems at commercial scale. UEP does not offer specific guidelines for hen exposure to dust. Human exposure limits are 15 mg/m^3 for total dust and 5 mg/m^3 for respirable dust by 8 h time weighted average (OSHA, 2006). Several articles reported emission values (Wathes, 1998; Liang et al., 2005), but measurements at bird level were not found for the different systems and would be expected to vary considerably.

Fly control. Fly control strategies have been developed for cage facilities, including frequent manure removal for manure-belt houses and applications of insecticide in high-rise houses (Bell, 2002). No reports or comparisons for alternative houses were found in the literature.

Facilities maintenance

Equipment maintenance is essential for any system with automated egg collection, feeding, drinkers, ventilation, etc. The more elaborate the automation, the more requirements for maintenance. More extensive housing systems require additional maintenance for furnishings. Dustbaths within enriched cages can be especially challenging; though mat scratching areas, an acceptable alternative, alleviate the challenge of dustbaths.

Environmental responsibility

Several important areas for management to consider carefully are related to land usage, land impacts, and air pollution. Part of management must address: minimizing environmental impact, reducing pollution, reducing soil erosion, improving working environment for people, minimizing resources utilization while maximizing animal performance (Estevez, 2002).

Cage houses make the most efficient use of space, housing a large number of birds in a smaller area. The most land demanding system is free-range. One major problem for free-range producers is the space requirement for ranging outdoor birds and the potential environmental destruction of large numbers of birds on pasture. One pasture management practice involved rotation schedules between birds and crops (DEFRA, 2001; Glatz et al., 2005). Since hens receive no nutritional benefits from pasture, the primary benefit of a pasture-raised system of egg production may be environmental or ethical (Clancy, 2006).

A review by Wathes (1998) highlights several European studies that document emissions of ammonia, methane, nitrous oxide, odor, and dust from poultry farms. Atmospheric ammonia is the predominant pollutant gas in poultry production facilities, and emission rates are higher for high-rise than manure belt for conventional cage systems (Liang et al., 2005). Many large farms have been identified as sources of high ammonia emissions.

Under EU legislation, either aviary or floor-raised systems are acceptable cage-free systems (Europa, 2006). deBoer and Cornelison (2002) developed a method for assessing sustainability of housing systems. Based on equal importance of all indicators, the traditional cage system was most sustainable, followed by aviary, then floor-raised. Improvements to

economic performance of cage-free systems or alterations to the weighting of indices may change the result.

Welfare Considerations for Housing Systems

Evaluating a housing system based on the five freedoms includes consideration for all facets of the environment. Proper nutrition and adequate water can be provided in all systems, though competition amongst birds may vary. Proper feeder space is essential for any system, but is not agreed upon within the literature or the regulatory communities (Faure, 1986; EFSA, 2005; UEP, 2006). Control of temperature and air quality was discussed under the *Management* section, and will not be repeated in this section. There are many factors to consider for optimizing cage configuration: cage size, feeder space per bird, group size, genetic strain, housing type, number of cage levels, lighting program, nutrition, and dozens of others (Bell, 2002).

Stocking density has been the topic of ongoing debates in the US. The UEP recommends space allowance between 432 to 555 cm^2/bird (67 to 86 in^2/bird) for white and brown genetic varieties, and McDonald's requires a minimum of 465 cm^2/bird (72 in^2/bird) from its suppliers. However, for the few producers who are not UEP members and do not contract with McDonald's or a similar buyer, compliance with these recommendations are voluntary, and some farms stock as densely as 310 cm^2/bird (48 in^2/bird). For the original cage system, Collignon reportedly recommended 800 cm^2/bird (124 in^2/ bird), based on his own field experience, though no specific details were given in the reference (Studer 2001).

Numerous studies have shown benefits, not only for bird welfare, but for production parameters for lower stocking density. Reduced space allowance beyond a critical point has

many negative effects: 1) increased mortality, 2) decreased hen-day production, 3) more egg breakage, 4) reduced net profit per bird, and 5) variable effect on total profits (Bell, 2002). Dawkins and Hardie (1989) showed that hens (Ross Brown, 1.9 to 2.6 kg) require 540 to 1006 cm^2 (84 to 156 in^2) to turn around, 653 to 1118 cm^2 (101 to 173 in^2) to stretch wings, 860 to 1980 cm^2 (133 to 307 in^2) to flap wings, 676 to 1604 cm^2 (105 to 249 in^2) to ruffle feathers, 814 to 1270 cm^2 (126 to 197 in^2) to preen, and 540 to 1005 (84 to 156 in^2) cm^2 to scratch the ground. Because behavior patterns are socially initiated, it is important for birds to perform behaviors simultaneously. The hens in this study were larger than many common strains in the US (CV-20, W-36). Nevertheless, in comparison, the largest recommended cage-space allocation in the US is 432-555 cm^2/hen, (67 to 86 in^2) (UEP, 2006).

Studies have demonstrated that sufficient space, both physical and social space, is important to hens (Hughes (1975), Keeling, 1995, Lindberg and Nicol, 1996). Dawkins (1981) showed that hens prefer a larger cage with more space (0.76 m by 0.86 m versus 0.38 m by 0.43 m), but place a higher priority on flooring (litter versus wire mesh). Keeling (1994) revealed that when insufficient space was available, not all behaviors were performed in cages. Nicol (1987) showed that increasing cage height and cage area increased the rate of performance of positive behaviors, including head stretching and scratching, and reduced negative behavior, including cage pecking. The study concluded that spatial restriction may increase the cost of performing certain comfort activities and reduce the rate of performance of those activities. When offered unrestricted space in an enriched environment, hens demonstrated predictable behavior patterns (Mishra et al., 2005).

Savory et al. (1999) reported that feather damage varied with group size and stocking density interactions, and was greater for large groups (20 birds) at higher stocking density

(186 cm^2/bird, 29 in^2/bird) and least for small groups in a lower stocking density (10 birds at 744 cm^2/bird, 115 in^2/bird) for bantams in wire mesh cages. On the contrary, Moinard et al. (1998) reported that feather condition was independent of cage space allowance.

Production numbers consistently favor lower stocking density (Lee and Moss, 1995; Altan et al., 2002; Anderson et al, 2004). Cook et al. (2006) reported no difference in feed intake or meal duration for hen housed at 348 to 465 cm^2/bird (54 to 72 in^2/bird). Many farmers opt to use higher stocking density to increase total production and reduced overhead costs per dozen eggs. This is not always the most economical decision because when economic margins are low, higher densities are less profitable (Bell, 2002). Using fuzzy logic based on performance parameters of egg production and mortality, Roush and Cravener (1990) determined the crossover point between a crowded and uncrowded cage was between 3 and 4 birds in a cage 4645 cm^2 (1161 and 1548 cm^2/bird, 180 to 240 in^2/bird respectively) and 3 birds for cage size 1548cm^2 or 3097 cm^2 (516 or 1032 cm^2/bird 240 to 480 in^2/bird). The results showed the larger cage was crowded at a lower stocking density.

A 1997 review highlighted the need for additional understanding of interactions between space allowance and group size (Barnett and Newman, 1997). Numerous studies investigated behaviors and motivations at different stocking densities. However, effects of space allowance on micro-environment and tolerance of heat stress were not found in the literature; neither was the effects of space allowance on rates of mortality and cannibalisms.

A review by Jones (1996) considers fear responses, and notes the importance of neither understimulating or overstimulating hens. Important considerations of this review highlighted that many farm systems prevent responses from fear (like fleeing a stimulus). Conventional cages reduce the incidence of frightening events, but that, in turn, precludes a

wide range of sensory inputs and results in inadequate behavioral repertoire and stereotypic behavior patterns and vices. Environmental enrichment, handling regularly, genetic selection all show potential to improve fear responses. Jones (1996) suggests a goal of providing stimulating, safe, economically viable environment.

Barnett et al. (1994) reported that increasing human contact by 15 min/day reduced fear and immunological responses of caged layers, and also increased production. It may additionally be possible to manipulate the fear of human response to improve welfare of hens, and well as performance (Hemsworth et al., 1993).

Current debates within the US are just beginning to address the main welfare areas in which traditional cages fall short. Duncan (1998) considers that performance of necessary behaviors leads to increase in health or physical condition. A behavioral 'need' will inevitably arise and is controlled by internal factors present no matter what type of environment is provided. Braastad (1990) demonstrated the potential for redirecting abnormal behaviors by addition of cage furnishings. Clarke and Jones (2000) used video screens and approach-avoidance tests to show the importance of considering outside as well as inside cage environment, to assess enrichment.

Enriched cage systems address several inadequacies of conventional cages regarding welfare and behavioral expression by providing greater space allowance, a nest area, areas for perching, and an area with litter for pecking, dust-bathing and scratching. Group sizes within modified cages range from 4 up to 60 birds, and optimal group size has not been determined (Appleby and Huges, 1995; EFSA, 2005). The enriched cage systems reduce disadvantages of traditional cages for welfare but retain most of the advantages.

Numerous studies have looked at the benefits, viability and usefulness of furnished cages. Behavioral improvements have been observed in increased normal behaviors, reduced abnormal behavior, as well as decreased aggressive behavior. In one study looking at different furnished cages and bird strains, no cannibalistic behavior or severe pecking was observed (Wall et al., 2004). Cordiner and Savory (2001) demonstrated that perches and nest boxes reduced aggressive acts by allowing subordinate hens to avoid dominant hens by day, another benefit of enriched cages.

The design of the nest box (placement, construction, appearance) can drastically affect its usage in both caged and cage-free houses (Appleby et al., 1988b; Appleby, 1990; Appleby et al., 1991; Wall et al., 2002; Struelens et al., 2005). In one study, 90.6% of eggs were laid in the nest box for one bird strain tested (Wall et al., 2004). In another study, up to 95% of eggs were laid in the nest box (Smith et al., 1993). Reed and Nicol (1992) reported 100% of eggs laid in the nest in a cage facility, with hens preferring solid rubber mat in nest floor over wire mesh. Additionally, the hens preferred nest with a small strip of artificial grass attached to the nest box wall, even though it was not available for use. Struelens at al. (2005) also demonstrated that artificial turf and peat were preferred for nest box floor over wire mesh. Management of the nest box is important. The nest box should be closed at night to prevent roosting and fouling of the nest floor. The nest box is also not used for other activities and should not take up part of space allowance (Appleby, 1990); therefore the area of the nest box is not included in the term ‘usable’ space. Sherwin and Nicol (1993a) demonstrated benefits for nesting in enriched cages, and demonstrated that age at transfer and rearing method affect number of floor eggs within enriched cages (1993b).

The design of the perch is critical for its effective usage in enriched cages (Appleby et al., 1992; Duncan et al., 1992; Appleby et al., 1998). One study reports as high as 95% of birds using perch regularly when perch space was adequate (Appleby, 1995). Another reports 60-99% of roosting at night, depending on placement and spacing, and variation in behaviors performed on the perch for placement (Duncan et al., 1992). Other studies reported 80-100% and 90-94% of birds roosting on perches (Tauson, 1984; Appleby et al., 1998). In cages with perches, reduced injurious pecking and improved bone strength (Duncan et al., 1992) have both been reported. One criticism of perches, and the rationale for one source not recommending them (Bell, 2002), is that the number of cracked eggs is higher when perches are available (Tauson, 1984). This is true because hens may lay eggs while perching (Duncan et al., 1992). However, when a properly designed nest box is available in addition to the perch, hens consistently lay in the nest box and cracked eggs are not a problem (Appleby et al., 1998). Hens showed no preference for perch construction material, which could make perches more hygienic and easier to clean (Lambe and Scott, 1998).

The litter provision requirement is the most difficult and costly to meet. Most systems provide an enclosure similar to the nest box with sand or other substrate which can have a timed opening and closing for the door to reduce eggs laid there. One alternative to the dustbath is a system that provides a small solid area (generally artificial turf) on the cage floor with a small amount of the feed dropped there by conveyor to provide an area for pecking and scratching (Savory, 2004). The birds can express foraging behavior and sham dustbathing (which may be sufficient to fulfill the desire to dustbathe). The necessity of a formal dustbath has been largely debated and the argument for its ‘necessity’ has not been

widely accepted. Lindberg and Nicol (1997) concluded that an area for sham dustbathing appears to be sufficient, though actual dustbaths may have additional welfare benefits.

Appleby et al. (1993; 2002) reported more feather and foot damage for traditional versus enriched cages. In a commercial facility, environmental control of temperature was achieved with ventilation, and feather and foot damage was improved over traditional cage houses. Mortalities reported were higher for traditional versus enriched cages (Guesdon et al., 2004).

There are still several unanswered questions about the unmet needs of hens in a furnished cage system. One study revealed that even when ample space is available in the furnished cage, some comfort behaviors such as wing flapping are not performed (Albentosa and Cooper, 2004). This is possibly because the activity is still inhibited or thwarted by the cage housing (the psychological perception or physical aversive contact with cage or penmates) or hens have little inclination to perform these activities in cages (maybe additional space and furnishings allow sufficient opportunity to express body maintenance activities).

Early exposure to furnishings is important for development of spatial cognitive abilities (Gunnarsson et al., 2000), and may affect the normal development of a behavior (Olsson et al., 2002). While early exposure may affect the development of the behavior, such as dustbathing, lack of early exposure does not remove the desire to perform the behavior (Nicol et al., 2001). Based on previous experiences with learning in hens, social learning may contribute to the development of damaging behavior as well (Nicol, 2004).

Mench et al. (1986) concluded hens neither better nor worse in cages than floor pens, based on production and physiological data. Behavioral expression varied greatly between

floor pens and all three cage configurations investigated, even when cages offered ample space for collected behaviors (1394 cm^2 (216 in^2) for 1 hen, 1394 cm^2 (216 in^2) for 2 hens, and 2788 cm^2 (432 in^2) for 2 hens). Lindberg and Nicol (1996) concluded that small groups with sufficient space is more important to hens than a large group or large space.

Hansen (1994) reported more abnormal behaviors in cages than in an aviary system for white Leghorns. Young white Leghorn layers showed more comfort behaviors and greater range of activities in an aviary system than cages, and fewer incidences of feather pecking (Tanaka and Hurnik, 1991), though rearing pullets in floor pens instead of cages may affect results. Mature white Leghorn layers had the same result as young ones; additionally, stereotypes (feather pecking, object pecking, head flicking, head bobbing, pacing) were more frequent in conventional cages than in aviary (Tanaka and Hurnik, 1992). Most aggressive acts within the cage-free houses occur in the litter area or nest areas (Oden et al., 2002). Increased aggressive pecks have been associated with decreased body weight and increased feather damage in floor pens, and larger groups (60 and 120 birds) showed most feather damage (Bilcik and Keeling, 1999). Larger group sizes (120 versus 15) and unfamiliar social environments have been shown to increase duration of tonic immobility in hens (Bilcik et al., 1998), another fear response.

Savory (1995) summarized a working discussion regarding the issue of increased feather pecking and cannibalism in colony housing systems. It has been hypothesized that feather pecking is redirected ground pecking, which is separate from aggressive pecking to determine dominance hierarchy. Provision of flooring, adequate feeder and drinker area to reduce competition, addition of perches, and provision of nest were all identified as impacting feather pecking. Placement of perches in an aviary system is important to

increase usage and minimize the risk of injury, and should be placed with no more than 45^{o} angle between horizontal perches at different heights (Scott et al., 1997). Downward jumps are most difficult for hens to complete (Moinard et al., 2004).

In a 3-year study of traditionally caged vs. aviary birds, the aviary system had several advantages over the cage system (Taylor and Hurnik, 1994). The aviary birds had better feather coverage, fewer overgrown claws, and less toe damage. However, the aviary system required a higher level of management of birds and of litter, but that higher level of management resulted in fewer sole lesions than some litter systems and fewer foot problems than traditional cages.

One review revealed that cage-free houses expose birds to higher disease risk and aggression than cages (Appleby and Hughes, 1991). Koekelbeck and Cain (1984) reported no difference in plasma cortisol of caged versus range hens, but levels were higher for floor-raised hens. This may result from balance between the stress of confinement in cages and social stressors in cage-free. Mench et al. (1986) also reported elevated plasma cortisol for cage-free hens, but this was reduced after altering the capture method for floor hens.

In a more recent on-farm assessment of commercial cage-free floor houses, Nicol et al. (2006) reported good conditions while flocks were young, but poor welfare conditions for all houses visited by the end of lay. Mortality was higher for higher stocking density (7 or 9 birds/m^2 versus 12 birds/m^2), but no other differences were observed between houses.

The additional benefits of the free-range system over cage-free barns allow for natural foraging behavior, full locomotion, and exercise. The additional risks to the hens result from predators and disease exposure from wild birds. Moberly et al. (2005) reported that British free-range flocks experience only approximately 0.5% losses to fox predation over the life of

the flock, relatively minor in comparison to other losses. Also, the group sizes are much larger than in a natural setting, creating potential problems with competition, social organization, and aggression.

One problem of free-range systems occurs due to lack of cover on many range sites. The birds may experience fear from exposure and cannot escape from aggressive behavior from other birds; therefore, frequently only a small percentage of birds use the outdoor space. One study investigated the possibility that this may be an effect of familiarity difference between inside and outside the barn (Grigor et al., 1995). Some farmers have taken the approach of offering pasture in wooded areas to provide cover or planted plots of kale on highly exposed plots. Both solutions report increased numbers of hen ranging and more time spent on range (BFREPA, 2006).

Based on the literature, extensive systems have the potential to improve quality of life for hens when managed adequately. Consequences of poor management on welfare are more severe for more extensive systems, where management of furnishings is essential (Appleby and Hughes, 1991).

Some welfare concerns are common to multiple systems. Osteoporosis is a severe welfare problem (Whitehead and Fleming, 2000; Korver, 2004; Webster, 2004) for both cage and cage-free systems. Traditional caged hens have weaker bones than those in alternative systems and are highly susceptible to breakage when handled for depopulation. However, bone breakage rates prior to depopulation may be higher in cage-free systems due to birds colliding with one another. There is little evidence to indicate osteoporosis is directly linked to loss of calcium to egg shells (Whitehead, 2004). Instead, several studies attribute osteoporosis to hens receiving inadequate nutrition and poor bone structure as an effect of

genetic selection for high egg yield. Bone breakage is not only a problem for bird welfare, but it is also a problem for meat processors who are at risk for bone contamination of meat products (Newman and Leeson, 1997). A review noted that broken bones resulted from handling and collisions, and improvements can be made by increasing bone strength and better design of housing to prevent collisions (Knowles and Wilkins, 1998). Genetic effects of bone strength reveal stronger for brown varieties than white varieties (Riczu et al., 2004).

Hens in battery cages had the weakest bones, the least movement, and the most bone breakage. The results were better for two different cage-free systems (Knowles and Broom, 1990). Furnished cages and aviaries show improved bone strength over traditional cages (Leyendecker et al., 2005). Enrichment was shown to improve fear injuries and responses during depopulation (Reed et al., 1993).

Norgaard-Nielson (1990) found that vigorous wing movements were highest in a deep-litter system, with half the observations for a modified cage system, and none for a traditional cage system. Correspondingly, humerus strength was reduced by 9% for hens in furnished cages and 45% for hens in traditional cages. Tibial breaking strength was also reduced for caged hens. Moinard et al. (1998) reported no difference in tibia strength, increased humerus strength for taller cages (40 versus 60 cm), and fewer broken bones after slaughter but increased mortality for taller cages. Fleming et al. (1994) reported that hens in conventional cages had poorest bones compared to three cage-free alternatives, and concluded that the amount of movement allowed affects bone structure.

Reports of parasite and disease prevalence in the different systems is inconsistent within the literature. This is discussed for foodborn pathogens under the *Food Safety* section.

Another potential welfare issue in all systems is competition for resources. It is important that within any system, equal resources are available to all birds because there is a strong synchronization of behaviors (Webster and Hurnik, 1994). This is especially important when considering feeder space and stocking densities. This synchronization occurs because one bird eating stimulates the other birds to eat. If a dominant hen does not allow a subordinate hen access to the feeder, the subordinate hen may not be stimulated to eat when the other hens have moved away. The subordinate hen may then be affected by nutritional deficiencies.

Feather pecking is a well-documented problem. Studies have shown genetic correlations with feather pecking, both for greater feather pecking for brown strains over white and differences within different white strains (Kjaer and Sorensen, 1997; Kjaer, 2000; Oden et al., 2002). Genetic effects of mortality of free-range hens may be related to feather pecking (Kjaer and Sorensen, 2002). Genetic variation was also identified as an important factor in feather pecking (Savory, 1995). Studies have shown potential for addition of string furnishings to reduce incidence of feather pecking (Jones et al., 2000; Jones et al., 2004; McAdie et al., 2005). Incidence of feather pecking positively correlates with corticosterone concentration, indicating a relationship between stress and feather pecking (Vestergaard et al., 1997).

Lighting is important for welfare of laying hens (Prescott et al., 2003), including visual cues for recognition among hens in small flocks or groups (D'Eath and Stone, 1999; Hauser and Huber-Eicher, 2004). Low lighting typically improved performance; but effects of low light on welfare may result in sensory deprivation for primarily visual animals (Manser, 1996).

Genetic manipulation also shows potential for addressing some of the negative aspects in the different housing systems. Potential has been demonstrated for increased production and improved behavior (reduced negative behaviors), thus increasing welfare (Muir and Craig, 1998). Genetic variation has been shown to yield significant interactions between housing methods, based on production performance; and it is important to pair appropriate strain to appropriate housing system (Lee and Craig, 1981).

Economic Considerations for Housing Systems

Cost of egg production varies by each housing system, and generally is least for traditional cages, more for modified cages and cage-free, and greatest for free-range systems. A thorough standardized comparative economic analysis for the systems discussed at commercial scale, including effects on both producer and consumer, was not found in the literature. In general, total initial investments for cage systems are higher than for cage-free systems. Enriched cage systems require the greatest investment for inclusion of furnishings, as well as more labor to manage than traditional cages. The higher cost of production results in higher cost of eggs to the consumer.

While many of the current practices with traditional cages resulted from maximizing profits with less consideration of bird welfare, there is strong evidence to show that good welfare is important for consistent profits. Studies have shown higher per-bird production for reduced stocking density (Satterlee et al., 1984; Anderson et al., 2004) and also better feed conversion, better weight gain, and lower mortality (Satterlee et al., 1984). As noted by Bell (2002) and Hann and Harvey (1971), when economic margins are low, the effects of

high densities often result in lower total profits for a farm even though the number of birds may be 33% greater.

Conflicting reports were found in the literature for production rates in the different systems. Studies report no difference in production for traditional versus enriched cages (Duncan et al., 1992; Abrahamsson et al., 1995; Appleby et al., 2002). Others report higher production for traditional cages (Walker et al., 1998). No differences were observed for production comparison of traditional versus enriched cages, but attractive nest is important to minimize the number of broken eggs in enriched cages (Guesdon et al., 2004). Wall and Tauson (2002) reported the number of broken eggs in an enriched cage system could be reduced by addition of egg-savers (simple wire devices that slow eggs as they roll from the nest into the collection cradle) and nest curtains, which may be effective when egg lay is concentrated to a small area, such as the nest box.

Discrepancies in the economic reports are likely the result of different management experience and practices. Wegner (1990) reported that after 10 years of experience with an enriched cage system (getaway cages), performance levels of the hens were similar, and production costs were approximately 5% higher for the enriched cages.

Studies report no difference in production rates for traditional cages versus cage-free barns (Tanaka and Hurnik, 1992; Taylor and Hurnik, 1996). Others report better production variable for caged layers over aviaries (Koelkebeck and Cain, 1984; Al-Awadi et al., 1995; vanHorne, 1996; Basmacioglu and Ergul, 2005). Small groups have been shown to have an economic advantage over large groups (Hann and Harvey, 1971).

Again, the discrepancies are likely attributable to experience in management. In Switzerland, 65% of hens are housed in aviaries (none in cages), which reportedly yield

production similar to traditional cages (Hane et al., 2000). In Sweden, the reported cost of production was 1.17, 1.31, and 1.34 $/kg for enriched cages, aviaries, and deep litter houses, respectively, after conversions were made using the exchange rate at the time of the report (IEC, 2005). Traditional cage production costs were not reported for Sweden (it composed less than 3% of egg production at the time), but production costs for traditional cages in the US were reported at 1.99 $/kg (higher than even the cage-free production in Sweden. However, to adequately compare these values, the economic state of each country should also be considered.

In one assessment, production costs were 8.2% greater for cage-free aviary than for traditional cages (VanHorne, 1996). In a 3-year study of traditional cages versus aviary system, there was little to no productivity difference between the systems, with regard to parameters including egg weight, egg cracking, and total hen-day production (Taylor and Hurnik, 1996). The major economic limitation of cage-free over traditional cages is the increased labor and management cost.

Nonetheless, traditional cage alternatives have proven to be profitable in Europe. One critical component to that success is the public awareness and support of attempts to improve animal welfare. The production costs for the alternative systems are higher, but consumers are willing to pay more for eggs from these systems. Some producers also take advantage of other price boosters for free-range production, including organic production (smaller flocks, fed organic feed) or Omega-3 feed enhancement. One retailer reports 50% of its egg sales are free-range eggs, and several other retailers now only sell free-range eggs and egg products (BFREPA, 2006). Not only are EU consumers willing to absorb the cost difference, but producers have become more efficient and effective at managing these

systems, which reduces their production costs and increases profits. After 16 years with cage-free systems, the Swiss cage-free systems showed only a 15% increase in production costs per egg over production costs for traditional cages (Studer, 2001). It is not known whether a majority of US consumers may also perceive value from alternate production methods.

In one review, Craig and Swanson (1994) highlighted attempts to quantify costs, but noted the lack of information for non-cage systems; on a relative scale, free-range egg was most expensive to produce, followed by floor-raised, aviary, and enriched cage. The least expensive egg to produce was from the traditional cage. Since then, much more information has been generated, but was not found to be summarized in a similar analysis.

Food Safety

Because it is not feasible to attain a housing environment with no pathogens, defensive strategies to reduce the risk are the most feasible approach. The single most important factor in controlling bacterial populations is proper ventilation to achieve proper (dry) litter and manure management (Mollinson et al., 2001).

Risks of food contamination are reduced if pathogens are not present, or low in prevalence. *Salmonella* and *Campylobacter* are documented foodborne pathogens found in laying hens (Stern et al., 2003; Messens et al., 2007). Little information regarding prevalence of *Campylobacter* for different housing systems was found in the literature for laying hens. Prevalence of *Salmonella* in different housing systems is inconsistent in the literature, and has been reported to vary with housing system, diet, season, and bird age. Cages restrict bird movements (Vits et al., 2005), which should impede transmission of

pathogen within the flock. One California study reported fewer *Salmonella enteritidis* in caged birds (1.7%) than in free-range birds (50%) with a similar pattern overall for group D *Salmonella* prevalence in caged (1.5 per 10,000) and free-range (14.9 per 10,000) hens (Kinde et al. 1996). Likewise, significantly more *Salmonella* were isolated from floor pens than from batteries of caged layer hens (Geue and Schluter, 1998). *Salmonella* prevalence in non-caged barn layers (61.5%) and free-range (54%) layers exceeded estimates for caged birds (34%) in the United Kingdom (Davies et al. 2001). Similarly, among the multiple risk factors for *Salmonella* infection in laying hens of the same age, keeping birds in a cage lowered the risk of *Salmonella* when compared to free-ranging hens (Mollenhorst et al. 2005). On the contrary, Methner et al. (2006) concluded that *Salmonella* prevalence was highest in layer hens in conventional cage systems (46.3%) and lowest in birds in free-range flocks (21.9%). Additionally, quality (egg shell thickness, egg weight, egg yolk color) and *Salmonella* contamination of eggs laid by caged hens was negatively impacted when compared to free-range birds, especially under heat stress (Barbosa-Filho et al. 2006). Further, while eggs obtained from free-range hens exhibited a lower Salmonella penetration rate (6%) than eggs from hens in conventional battery cages (16%), a number of factors, including the strain of layer hens and diet were critical (Messens et al., 2007). De Buck et al. (2003) considered pathways for *salmonella* to contaminate eggs and revealed that isthmal secretions may result in incorporation of the bacteria into the shell membrane.

The risk of contamination may be lowered for reductions in contact of eggs with fecal excretions. Cage facilities offer simple egg collection, eliminating floor eggs. Frequency of dirty eggs was no different for traditional versus enriched cages with only a perch (Tauson, 1984). More dirty eggs were reported from floor-raised system than traditional cages

(Appleby et al., 1988a). Cage- free houses also offer the opportunity for eggs laid outside automated collection areas, making it possible for eggs to remain uncollected for days.

The amount of time between egg lay and refrigeration plays an important role in shelf life. Larger farms tend to collect eggs continuously throughout the day, while smaller farms typically collect once or twice per day. Additionally, many larger farms wash, package, and refrigerate eggs on-site.

Summary of System Comparisons

The ideal laying-hen housing system should provide simplest management requirements, all necessary welfare benefits, maximum profit, and the safest product to consumers. Unfortunately, these driving forces often work against one another, making the ideal system impossible to achieve. Part of the controversy over farm animal welfare issues is the apparent conflict of interest because practices that may increase farm profitability may negatively impact welfare (for example, increased stocking density) (Estevez, 2002), and vice versa. The best housing system should create a balance among them. To assess this balance, priorities must be determined before the systems can be ranked. Herein lies the main problem, the assignment of 'importance', which will vary greatly among the company executives, farm managers, farm workers, hens, and consumers. Preference for a system will largely rely on weights of importance for the categories outlined, and these weights will depend on the parties in question (animal, producer, consumer), their previous experiences and perceptions, their understanding and education of egg production, and any moral code to which they are bound. Barnett and Newman (1997) highlight the importance of public attitudes on the success of adopting new husbandry techniques, such as enriched cages.

Rogers et al. (1989) in Edinburgh, Scotland revealed similarities and differences in perceptions of people with varying experience with agriculture and correlated their rankings to system acceptability with their ranking of priorities and importance. They revealed widespread misconceptions regarding disease risk for housing systems, even among those with agricultural experience. In a review, Savoy (2004) raises the question of the extent to which welfare standards should represent a compromise between bird welfare, practicalities, public pressure, and commercial interests.

Challenges that arise when ranking systems include accounting for variations from producer to producer. It is also difficult to quantify intangibles and quality factors such as space, even though their importance may be greater.

In this assessment, our premise was the system under discussion was properly designed and , adequately managed by properly trained workers, and efficiency and least labor-intensiveness were valuable characteristics. For the final summary table, no level of importance was assigned to any of the assessment parameters, which is likely not representative of any particular system. The result is an attempt to aid in visualization of the comparisons. The last rows of Table 1 summarize the '+' and '-' frequencies for each system. Traditional cages and free range systems yielded the most extreme rankings ('++' and '--'). There are still many unknowns and contradictions in the literature for enriched cages and cage-free barns that may affect the overall rankings.

With good management and responsible decision-making, any of the described systems may be profitable and, as observed, each has specific benefits and problems. When considering the rationale for the current EU legislation, the regulation was not simply based on science and economics. The evidence shows pros and cons for all systems but not

conclusively that one system is overall far better or worse than another. One review of the legislation stated that the ban on traditional cages was influenced more by public perception than by scientific and commercial evidence and that the ban was initiated for political reasons (Savory, 2004). As stated by several scientists, the argument over housing systems cannot be purely founded in science because questions arise that cannot be answered with a scientific study (Estevez, 2002). There is an ethical component that must be answered and dealt with accordingly. It is the intent of this paper to critically assess each system based on scientific evidence, and while it has been acknowledged, it is not the intent of this paper to assess this question of ethics.

Identified Gaps for Future Research

This review has highlighted many areas of inadequate information in the literature for comparing housing systems and anticipating consequences of altering housing schemes. Fraser (2001) criticizes the polarizing information presented by organizations on both 'sides' of the current animal welfare debates, and similarly criticize scientists for also generalizing the issues, and falling into the polarizing banter. In order to provide useful guidance, scientists must consider the issues as research problems worthy of genuine investigation and analysis.

In general, this review has revealed a number of inadequacies in our understanding of the housing systems discussed. Based upon the current state of knowledge and the comparison presented in the summary table, highlights of researchable areas include:

- Better predictions for effect of changes to a system (such as increasing space allowance on environmental control and hen physiology)

- Field comparison of environmental conditions and hen health within housing systems
- Examination of environmental control (design, operation, and effectiveness) for cage-free houses
- Examination of bird health parameter and disease prevalence in cage-free houses
- Quantification of economic comparisons at commercial production, and impacts on producers and consumers
- Development of ranking system for priorities
- Better understanding of hen perceptions and ranking of priorities (including preferences with multiple environmental factors)
- Development of a housing system scoring method
- Optimization of housing including multiple parameters in model

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Table 2.1: Comparison of advantages and disadvantages[1] of current and emerging housing systems for laying-hen production, assuming properly designed, well-managed facilities.

	Traditional Cage	Enriched Cage	Cage-Free Floor Raised	Cage-Free Aviary	Free Range
Management					
Husbandry and labor required					
-general bird observation	+	+	+	+	+
-individual bird observation	-	-	+	+	+
-bird handling	+	+	-	-	-
-egg collection	+	+	-	-	-
-feeding	+	+	+	+	+
-removal of mortalities	-	-	+	+	+
-worker training	+	-	-	-	-
Manure management					
-collection	+	+	-	-	-
-removal	-	-	+	+	+
-storage/disposal	-	-	-	-	+
-emissions	-	-	?	-	?
Environmental control					
-temperature in cold weather	+	+	?	+	-
-ammonia in cold weather	-	-	?	?	?
-temperature in hot weather	-	-	+	?	+
-ammonia in hot weather	+	+	?	?	?
-dust	?	?	?	?	?
-fly control	-	?	?	?	?
Facilities maintenance					
-equipment	-	-	+	-	+
-furnishings	+	-	+	-	+
Environmental responsibility					
-land usage	+	+	-	-	--
-land impacts	-	?	?	?	?
-air pollution	-	-	?	-	?
Hen Welfare					
Nutrition					
-clean and adequate feed	+	+	+	+	+/-
-competition for feed	??	??	?	?	?
-clean and adequate water	+	+	+	+	+/-

Table 2.1 *(cont.)*: Comparison of advantages and disadvantages[1] of current and emerging housing systems for laying-hen production.

	Traditional Cage	Enriched Cage	Cage-Free Floor Raised	Cage-Free Aviary	Free Range
Environment					
-space, access	-	-	+	+	+
-space, quality	-	+	+	+	+
-temperature	+	+	?	+	?
-fresh air	+	+	+/-	?	+/-
Health					
-control of parasites	?	?	?	?	?
-control of disease	??	?	??	??	??
-foot problems	-	+	?	?	+/-
-osteoporosis	-	+	+	+	+
-broken bones	-	+	-	-	+
-injuries	?	?	-	-	+
Behavior					
-standing, sitting	+	+	+	+	+
-locomotion	-	-	+	+	++
-eating, drinking	+	+	+	+	+
-scratching	-	+	+	+	++
-pecking	-	+	+	+	++
-foraging	-	-	-	-	++
-perching	-	+	+	+	+
-nesting	-	+	+	+	+
-abnormal behaviors	-	+	+	+	++
Fear and distress					
-contact with other birds	?	?	?	?	?
-group size	+	+	-	-	-
-instances of aggression	+	+	-	-	-
-ability to escape aggression	-	-	+	+	+
Consequences of poor management	+	-	--	--	--
Economics					
Investment per bird	-	--	+	-	+
Production costs to producer	++	-	-	-	--
Bird productivity	+	+	??	??	--
Product cost to consumer	++	+	-	-	--
Consumer value	?	?	?	?	?

Table 2.1 *(cont.)*: Comparison of advantages and disadvantages[1] of current and emerging housing systems for laying-hen production.

	Traditional Cage	Enriched Cage	Cage-Free Floor Raised	Cage-Free Aviary	Free Range
Food Safety					
Cleaner eggs	+	+	-	-	-
Egg non-contact with feces	+	+	-	+/-	-
Presence of foodborn pathogens in flock	?/??	?/??	?/??	?/??	?/??
Time of egg lay to refrigeration	+	+	-	+	-
TOTAL, frequency of notation					
'++'	2	0	0	0	4
'+'	23	30	25	23	24
'-'	23	17	17	20	14
'--'	2	1	1	1	5
'?'	6	9	15	13	13
'??'	3	2	3	3	2
SCORE[2], points	0	11	6	1	8

[1]Assessment notations (and score values)
++ advantage (2 pt) + positive (1 pt) - negative (-1 pt) -- disadvantage (-2 pt)
?? literature not consistent (0 pt) ? absent from literature (0 pt)
[2]Score based on frequency of notation and assignment of points as indicated in footnote 1

Figure 2.1: A traditional cage system with manure belt beneath cages for waste removal. (Photo source, A.R. Green, author)

Figure 2.2: Modified cage system including nest box, perch, and dustbath, for housing groups of 8 birds/cage (Photo source: EFSA, 2005)

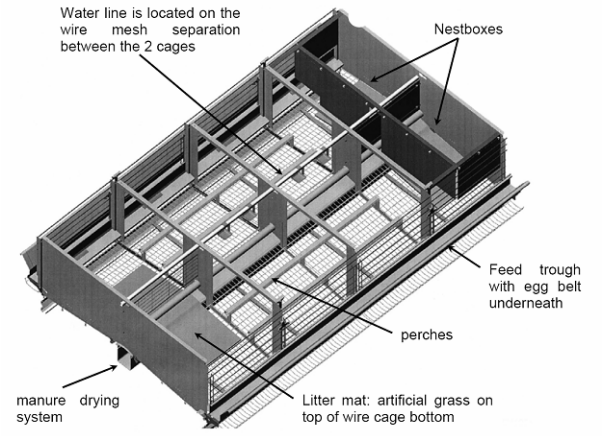

Figure 2.3: Modified cage system including nest box, perch, and litter mat, for housing groups of approximately 18 birds/cage (2 cages shown). (Photo source: EFSA, 2005)

Figure 2.4: Deep-litter system with hens raised on partially littered floor (Photo source: A.R. Green, author)

Figure 2.5: Aviary system with access to multiple levels. (Photo source: Studer, 2001)

Figure 2.6: Free-range hens have the option to spend partial time outdoors daily. (Photo source: BFREPA, 2008)

Chapter 3

Field Evaluation of Air Quality and Bird Health Status in Three Types of Commercial Egg Layer Houses

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Abstract. *In this field observational study, three types of laying-hen houses, i.e., high-rise (HR), manure-belt (MB), and cage-free floor-raised (FR), were monitored for air temperature, relative humidity, carbon dioxide (CO_2), and atmospheric ammonia (NH_3) during winter and summer conditions in Iowa. Under winter conditions, the HR and MB houses had more comfortable temperature and NH_3 levels than the FR houses where NH_3 level reached 85-89 ppm and house temperature varied more with outside conditions. Under summer conditions, house temperature showed the least rise above ambient in the FR houses, and NH_3 level was similar for all housing types. Examination of the hen health status revealed differences in pathogen frequency between housing systems for winter and summer, but not conclusively in favor of one system over another. The results of this study indicate that the benefits of each system were season-dependent. Further monitoring of the environment, bird health and production performance over an extended period (e.g., one year) to quantify the benefits and limitations of each system is warranted. Information of this nature will aid in optimization of hen housing systems for enhanced bird welfare and sustained production efficiency for the egg industry.*

Keywords: Ammonia, temperature, *Campylobacter*, *Salmonella*, high-rise, manure-belt, cage free floor-raised

Introduction

Animal welfare is an increasing issue of concern for the egg industry. Housing systems play a critical role in welfare of laying hens, and various systems are implemented throughout the world. A segment of the U.S. egg industry has begun modifying housing systems from conventional cages to alternative (non-caged) systems, although this trend is more prevalent in Europe. Behavioral benefits of cage-free systems are well documented, as are disadvantages (van Emous and Fikls-van Niekerk, 2004; Vits et al., 2005). Caged systems offer opportunities for better management, reduced production costs and more efficient use of resources. Important considerations for welfare also include environmental conditions (including air quality) and hen health, but these parameters are not well documented for different laying-hen housing systems.

Different housing systems create unique management scenarios, and may result in different microclimates for the same weather. Environmental temperatures not only influence hen comfort and performance, but affect other environmental parameters, such as ammonia and dust levels in poultry houses (Carlile, 1984). Ammonia emissions from layer houses have been shown to differ considerably among high-rise, manure-belt, and cage-free systems (Koerkamp and Bleijenberg, 1998; Liang et al., 2005). Ample literature has documented the adverse effects of elevated atmospheric ammonia levels on poultry, e.g., reduced production performance and poor health of broilers (Charles and Payne, 1966a; Deaton et al., 1984; Miles et al., 2004; Miles et al., 2006), reduced egg production (Charles and Payne, 1966b),

damaged respiratory tract (Nagaraja et al., 1983; Al-Mashhadani and Beck, 1985), increased susceptibility to Newcastle Disease Virus (Anderson et al., 1964), increased incidence of air sacculitis (Oyetunde et al., 1978) and keratoconjunctivitis (blind eye) (Faddoul and Ringrose, 1950), and prevalence of *Mycoplasma gallisepticum* (Sato et al., 1973). Egg quality may also be adversely affected by high levels of atmospheric ammonia as measured by reduced albumen height, elevated albumen pH, and albumen liquefaction (Cotterill and Nordsog, 1954). To ensure good bird health and performance, it is recommended that atmospheric ammonia in poultry houses not exceed 25 ppm (UEP, 2006), which may be difficult to achieve in some housing types in winter. During summer it may be problematic for houses with high numbers of birds to provide sufficient ventilation to maintain comfortable temperatures, even at maximum ventilation rates.

Health concerns impact not only the welfare of the birds, but also the microbial food safety of the consumer. Consumption of contaminated poultry is a major risk factor for human infections with *Salmonella* and *Campylobacter* (Altekruse and Tollefson, 2003). *Campylobacte*r spp, which is a major cause of bacterial enteritis worldwide, cause nearly 2 million cases of foodborne illness, 10,000 hospitalizations and 100 deaths annually (Mead et al., 1999). *Salmonella* causes an estimated 1,343,000 cases of foodborne illness, 15,000 hospitalizations and 500 deaths each year (Mead et al., 1999). *Salmonella, C. jejuni* and *C. coli* frequently colonize clinically healthy live birds and are present in retail purchased poultry. In addition, *Salmonella*-contaminated eggs are a vehicle of transmission to humans. The ability of *Campylobacter* to laterally transfer genes encoding antimicrobial resistance to other bacteria in the avian intestine are of public health concern. To reduce human foodborne

illness, on-farm pathogen reduction strategies strive to deliver poultry, meat, and eggs to the American consumer free of *Campylobacter* and *Salmonella*.

Epidemiological studies indicate that the prevalence of either *Salmonella* or *Campylobacter* varies with housing system, diet, season and age of birds (Avrain et al., 2003; Bailey and Cosby, 2005; Heuer et al., 2001; Huneau-Salaun et al., 2007; Tresierra-Ayala et al., 1995; Wittwer et al., 2005). A California study reported fewer *Salmonella enteritidis* in caged birds (1.7%) than in free-range birds (50%) with a similar pattern for other group D *Salmonella* in caged (1.5 per 10,000) and free-range (14.9 per 10,000) hens (Kinde et al., 1996). Likewise, significantly more *Salmonella* were isolated from floor pens than from batteries of caged layer hens (Geue and Schluter, 1998). *Salmonella* prevalence in non-caged barn layers (61.5%) and free range (54%) layers exceeded estimates for caged birds (34%) in the United Kingdom (Davies and Breslin, 2001). Similarly, among the multiple risk factors for *Salmonella* infection in laying hens of the same age, confining birds to a cage lowered the risk of *Salmonella* when compared to free-ranging hens (Mollenhorst et al., 2005). In contrast, others have reported that *Salmonella* prevalence was highest in laying hens housed in conventional cage systems (46.3%) and lowest in free-range flocks (21.9%) (Methner et al., 2006). Still others report no significant differences in *Salmonella* status when free versus caged layers were evaluated (Posadas-Hernandez et al., 2005). No studies have compared the *Campylobacter* prevalence in layers maintained in different housing systems.

To fully assess the welfare of birds in a specific system, it is important to evaluate the system as a whole, including aspects of health, environment, behavior, handling and management practices, worker education and training, and economics. Few studies compare air quality at bird level in high-rise caged (HR), manure-belt caged (MB), and cage-free

littered floor raised (FR) laying-hen facilities. Information regarding hen health status and prevalence of foodborne pathogens in these housing systems yields conflicting reports.

Therefore, the objective of this field research was to monitor the air quality and hen health status in three types of housing systems – HR, MB, and FR for both warm and cold climatic conditions in Iowa. This paper summarizes the results of this monitoring that may be used by decision makers to improve laying hen husbandry.

Materials and Methods

Description of the Layer Houses Monitored

Four houses from each of the three hen housing systems (HR, MB, and FR) were selected based on farm access and availability. The characteristics of the houses are described below and summarized in Table 1.

The four FR houses, located at three separate sites (Site 1, 2, 3) within 16 km (10 mile) of one another, featured floors that were partially or fully available to the hens and covered with litter. All FR houses were equipped with automated feeding, watering, and egg collection and nest boxes for the hens. Hens in one house produced organic eggs and were allowed daily access to pasture under suitable weather conditions. Two houses had a partially slatted floor located along the center of the house and manure accumulated beneath the slatted floor was periodically removed. Three houses were naturally ventilated, and one was mechanically ventilated. Three houses had an east-west orientation, and one had a north-south orientation. The MB houses monitored were located at one commercial egg-production site (Site 4, Table 1). Manure was removed daily. The HR houses monitored were located at one commercial egg-production site (Site 5, Table 1). Manure was scraped from the

dropping board into the lower-level storage four times daily. Manure remained in the house for about a year before a complete clean-out.

Monitoring of Environmental Conditions

Environmental variables measured near bird level included: ammonia (NH_3), carbon dioxide (CO_2), air temperature (T), and relative humidity (RH). Each house was monitored continuously over a 20 to 24 h period in winter and summer. All 12 houses in the study contained adult laying hens of various ages, but hens within a house were of the same age (Table 1). Ammonia and CO_2 concentrations inside the barns were measured at 30 min intervals using portable monitoring units (PMUs) previously developed for monitoring poultry building ammonia emissions (Xin et al., 2002). A three-location composite air sample across the width of the house and near 1/3 into the length of the house was taken for the air sampling (Figures 1 & 2). Air temperature and RH of both inside and outside the barns were recorded at 5 min intervals using programmable, portable temperature and relative humidity (T/RH) loggers (H08-032-08, Hobo Pro, Onset Computer Company, www.onsetcomp.com). One T/RH logger was placed at each sampling port. For caged houses, an additional T/RH logger was placed in the cage aisle near each sampling port (approx. 1.5m or 5ft distance).

Examination of Hen Health Status

Ten birds were randomly selected from each house on the day of monitoring for assessment of health status, tracheal condition and prevalence of *Campylobacter* and *Salmonella*. Blood samples were taken from each hen and sera from these samples were

subsequently tested for the presence of antibodies against *Mycoplasma gallisepticum* and *Mycoplasma synoviae* by the serum plate agglutination test. Birds were euthanized via injection of sodium pentobarbital, and trachea, small intestine, and ceca samples were collected.

Tracheal Analysis. Tracheas were fixed in 10% neutral buffered formalin, dehydrated in a graded series of ethanol, and embedded in paraffin. Sections were cut (4 microns in thickness) and stained with hematoxylin and eosin for examination by light microscopy.

Intestinal Homogenates. Ceca and small intestine were collected, refrigerated (4^{o}C) and a 10% (wt/vol) homogenate prepared in buffered peptone waster as previously described (Wesley et al., 2005).

Detection and Identification of Campylobacter spp. Presumptive *Campylobacter* isolates were confirmed and speciated as *C. coli* or *C. jejuni* by PCR (Polymerase Chain Reaction) as previously described (Wesley et al., 2005).

Detection and Identification of Salmonella. The buffered peptone water homogenate (10% wt/vol) was incubated (24 h, 37^{o}C) aerobically. Following incubation, 1 ml of the enrichment was transferred to 10 ml Tetrathionate Hajna broth (Becton Dickson, Sparks MD) and incubated (24 h, 42^{o}C) aerobically.

Data Analysis and Presentation

For environmental conditions, data were summarized for each house and combined into mean plots for each variable during each monitoring period. To describe the combined effects of T and RH under warm conditions, Temperature Humidity Index (THI) was calculated using the relationship $THI=0.6T_{db}+0.4T_{wb}$, where T_{db} = dry-bulb temperature and

T_{wb} = wet-bulb temperature (Zulovich and DeShazer, 1987). For the health status data, two-factor repeated measures analyses were used in two different comparisons between Table 5 and Table 6 prevalence of *Campylobacter* data. The first comparison examined differences among birds under the three housing schemes over two trials (winter and summer) using four replicates. The second comparison examined differences between caged and non-caged birds over winter and summer with an unequal number of replicates. Differences were considered statistically significant at $P < 0.05$.

Results and Discussion

Bird-Level Environmental Conditions

Environmental conditions differed for all three housing types. There was greater variability from house to house for the FR system flocks, which were independently-operated sites with different house configurations and flock management. Variability was less for houses located on the same site and operated under the same management, as was the case for the MB and HR houses. House ventilation systems differed, explaining some of the observed variation in environmental conditions. Additionally, the FR houses provide only one level of birds with 3 to 5 times more space per bird than the cage facilities, resulting in much less heat production for the system as well as lower CO_2 concentrations.

Winter. The 24 h mean, maximum and minimum values of each variable for each housing system in winter are summarized in Table 2 and depicted in Figure 3. Temperatures and NH_3 levels remained within comfortable or recommended ranges during the entire monitoring period for the cage (HR and MB) systems. In comparison, NH_3 concentrations substantially exceeded the recommended level of 25 ppm for laying hens for the FR system,

with a daily mean of 46 ppm, as compared to 14 ppm for HR and 7 ppm for MB. The maximum concentration in the FR houses reached 85-89 ppm. Temperatures in the FR houses tended to fluctuate with the outside conditions. The temperature at bird level was considerably cooler in the FR houses than in the cage houses, averaging 15.5 (± 1.5) ^{o}C vs. 20.6 (± 0.8) ^{o}C for HR houses and 24.6 (± 1.0) ^{o}C for MB houses. The lower potential for heat production by the birds in FR houses contributes to the cooler temperatures. Interestingly, CO_2 concentrations tended to be lower in FR houses (mean ± SE of 2021 ± 199 ppm) than in the HR (2433 ± 95 ppm) or MB (3072 ± 36 ppm) systems, presumably a result of lower bird density and thus less CO_2 generated from bird respiration in the FR houses.

Animal welfare standards promoted by the United Egg Producers state that ammonia levels in chicken houses should ideally be less than 10 ppm and should not exceed 25 ppm (UEP, 2006). Studies have shown that laying hens find atmospheric ammonia highly aversive at concentrations of 25 ppm (Kristensen et al., 2000). Air quality for the humans working in poultry houses is also a concern. The National Institute for Occupational Safety and Health (NIOSH) has established a limit of 25 ppm ammonia, time weighted average (TWA) over 8 hours for humans (NIOSH, 2005). The Occupational Safety and Health Administration (OSHA) permissible exposure limit for humans is 50 ppm ammonia TWA over 8 hours (OSHA, 2002).

Frequent (daily, in this case) removal of manure in the MB houses greatly reduced ammonia concentrations. This result was consistent with those previously reported (Liang et al., 2005).

Simple operating adjustments could have improved the conditions in the naturally ventilated FR houses. For these FR houses, addition and operation of minimum ventilation

fans could have significantly reduced NH_3 concentration during the night when side curtains were closed. Litter management likely had a significant impact on NH_3 generation, with drier litter lessening NH_3 volatilization. A thin layer of wood shavings was periodically spread over the litter in house FR3, which subsequently had lower levels of NH_3 in winter, even at night when the curtains were closed. During winter, FR3 had the best air quality, which was likely not a function of its orientation. Instead, ventilation of house FR3 was likely enhanced by the chimneys located longitudinally along the center of the house.

Summer. The 24 h mean, maximum and minimum values of each variable in summer are summarized in Table 3 and depicted in Figure 4. Maximum NH_3 concentrations were within the recommended level (25 ppm) for all houses, with the exception of FR3 (42 ppm) and FR4 (29 ppm). All daily mean NH_3 levels were below 25 ppm. Temperatures in the FR houses showed less rise above the ambient than the cage houses (average rise or percent rise with respect to ambient: 0.3^{o}C or 1% for FR, 1.2^{o}C or 4% for HR, and 4.7^{o}C or 18% for MB). Temperature Humidity Index also showed less rise above ambient for cage free versus cage houses, and HR houses had the greatest THI rise above ambient. The reduced bird density in FR houses created an advantage here for temperature control in warm temperatures.

For conditions in Iowa, orientation of the houses for natural ventilation (E-W) is critical in summer months when wind drives the air exchange. House FR3 was oriented N-S and had the poorest air quality during the summer study period.

The tunnel ventilation used in the MB houses in this case needs to be configured properly; namely, the eave inlet dampers must be properly adjusted to achieve the relatively uniform air distribution along the length of the building. Some ventilation dead spots were

noted in the MB houses during the summer, leading to poor air quality at these locations. However, it is uncertain if these stagnant areas were reflected in the measurements. Even so, the temperature distribution was more uniform in the MB houses than in the HR houses, particularly during summer.

Temperature Stratification between Cages and Aisles

Figures 5 and 6 display the differences in T and RH between the aisle and the cage interior. Air temperature tended to be higher inside the cages than in the aisle during both winter and summer, especially for the MB houses. As expected, the differences were more apparent in winter than in summer due to lower ventilation rate in winter. The magnitude of the differences tended to be smaller in the HR houses than in the MB houses, even though the differences fluctuated more in the HR houses.

Temperature differences between cage interior and aisles likely resulted from several factors. The main factor was likely that air movement was impeded by cage fixtures and the presence of birds. Additionally, birds contribute heat to their microenvironment that would not be detected by a thermostat located in the aisle. Because the cage temperature was monitored inside an adjacent empty cage, the differences in microclimate experienced by the birds may have been even greater than measured. This outcome suggests that it may be prudent to periodically monitor the cage interior temperature, and adjust the temperature setpoint, when necessary, to reflect the microenvironment that the birds are experiencing. Alternatively, consider locating the thermostat temperature sensors near the bird microenvironment.

Hen Health Status

Tracheal Analysis. Antibodies against *Mycoplasma synoviae* (MS) and/or *Mycoplasma gallisepticum* (MG) were detected in sera from all hens except in samples from house FR2 (winter) and houses FR1, FR2, and FR4 (summer) (Table 4). Microscopic examination of hen tracheas revealed abnormally high numbers of lymphocytes within the lamina propria layer of the tracheal wall in birds from all houses except from house FR2. Intact cilia were present on the respiratory surface of all birds from all houses, and no eye lesions were observed.

The presence of antibodies against *Mycoplasma synoviae* (MS) or *Mycoplasma gallisepticum* (MG) in the sera of chickens indicates that flocks were infected with these pathogens. Although not observed in this study, infection with *Mycoplasma* sp. typically results in damage to cilia on the mucosal surface of the trachea and increased the susceptibility of infected chickens to inhaled dust-borne pathogens. The immune response of hens to the presence of avian mycoplasmas colonizing respiratory epithelium of the trachea is manifested by the accumulation of lymphocytes within the underlying lamina propria. Hens in FR2 were not infected by MG or MS, did not mount an immune response, and consequently did not have significant numbers of lymphocytes in the tracheal wall during winter or summer. Because most hens in this study were infected with *Mycoplasma*, microscopic changes observed in the tracheas could not be distinguished from changes that might have resulted from exposure to ammonia or particulate matter in the air.

Intestinal Homogenates. Campylobacter and *Salmonella* were detected in winter (Table 5) and summer (Table 6). For winter conditions (Table 5), *Campylobacter* spp. prevalence was higher in FR than in HR houses (80.0% vs. 37.5%, $P<0.05$), but there was no

difference in overall *Campylobacter* spp. prevalence between FR hens (80.0%) and MB hens (62.0%). The prevalence of *C. coli* was higher in FR hens than HR or MB hens (55.0% vs. 25.0% or 25.6%, respectively, $P < 0.05$). No differences were detected when *Salmonella* prevalence was correlated with housing systems. Prevalence numbers were too low to perform χ^2 tests for birds dually infected with *C. jejuni/C.coli*. For summer conditions (Table 6), results from bacteriological isolation of *Campylobacter* showed lower prevalence of *Campylobacter* and *C. jejuni* for FR hens and HR hens than for MB hens (27.5% and 20.0% vs. 65.0%; and 7.5% and 20.0% vs. 52.5%, respectively, $P < 0.01$). When winter and summer data based on bacteriological isolation are compared, the prevalence of *Campylobacter* spp. in the FR birds was higher in winter than in summer (80.0% vs. 27.5%, $P < 0.05$).

Monitoring for bacterial foodborne pathogens showed seasonal differences between the housing systems. Factors contributing to the higher prevalence of *Campylobacter* spp., specifically *C. coli,* in winter for the FR birds may include: more direct contact with manure which facilitates fecal-oral transmission of enteric pathogens, different breeds of laying hens used in the FR houses, and bird housing densities. Most significantly, during periods of inclement weather in the winter months, the FR birds were confined indoors which facilitates fecal-oral transmission of *Campylobacter* within the flock. In contrast, for the summer monitoring, the prevalence of *Campylobacter* spp. and *C. jejuni* was significantly lower in FR and MB birds when compared to the HR hens ($P<0.01$).

Observational Nature of This Study

Results from this study should be regarded as observational only. Because the monitoring was conducted at a system level, results could not be interpreted to specifically discern the

source(s) of differences. It also should be acknowledged that data from 24 h environmental monitoring would not be sufficient to yield concrete conclusions about different housing types. Nevertheless, the data confirmed that under similar weather patterns, different environmental conditions would exist for different housing systems and different management schemes. Also, the results indicate seasonal differences among housing systems for prevalence of bacterial foodborne pathogens, but the results do not conclusively show that one system yields lower pathogen frequencies than another, as reported in the Netherlands (van Emous and Fikls-van Niekerk, 2004). Further studies should include multiple representations of each housing type, encompassing different management and housing configurations to better delineate the cause-effect relationships. Future studies should also consider collecting environmental, physiological, and production data collected periodically over an extended period of time (e.g., one year).

Conclusions and Applications

- Observational data to assess environmental conditions (T, RH, CO_2, and NH_3) and bird health status in winter and summer were collected for three types of laying-hen housing in Iowa: a) cage-free floor-raised (FR), b) caged high-rise (HR), and c) caged manure-belt (MB).
- Differences in environmental conditions and/or pathogen frequency were observed among all three housing types during summer and winter conditions. During winter, NH_3 levels were much higher in the FR housing systems than in HR or MB systems. Air temperature in the FR houses also fluctuated more, following the outside temperature.

- Results of this study were unable to identify the specific sources of benefits associated with each system because all houses were different in some aspect, and were operated under different management practices.
- Differences observed in the air quality and pathogen frequency merit further research to quantify and identify sources of these differences.
- It may be prudent to periodically monitor the cage interior temperature, and adjust the temperature setpoint, when necessary, to reflect the microenvironment that the birds are experiencing. Alternatively, consideration should be given to locating the thermostat temperature sensors near the bird microenvironment.

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Table 3.1: Description of the laying hen houses monitored

		Floor-Raised				Manure belt					High-rise				
	House	FR1	FR2	FR3	FR4	MB1	MB2	MB3	MB4	MB5	HR1	HR2	HR3	HR4	HR5
	Site	1	1	2	3	4	4	4	4	4	5	5	5	5	5
	Ventilation	Natural	Mechanical, side inlets and fans	Natural, with chimneys	Natural	Mechanical, tunnel with lengthwise inlet					Mechanical, ceiling inlet with side fans in manure storage area				
	Orientation	E-W	E-W	N-S	E-W	E-W	E-W	E-W	E-W	E-W	E-W	E-W	E-W	E-W	E-W
	Manure management	Litter, wood shavings added once at start of flock	Litter, wood shavings added once at start of flock, partial slatted floor with auger removal	Litter, sawdust added every 2 weeks, partial slatted floor with auger removal	Litter, wood shavings added once at start of flock	Removed daily					Removed between flocks				
WINTER 2006	Date monitored	Jan 10-11	Jan 10-11	Jan 10-11	Jan 10-11	Jan 16-17	Jan 16-17	Jan 16-17	Jan 16-17	DNM[1]	Jan 21-22	Jan 21-22	Jan 21-22	Jan 21-22	DNM[1]
	Bird age	76 weeks	36 weeks	32 weeks	53 weeks	39 weeks	98 weeks	45 weeks	109 weeks	DNM[1]	42 weeks	46 weeks	93 weeks	142 weeks	DNM[1]
	Flock size (initial)	3500	6000	8700	10,000	104,500	106,400	106,400	93,200	DNM[1]	66,061	65,141	64,727	80,174	DNM[1]
SUMMER 2006	Date monitored	Aug 7-8	Aug 7-8	Aug 7-8	Aug 7-8	DNM[1]	DNM[1]	Aug 1-2	Aug 1-2	Aug 1-2	DNM[1]	Jul 24-25	Jul 24-25	Jul 24-25	Jul 24-25
	Bird age	43 weeks	67 weeks	63 weeks	36 weeks	DNM[1]	DNM[1]	76 weeks	32 weeks	50 weeks	DNM[1]	99 weeks	22 weeks	72 weeks	39 weeks
	Flock size (initial)	3500	6000	8700	10,000	DNM[1]	DNM[1]	106,400	106,400	106,400	DNM[1]	65,141	63,006	73,600	66,061
	House dimensions	40 x 160 ft 12 x 49 m	50 x 210 ft 15 x 64 m	40 x 300 ft 12 x 91 m	66 x 180 ft 20 x 55 m	60 x 520 ft 18 x 158 m	60 x 520 ft 18 x 158 m	60 x 520 ft 18 x 158 m	60 x 520 ft 18 x 158 m	60 x 520 ft 18 x 158 m	48 x 430 ft 15 x 131 m	48 x 430 ft 15 x 131 m	48 x 430 ft 15 x 131 m	48 x 430 ft 15 x 131 m	48 x 430 ft 15 x 131 m
	Bird housing	Cage-free, free range organic	Cage-free	Cage-free	Cage-free, Omega-3 diet	Caged	Caged	Caged	Caged	Caged	Caged,	Caged	Caged	Caged	Caged
	Birds per group (W/S[2])	3500	6000	8700	10,000	10	6	6	6	6	8	8	9/8	6/5	8
	Breed	Brown (U[3])	Brown (U[3])	Brown (U[3])	Brown (U[3])	W-36	W-36	W-36	W-36	W-36	W-36	W-36	W-36	W-36	W-36
	Space allowance, in[2]/bird (W/S[2]) cm[2]/bird (W/S[2])	263 + dpa[4] 1698	252 1626	199 1284	171 1103	54 348	54 348	54 348	54 348	54 348	59 381	59 381	56/61 361/394	56/61 361/394	59 381
	Water treatment	peroxide	peroxide	peroxide	none, well-water	ozonated	ozonated	ozonated	ozonated	ozonated	none	none	none	none	none

[1]DNM = did not monitor
[2]W/S = winter/summer
[3]U=unknown, breed not documented
[4]dpa=daily pasture access

Table 3.2: Winter conditions: 24 h mean, maximum and minimum values for each laying-hen house and resulting overall mean and standard error (SE) for each type of housing system.

24-h Means	**Floor-Raised**						**Manure Belt**						**High-Rise**					
	FR1	FR2	FR3	FR4	**Mean**	SE	MB1	MB2	MB3	MB4	**Mean**	SE	HR1	HR2	HR3	HR4	**Mean**	SE
NH3 Mean	59	57	20	50	**46**	9	6	8	7	6	**7**	0	8	10	20	17	**14**	3
Max	85	86	30	89	**72**	14	9	10	10	9	**9**	0	11	14	26	24	**18**	4
Min	45	46	3	20	**28**	10	4	6	6	5	**5**	0	7	9	16	8	**10**	2
CO2 Mean	2150	2376	1451	2108	**2021**	199	3122	2987	3037	3142	**3072**	36	2455	2260	2691	2326	**2433**	95
Max	2713	3159	2161	4261	**3073**	445	3986	3434	3469	3885	**3694**	141	2643	2678	2953	2643	**2729**	75
Min	1369	2091	884	919	**1316**	281	2507	2472	2713	2643	**2583**	57	2091	2056	2437	2056	**2160**	93
T Mean	16.8	18.6	11.4	15.3	**15.5**	1.5	27.1	25.1	23.8	22.6	**24.6**	1.0	22.8	18.8	20.2	20.6	**20.6**	0.8
Max	17.8	19.5	14.9	20.7	**18.2**	1.3	28.3	26.5	25.3	23.8	**26.0**	1.0	24.7	19.3	21.1	21.3	**21.6**	1.1
Min	14.8	17.5	8.2	9.4	**12.5**	2.2	24.9	23.1	22.3	21.7	**23.0**	0.7	20.4	18.3	18.8	19.8	**19.3**	0.5
RH Mean	69	64	66	62	**65**	1	36	37	47	41	**40**	2	41	51	56	50	**50**	3
Max	72	79	72	69	**73**	2	44	41	54	46	**46**	3	45	56	62	64	**57**	4
Min	63	59	59	55	**59**	2	29	33	40	37	**34**	2	37	49	52	42	**45**	3
THI Mean	14	13	7	10	**11**	2	15	14	15	13	**14**	1	13	11	13	13	**12**	0
Max	16	16	10	15	**14**	1	17	15	16	14	**16**	1	14	12	15	16	**14**	1
Min	11	12	4	5	**8**	2	13	12	13	11	**13**	1	11	11	13	10	**11**	1
Ambient T Mean	11.9	13.4	8.0	11.6	**11.2**	1.1					**2.2**						**-0.1**	
Max	14.6	16.5	10.7	14.8	**14.1**	1.2					**21.3**						**1.4**	
Min	10.2	12.3	6.1	7.1	**8.9**	1.4					**-4.7**						**-1.2**	
Ambient RH Mean	71	69	70	67	**69**	1					**92**						**89**	
Max	76	83	76	71	**77**	2					**100**						**93**	
Min	60	64	60	59	**61**	1					**51**						**82**	
Ambient THI Mean	8	9	5	8	**7**	1					**0**						**-1**	
Max	9	12	6	11	**10**	1					**4**						**0**	
Min	7	8	3	4	**5**	1					**-4**						**-2**	
T Rise Above Ambient	4.8	5.2	3.4	3.7	**4.3**	0.4	24.9	22.9	21.6	20.4	**22.5**	1.0	22.9	18.8	20.2	20.6	**20.6**	0.8

[1]NH_3=ammonia, CO_2=carbon dioxide, T=temperature, RH=relative humidity, Amb = ambient
[2]FR=floor-raised, MB=manure-belt, HR=high-rise

Table 3.3: Summer conditions: 24 h mean, maximum and minimum values for each house and resulting overall mean and standard error (SE) for each type of housing system.

24-h Means	**Floor-Raised**						**Manure Belt**					**High-Rise**					
	FR1	FR2	FR3	FR4	**Mean**	SE	MB3	MB4	MB5	**Mean**	SE	HR2	HR3	HR4	HR5	**Mean**	SE
NH3 Mean	3	3	14	15	**9**	3	2	8	5	**5**	2	3	2	3	4	**3**	1
Max	6	6	42	29	**21**	9	4	14	7	**8**	3	5	3	7	8	**6**	1
Min	0	1	3	5	**2**	1	0	3	3	**2**	1	3	0	2	3	**2**	1
CO2 Mean	451	406	631	641	**532**	61	853	1043	1140	**1012**	73	541	442	475	621	**520**	40
Max	643	578	1059	1059	**835**	130	1059	1264	1435	**1253**	94	678	608	643	884	**703**	62
Min	368	333	368	438	**376**	22	643	884	884	**804**	70	473	368	403	508	**438**	32
T Mean	24.0	25.1	25.2	25.5	**25.0**	0.3	30.0	31.0	30.3	**30.4**	0.3	30.1	28.8	28.3	28.7	**28.9**	0.4
Max	28.5	30.3	30.1	30.0	**29.7**	0.4	32.1	32.8	32.1	**32.3**	0.2	33.8	33.3	34.9	33.4	**33.9**	0.4
Min	21.3	22.4	21.9	22.8	**22.1**	0.3	27.4	28.4	28.0	**27.9**	0.3	25.9	24.1	24.3	23.9	**24.6**	0.5
RH Mean	66	61	62	62	**63**	1	73	71	71	**72**	1	46	47	53	52	**50**	2
Max	76	70	70	70	**71**	2	78	78	77	**78**	0	54	57	63	63	**59**	2
Min	50	42	46	46	**46**	2	66	64	65	**65**	1	35	37	37	39	**37**	1
THI Mean	18	18	19	19	**19**	0	25	26	25	**26**	0	20	19	20	20	**19**	0
Max	20	20	22	23	**21**	1	28	29	28	**28**	0	23	22	23	22	**22**	0
Min	17	17	17	18	**17**	0	23	24	23	**23**	0	18	17	18	18	**18**	0
Ambient T Mean	24.0	25.0	24.8	24.7	**24.6**	0.2				25.7						27.7	
Max	30.0	32.3	29.8	30.2	**30.6**	0.6				32.3						33.4	
Min	20.8	21.8	21.9	22.0	**21.6**	0.3				21.3						21.5	
Ambient RH Mean	66	60	67	65	**64**	2				94						48	
Max	77	71	76	73	**74**	1				100						64	
Min	46	37	48	45	**44**	2				62						31	
Ambient THI Mean	18	18	19	19	**19**	0				**25**						**18**	
Max	20	20	22	23	**21**	1				**27**						**21**	
Min	17	17	18	18	**17**	0				**22**						**16**	
T Rise Above Ambient	0.0	0.1	0.4	0.8	**0.3**	0.2	4.3	5.3	4.6	**4.7**	0.3	2.4	1.1	0.6	1.0	**1.2**	0.4

[1]NH_3=ammonia, CO_2=carbon dioxide, T=temperature, RH=relative humidity, THI=temperature humidity index, Amb = ambient
[2]FR=floor-raised, MB=manure-belt, HR=high-rise

Table 3.4: *Mycoplasma synoviae* (MS) or *Mycoplasma gallisepticum* (MG) serology results (percent of birds (n=10) testing positive) from laying hens in three different housing systems.

Season		Floor-Raised				Manure-Belt					High-Rise				
	(% positive)	FR1	FR2	FR3	FR4	MB1	MB2	MB3	MB4	MB 5	HR1	HR2	HR3	HR4	HR 5
Winter	**MS**	0	0	100	100	100	100	100	100	DNM[1]	100	100	100	100	DNM[1]
	MG	100	0	0	0	0	100	0	0	DNM[1]	100	100	100	100	DNM[1]
Summer	**MS**	0	0	100	0	DNM[1]	DNM[1]	100	100	100	DNM[1]	20	100	100	80
	MG	0	0	0	0	DNM[1]	DNM[1]	0	0	0	DNM[1]	100	100	100	100

[1]DNM = did not monitor

Table 3.5: Prevalence (number of birds and percent of birds testing positive) of *Campylobacter, C. coli*, *C. jejuni* and *Salmonella* in winter by bacteriological isolation.

Bacterial Pathogens	Non-caged Floor-Raised (n=40)	Caged		
		Manure-Belt (n=40)	High-Rise (n=39)	Total (n=79)
Campylobacter	32 (80.0%)[a]	24 (62.0%)[a]	15 (37.5%)[b]	39 (49.4%)[b]
C. jejuni	7 (17.5%)	9 (23.0%)	4 (10.0%)	13 (16.5%)
C. coli	22 (55.0%)[a]	10 (25.6%)[b]	10 (25.0%)[b]	20 (25.3%)[b]
C. jejuni/C.coli	3 (7.5%)	5 (12.8%)	1 (2.5%)	6 (7.6%)
Salmonella	0 (0.0%)	2 (5.1%)	2 (5.0%)	4 (5.1%)

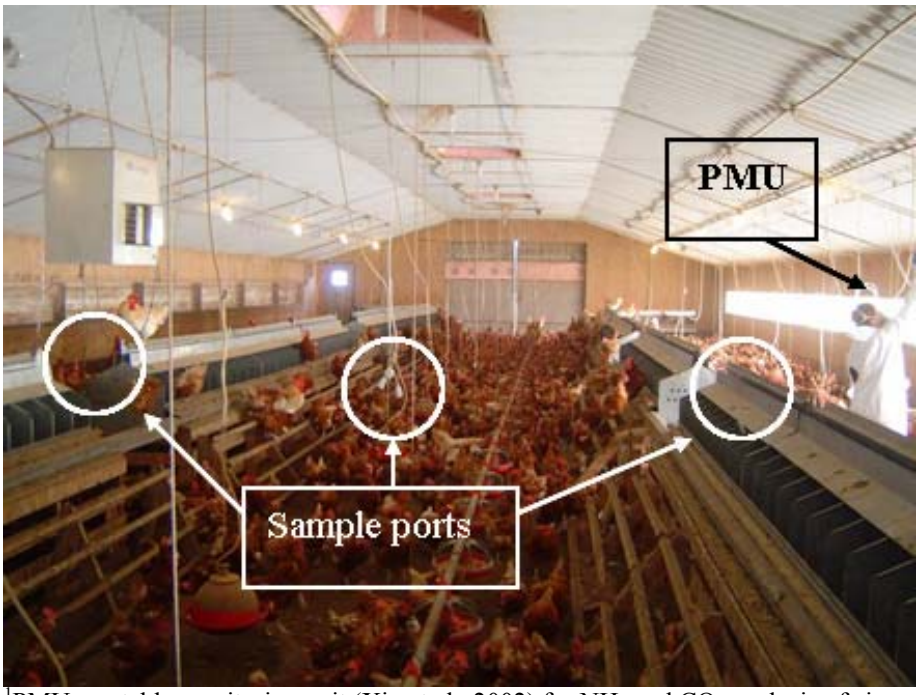
[a,b] Indicates a statistically significant difference ($P<0.05$), calculated using percentage of birds testing positive

Table 3.6: Prevalence (number of birds and percent of birds testing positive) of *Campylobacter, C. coli*, *C. jejuni* and *Salmonella* in summer based on bacteriological isolation techniques.

Bacterial Pathogens	Non-caged Floor-Raised (n=40)	Caged		
		Manure-Belt (n=40)	High-Rise (n=39)	Total (n=79)
Campylobacter	11 (27.5%)[b]	6 (20.0%)[b]	26 (65.0%)[a]	32 (45.7%)
C. jejuni	3 (7.5%)[b]	6 (20.0%)[b]	21 (52.5%)[a]	27 (38.6%)
C. coli	7 (17.5%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
C. jejuni/C.coli	1 (2.5%)	0 (0.0%)	5 (12.5%)	6 (8.6%)
Salmonella	3 (7.5%)	2 (6.7%)	1 (2.5%)	3 (4.3%)

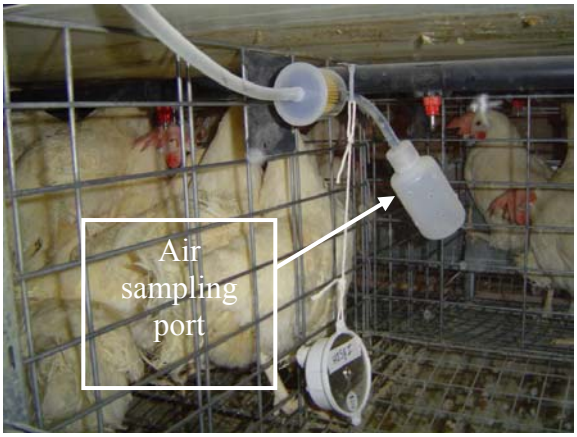
[a,b] Indicates a statistically significant difference ($P<0.01$), calculated using percentage of birds testing positive

Figure 3.1: A photographical view of monitoring configuration[1,2] in the floor-raised (FR) house.



[1]PMU=portable monitoring unit (Xin et al., 2002) for NH_3 and CO_2 analysis of air sample
[2]Circles indicate location of sample ports

Figure 3.2: Photographical views of the bird-level sampling port in a caged house (sampling port placed inside an adjacent empty cage, T/RH logger inside cage and in aisle)



[1]Distance approximately 1.5m or 5ft between T/RH logger inside cage and in aisle

Figure 3.3: Winter conditions (mean±SE) of NH_3, CO_2, T, and RH (I = inside, O = outside) in the floor-raised (FR), high-rise (HR), and manure-belt (MB) laying hen houses monitored.

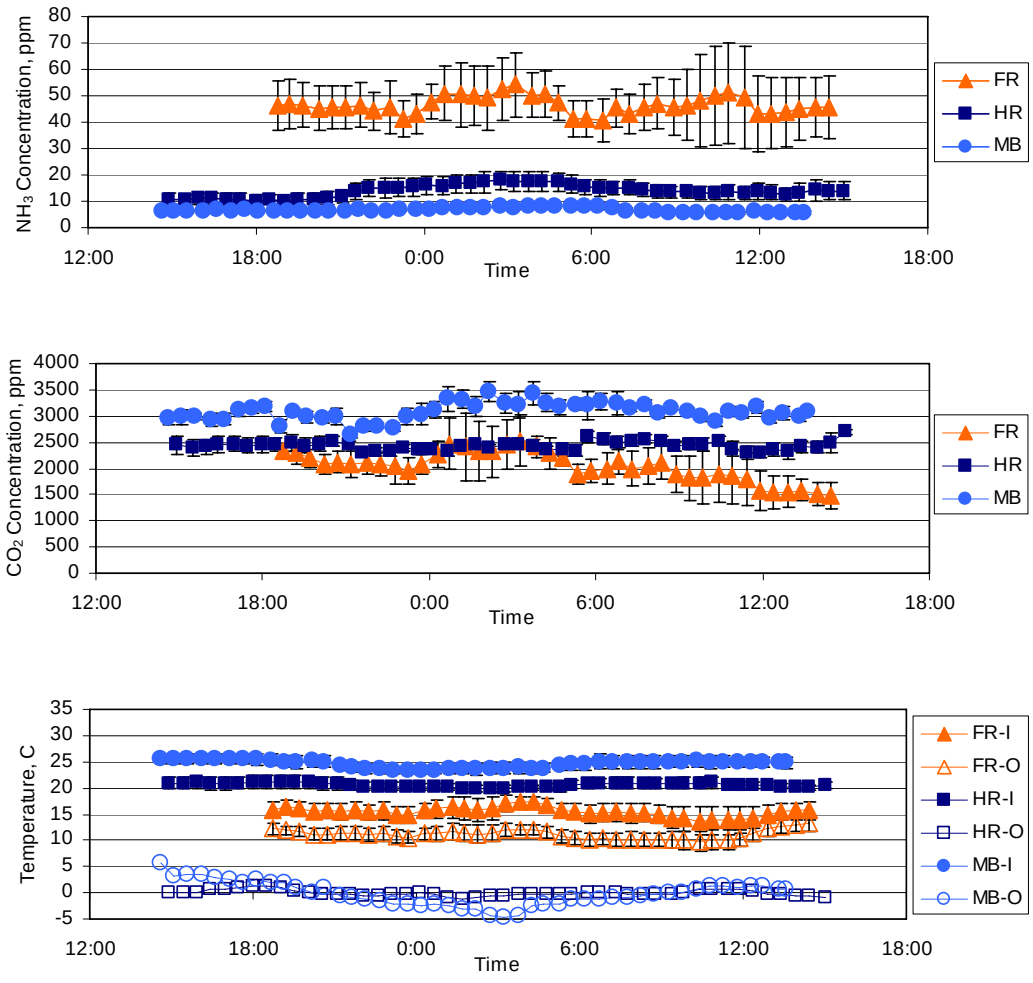
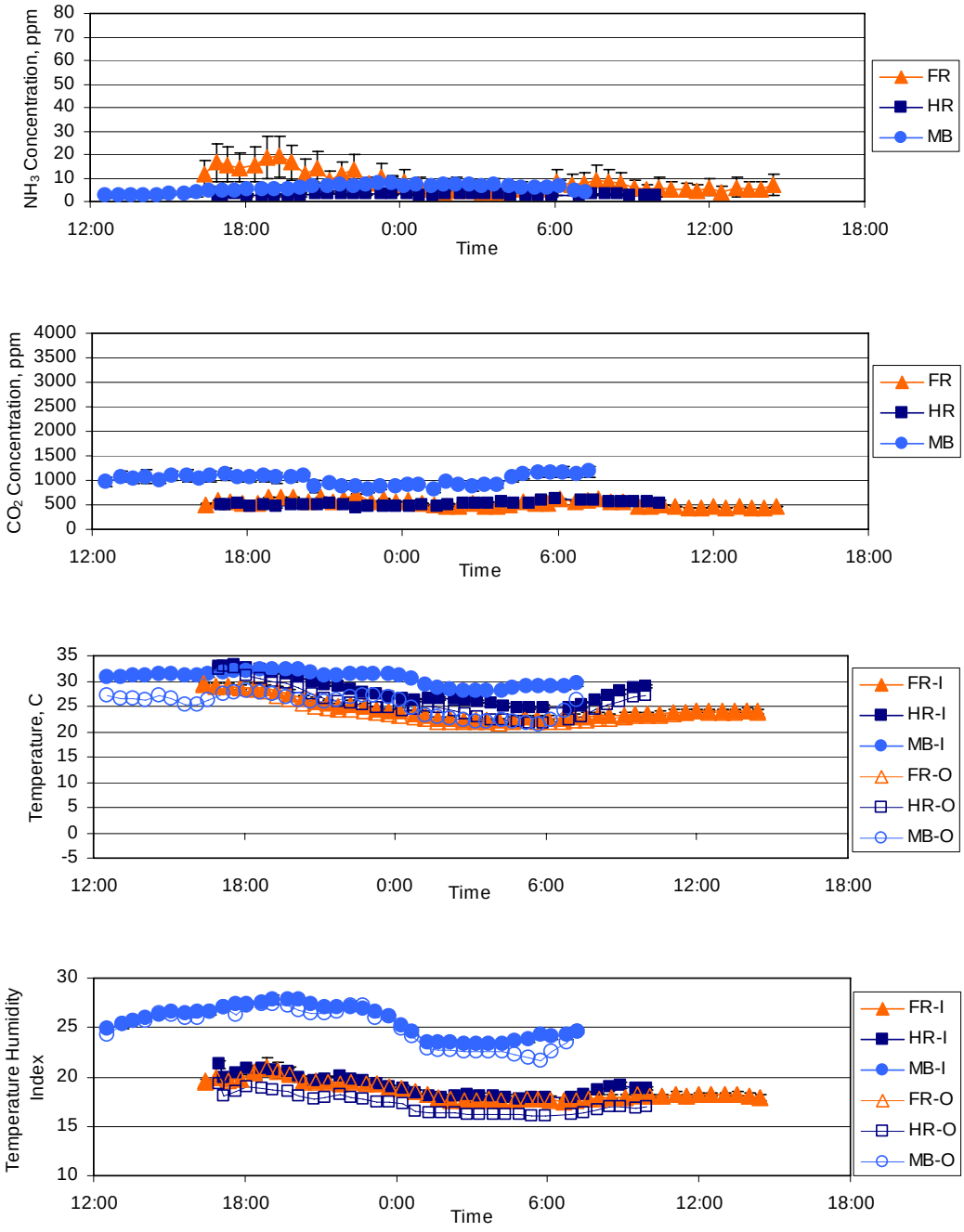


Figure 3.4: Summer conditions (mean±SE) of NH_3, CO_2, T, RH, and THI[1] (I = inside, O = outside) in the floor-raised (FR), high-rise (HR), and manure-belt (MB) laying hen houses monitored.



[1]Temperature Humidity Index: $THI=0.6\times T_{db}+0.4\times T_{wb}$ where db=dry-bulb, wb=wet-bulb

Figure 3.5: Winter conditions of mean temperature and relative humidity difference between cage interior and aisle for high-rise (HR) and manure-belt (MB) laying hen houses monitored.

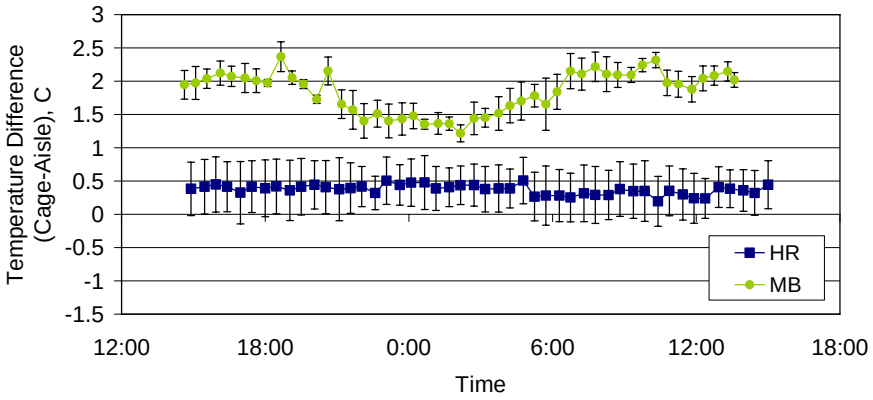
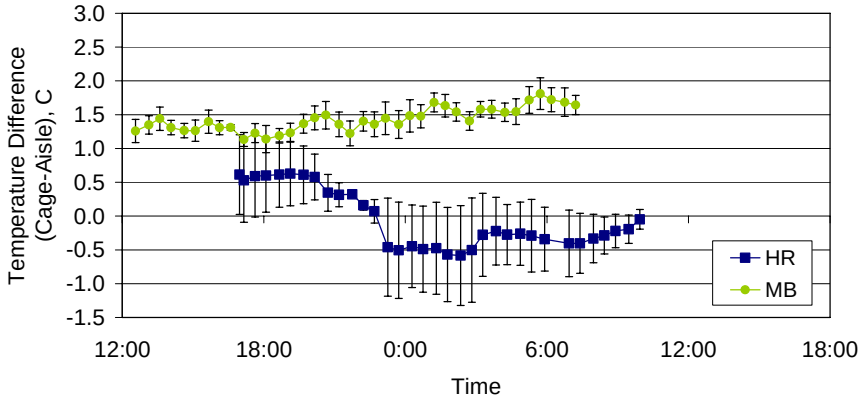


Figure 3.6: Summer conditions of mean temperature and relative humidity difference between cage interior and aisle for high-rise (HR) and manure-belt (MB) laying hen houses monitored.



Chapter 4

Effects of Stocking Density and Group Size on Laying Hens: Part I – Bioenergetics, Production, and Hen Condition under Thermoneutral and Heat Challenge Conditions

A manuscript prepared for submission to Transactions of the ASABE

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Abstract. *Current and relevant information regarding heat and moisture production (HMP) of laying hens is important for design and operation of ventilation systems for commercial layer housing. Different stocking densities are being adopted by the cage layer industry, but inadequate information is available to predict impacts of these changes on environmental control. A study was conducted with 24 groups of 48 hens (39 to 46 weeks old) to compare HMP, via indirect calorimetry, for four different stocking densities (348, 387, 465, or 581 cm^2/bird; 54, 60, 72, or 90 in^2/bird) and two group sizes (8 or 16 birds/cage). Additionally, comparisons were conducted to assess short-term effects of adopting reduced stocking density or varying group size on production and body and feather condition. Data were collected at thermoneutral (24C or 76F) and heat challenging conditions (32C or 90F and 35C or 95F). No notable differences in HMP were observed among the treatments under the experimental conditions (2.8 to 3.1, 3.5 to 3.7, and 6.4 to 6.6 W/kg 24-h time weighted mean SHP, MP, and THP, respectively, under 24C; 0.7 to 1.0, 4.9 to 5.2, and 5.6 to 6.1 W/kg under 32C; and -1.0 to -0.4, 5.9 to 6.5, and 5.4 to 5.7 W/kg under 35C). Differences were observed for bird condition, including greater wing damage for birds housed at 348 cm^2/bird ($P<0.04$)*

and more neck feather damage for birds housed in groups of 8 (P=0.02). Differences were also observed for production variables, including reduced feed conversion at 32C and 35C for 387 cm^2/bird (P<0.007). The results imply that for existing laying-hen houses, reducing stocking density and thus flock size may lead to difficulties maintaining desired temperatures without compromising air quality during cold weather, but may offer benefits for heat stress prevention and relief during hot weather. The differences do not clearly indicate favor for one housing regimen over another for condition and production.

Keywords: cage, layer, heat production, ventilation design, welfare, condition, production

Introduction

Proper ventilation is a critical aspect of controlling the environments within modern laying hen houses. Sizing of housing equipment to provide adequate ventilation and environmental control is partially based on heat and moisture production (HMP) data (ASHRAE, 2005); therefore it is important to have current and relevant information available. Most recent HMP data have shown differences for modern birds as compared to previous data. HMP of the modern laying bird is greater than that of bird strains 20 years ago (Chepete et al., 2004). In addition to effect of genetic strain, it is also likely that HMP is different for birds in different proximity to one another (as with varying stocking density), and it is possible that different group sizes and thus varying locomotion or microenvironment also affect HMP.

A growing sector of the US cage layer industry has adopted reduced stocking densities, though there is inadequate information available to predict the impact this change

will have on operation of existing barns. It is reasonable to hypothesize that birds in closer proximity to one another may have different thermoregulation needs and thus lower HMP than those farther apart. For example, a chicken closer to another warm chicken may need to produce less heat to maintain the same temperature, a 'sharing' effect, and vice versa for those farther apart. However, no evidence to support or refute this hypothesis was found in the literature.

Animal HMP may be assessed using direct or indirect calorimetry methods. Direct calorimetry directly measures heat lost by radiation, conduction, convection (i.e., the sensible mode), and evaporation (i.e., the latent mode). Direct calorimetry is tedious, more costly, and more complicated to operate as compared to indirect calorimetry. Indirect calorimetry takes advantage of the known metabolic relationship between heat production and exchange of respiratory gases (oxygen consumed and carbon dioxide produced). Compared with direct calorimetry, indirect calorimetry is more expensive to build, but less expensive to operate and more flexible to use.

Benefits and consequences of adopting reduced stocking density have been noted. Numerous studies have shown benefits, not only for bird welfare, but for production parameters for lower stocking density. Reduced space allowance beyond a critical point has many negative effects: 1) increased mortality, 2) decreased hen-day production, 3) more egg breakage, 4) reduced net profit per bird, and 5) variable effect on total profits (Bell, 2002). One study demonstrated that sufficient space is important to hens (Lindberg and Nicol, 1996). Another noted that it is important to not only consider the physical space of the birds but also the social space (Keeling, 1995). Production numbers consistently favor lower

stocking density (Lee and Moss, 1995). However, many farmers choose higher stocking density to increase total production and reduce overhead costs per dozen eggs.

Cage systems are frequently criticized for failing to provide space for behavioral functions. Increasing the cage floor space allows opportunity to perform some additional behaviors, and potentially space for escaping aggressive behaviors. The additional space also allows potential for increases in injury by colliding with cage walls and greater opportunity for a range of aggressive actions. Current information in the literature is insufficient to determine optimal space to achieve a balance between additional behavioral freedom and potential for injuries resulting from additional cage floor space.

The objective of this study is to compare HMP, feather and external body condition, and production parameters of W-36 laying hens over a range of stocking densities and group sizes under thermoneutral and heat challenging conditions.

Materials and Methods

Indirect calorimetry laboratory

This study was conducted at the Iowa State University Livestock Environment and Physiology Laboratory (ISU LEAP) that consists of four environmentally controlled indirect calorimeter chambers, each 1.52m x 1.83m x 2.40m (WxLxH, Figure 1). The calorimeter chambers had been used in several previous HMP studies (Xin and Harmon, 1996; Tanaka and Xin, 1997; Xin et al., 1998; Han and Xin, 2000; Xin et al., 2001; Chepete et al., 2004). The calorimetry system consists of an open-circuit, positive pressure arrangement (Figure 2), modified slightly from the arrangement described by Xin et al. (1998). Specifically, the infrared (IR) CO_2 analyzer in the original system was replaced with two IR sensors (Model

GMT222, 0-5000 ppm sensor, Vaisala, Inc., Woburn, MA) arranged in series. Operation and care of the system followed the protocol as outlined in Chepete (2002). The O_2 analyzer and CO_2 sensors were checked daily, and calibrated as necessary. Calibration gases applied were zero (pure N_2), 20.495% and 20.900% O_2 balanced in N_2, and 1500 ppm and 2500 ppm CO_2 balanced in N_2. Recovery tests were performed between each trial to verify operation of the calorimetry system.

The monitoring system was set to collect one air sample every six minutes, for a total of 30 min per cycle (four chambers + fresh air). Air samples were analyzed for O_2, CO_2, and dew point temperature (Figure 3). Following each switch to a new sample (ie. chamber 1 to chamber 2), the readings were allowed to stabilize for 5 min, and an average of the final 1 min of sampling was recorded. Air mass flow rates into each calorimeter chamber, barometric pressure, and temperature and relative humidity of the incoming air and exhaust air were also recorded on the same schedule.

Husbandry and treatment structure

Cages were constructed of 2.54 cm (1 in) square wire mesh attached to a frame of 2.54 cm (1 in) square steel tubing. The cages were assembled in a three tier arrangement, similar to that of a commercial house. Each tier housed 16 birds, for a total of 48 hens per chamber per trial. This number of birds provided sufficient changes in oxygen and carbon dioxide concentrations for accurate measurements. All cages had equal feeder openings (one per bird at spacing of 7.62 cm/bird or 3 in/bird) and drinker access (2 nipple drinkers on one port per 8 birds). Each cage had a sloped floor (approximately 8 degrees) and egg collection

area beneath the feeder (Figure 4). Manure trays were located beneath each cage tier, and manure was removed every 3 days.

Treatment combinations were based upon four stocking densities (348, 387, 465, or 581 cm^2/bird; 54, 60, 72, or 90 in^2/bird) and two group sizes (8 or 16 birds/cage). The variation in stocking density was achieved by varying only the depth of the cages, while maintaining constant feeder space. Group size was varied by addition of a removable section of wire mesh placed at the center of each tier, thus separating the tier into two groups of 8 birds or removing the divider for one group of 16 birds (Figure 4). Once assigned to a cage, birds remained in the same cage for the duration of the trial (Figure 5).

Hens for this study were acquired from a commercial egg production facility in central Iowa. Prior to the study, the hens were housed in cages approximately 51 by 61 cm (20 by 24 in), in groups of 8, 389 cm^2/bird (60 in^2/bird), under thermoneutral conditions. Feed during the trials was provided by the facility to maintain consistency, and Table 1 lists dietary compositions. The hens were randomly selected as needed for each trial from two houses of Hy-Line W-36 birds at 39 to 46 weeks of age. Prior to the start of the data collection, the hens were individually weighed and randomly assigned to cages. Twenty-four (24) groups of 48 hens were used in this study. Each group was allowed at least 2 days of acclimation under thermoneutral conditions (24C or 76F).

In a preliminary trial, four groups of hens were monitored for eight days at thermoneutral conditions to establish the required acclimation period to attain stability of HMP. Two days of acclimation was found to be sufficient to stabilize the bioenergetics responses, paralleling that observed by Chepete et al. (2004).

Following the acclimation, data were collected for 3 days at thermoneutral conditions (24C or 76F), immediately followed by 3 days at 32C or 90F, and finally by additional 3 days at 35C or 95F to simulate heat challenge conditions. Temperature was increased gradually over 6 h during each phase change. All hens were allowed *ad-lib* access to feed and water for the duration of the experiment. Feed was weighed and added; eggs were collected, counted and weighed; and drinkers were checked once per day. During heat challenge conditions, birds were observed and inspected twice daily, and mortalities were collected and documented.

One cage in each chamber (on the middle tier) was selected as a monitoring cage. Five random birds in this cage were tagged for individual identification. All birds were individually weighed and scored (for feather, neck, wing, and claw condition) at the start and end of each trial. Scoring method consisted of a subjective ranking score based on presence or absence of feathers overall and at neck (1=full coverage, 2=moderate coverage, 3=poor coverage; birds with overall poor feather coverage rejected from trial), wing damage (1=no damage, 2=feathers missing or scrapes visible), and quantification of broken claws (Figures 6 and 7). Additionally, the five tagged birds were weighed as a group every 3 days (at the end of each phase) for the duration of the trial. Egg production and total egg weight were documented daily. Feed disappearance and manure weight were documented between each phase of the trial. Manure samples were also collected for moisture content analysis. Manure samples were mixed by hand, a 5 g sample was placed into a clean, dry tray, weighed and oven dried at 100C for 10 h. The tray was removed from the oven and placed in a dry container with anhydrous $CaSO_4$ (Drierite, W.A. Hammond Drierite Co. Ltd, Xenia,

OH) to cool. The sample was weighed, and moisture content was calculated. Moisture content analyses were completed in replicates of five samples.

Statistical Design and Data Analysis

Treatment combinations (stocking density and group size) were assigned to chambers in a randomized incomplete block arrangement (Table 2). Three replicates of each treatment combination were completed during six trials between January and May 2007.

THP, MP, SHP and RQ were calculated based on the equations of indirect calorimetry as described in Appendix A (Xin and Harmon, 1996; Chepete, 2002). For the heat-challenge periods, body mass used in calculations was adjusted daily. Bird body mass was linearly interpolated, using values at the start and finish of each phase, and mortality was subtracted. The bioenergetics data were summarized as daily means as well as means by photoperiod for each temperature condition.

The bioenergetics data were analyzed using SAS PROC MIXED for main effects of stocking density, group size, chamber, trial, and interaction between stocking density and group size. Comparisons were completed for each temperature condition separately. Another analysis was completed with all data that also included a main effect of phase (ie. thermoneutral, heat challenge of 32C or 35C). Effects were considered significant at $\alpha \leq 0.05$.

Each bird condition and production data set was summarized and analyzed using SAS PROC MIXED for main effects of stocking density, group size, chamber, trial, and interaction between stocking density and group size. Significant effects were separated and compared using LSMEANS and PDIFF. Calculations were completed and comparisons were made for average daily feed disappearance, egg production, average egg weight, total egg

mass, feed conversion, manure production (wet basis), moisture content, and manure production (dry basis). Bird scores for feathers, neck, wings, and claws were compared including the main effect of phase for beginning and end. Effects were considered significant at $\alpha \leq 0.05$.

Results

Recovery tests of the indirect calorimeters system yielded similar results for each completion, RQ values ranging from 0.63 to 0.72, CO_2 recoveries from 90 to 102%, and O_2 recoveries from 89 to 97%, for all chambers for all trials.

Figures 8, 9, 10, and 11 demonstrate mean THP, MP, SHP, and RQ, respectively, for each treatment combination at each temperature condition. Tables 3, 4, and 5 summarize the bioenergetics data by daily means, as well as means separated by photoperiod, for each temperature stage, 24C, 32C, and 35C, respectively. During the thermoneutral phase, groups of 16 hens showed higher MP than groups of 8 hens during the dark period (3.30 *vs.* 3.17 W/kg, SE=0.04, P=0.04). No other differences were observed for any of the HMP analyses previously described Figure 12 demonstrates the collective mean for all treatments for THP, MP, SHP, and RQ. Overall results were different among the temperature phases of 24C, 32C, or 35C. Differences in body mass changes were noted between the temperature phases: begin to post-24, no difference; begin to post-32 (P=0.01); begin to end (P<0.0001); but none between stocking density and group size treatments or interactions.

Tables 6 and 7 show results of feed disappearance, feed conversion, manure production, egg production, usable eggs, average egg mass, and total usable egg output for all monitoring periods. Differences were observed for:

- Reduced feed disappearance at 24C for birds housed at 348 cm^2/bird than 387, 465, and 581 cm^2/bird (95 *vs.* 98, 98, and 99, SE=1, P=0.01, 0.02, and 0.006, respectively)
- Improved feed conversion at 32C for 387 cm^2/bird below 465 and 581 cm^2/bird (1.63 vs 1.78 and 1.76, SE=0.03, P=0.004 and 0.009, respectively) and for groups of 8 versus 16 (1.68 vs 1.76, SE=0.02, P=0.02)
- Improved feed conversion at 35C for 387 and 581 cm^2/bird below 348 and 465 cm^2/bird (1.44 and 1.48 vs 1.61 and 1.67, SE=0.04, P=0.007) and groups of 8 versus 16 (1.50 vs 1.60, SE=0.06, P=0.03)
- Greater wet basis manure production at 24C for groups of 16 versus 8 (91 vs 84 g/hen-day, SE=1, P=0.007), with greatest for treatment 581 cm2/bird and group of 16 (95 g/hen-day) and least for 581 cm^2/bird and 465 cm^2/bird and group of 8 (77 g/hen-day)
- Higher moisture content at 35C for 348 and 387 cm^2/bird than 465 and 581 cm^2/bird (66 and 67% vs 64 and 62%, SE=1, P=0.002), with highest MC for 348 and 387 cm^2/bird with groups of 16 (68 and 69%) and lowest for 581 cm^2/bird (59%)
- Greater dry basis manure production at 32C for 387 cm^2/bird over 348, 465, and 581 cm^2/bird (48 vs 43, 44, and 40 g/hen-day, SE=1, P=0.03, 0.04, and 0.002)

- More usable eggs produced at 24C for groups of 8 versus 16 (98 vs 96% of total eggs, SE=0.004, P=0.03)
- Greater total egg mass at 24C for groups of 8 versus 16 (2313 vs 2204 g/chamber-day, SE=25, P=0.02); and greater total egg mass at 35C for 387 and 581 cm^2/bird over 465 cm^2/bird (1052 and 1064 g/chamber-day, SE=42, P=0.04)

Mean bird condition results are shown in Figure 13. Differences were observed for greater wing damage for birds from beginning to end of each trial (1.56 vs 1.61, SE=0.01, respectively, P=0.004). Within treatments, greater wing damage was observed for birds housed at 348 cm^2/bird than at 465 or 581 cm^2/bird (1.62 vs 1.54 or 1.56, SE=0.02, P=0.0007 or P=0.04, respectively) and more neck feather damage for birds housed in groups of 8 than groups of 16 (1.98 vs 1.94, SE=0.01 respectively, P=0.02). More broken claws were observed overall from beginning to the end of each trial (0.48 vs 0.65, SE=0.02, respectively, P<0.0001), with no difference amongst treatments.

Discussion

Adequate acclimation was observed due to the repeatable HMP values for the three days of thermoneutral conditions. Complete production and feed disappearance information was not collected during the acclimation period, and cannot be compared.

The THP values from the current study compare well with most recent values for modern laying hens, 6.5 W/kg in this study vs 6.9 W/kg as reported by Chepete et al. (2004). The diurnal pattern can be easily observed from the higher THP during the light period and

lower THP during the dark period. Reduction of THP has previously been reported as 20% (Riskowski et al., 1977), 35% (MacLeod and Jewitt, 1984), or 25-26% (Xin et al., 1996) from light to dark period. This study showed approximately 25% THP reduction from light to dark.

Reduction of THP as ambient temperature rises has been previously shown (El Boushy and Marle, 1978; Xin et al., 2001). Increases in evaporative losses with rise in ambient temperature have also been documented (Chwalibog and Eggum, 1989). The results of this study also support this finding.

Mortalities affect the stocking density. No difference was observed in the mortalities; hence overall stocking density was decreasing at a similar rate (as mortalities were removed) for all treatments.

Previous studies have also shown that THP is related to physical activities (Boshouwers and Nicaise, 1985). Higher THP was anticipated for the largest cages at the largest group size because each hen had the greatest ability for increased activity (the largest accessible space by each bird). This increase was not observed. It is possible that the largest space was not sufficient to yield an increase in activity. Albentosa and Cooper (2004) found that even when space is sufficient, certain activities are still not performed in cages.

Because the droppings were exposed and not submerged in oil, the evaporation of moisture from the manure was included in the MP determination. The MP estimates reflect what would be expected of a manure-belt hen house system since manure was removed every 3 days. For the same number of birds, the surface area of the manure can vary since the variation in stocking density was achieved by varying cage depth. This could yield a difference in evaporation rate from the manure and thus differences in MP. This likely

explains the differences in MP between groups of 16 and 8, since groups of 16 are better able to use the floor space where the divider is located. The expected difference in MP between stocking densities was not observed. It was noted that birds defecated away from feeder and generally away from drinker in the deeper (Figure 14), thus not defecating over an area smaller than the entire usable floor area. This is particularly interesting to consider as potential to design behavior-specific housing and not worry about defecation through the entire house. Additional research is needed to fully characterize the motivation for defecation location in order to take advantage of this behavior.

The outcome of negative SHP during heat challenge period arose from the calculation of sensible heat production from the difference between total heat production and moisture production. The manure was not submerged in oil (so that the HP values would be reflective of system level). Supplemental heating was used to achieve the elevated temperatures in the chambers, and this additional sensible heat from the heaters (which would not be detected by the O_2 and CO_2 balance) evaporated moisture and inflated the MP result, resulting in lower than actual (and sometimes negative) values for SHP. This effect has no implications for the comparison of treatments or calculating ventilation rate during a heat challenge period (which should be determined for system level, and will not be governed by SHP).

The lack of difference in HMP among the varying stocking densities and group sizes has implications for the egg industry. The work done by Chepete and Xin (2004) showed that a 30% reduction in stocking density does not necessarily result in reduced ventilation (per bird basis) during colder weather because the critical mode for ventilation control is moisture or air quality control as opposed to temperature. The result of reduced number of birds, and thus total sensible heat load, in winter will be somewhat lower barn temperature.

This lower temperature, although not detrimental to thermal comfort of the bird, will likely increase production costs, as more feed energy will be used toward thermoregulation and thus less on egg production. Alternatively, if the barn was controlled only for temperature, the result could be build-up of excessive moisture which in turn leads to condensation problems or reduced air quality. Wathes et al. (1983) highlighted the problem of ventilation solely for thermal comfort may result in an environment with poor air quality. It should be noted that commercial laying-hen barns are typically not equipped with supplemental heating.

Production results do not indicate a clear advantage for one stocking density over another during all temperature phases. The relatively short duration of this study may not have allowed strong differences to be detected. Production numbers in previous studies have consistently favored lower stocking density (Lee and Moss, 1995; Altan et al., 2002; Anderson et al, 2004). Interestingly, manure characteristics during the heat challenge phases, were significantly different, with wetter manure for the larger group size. Manure contained broken eggs, so differences in broken eggs may affect the amount of waste produced, the moisture content of the manure, and also the calculated SHP. Additionally, feed wastage may have an elevated dry basis manure production values, as well as feed disappearance and feed conversion results. This was not quantified, but not observed to be excessive for any of the treatments.

Bird condition, body weight, and feather coverage was uniform at the start of each trial, and uniform at the end. General feather condition did not differ for any treatment or by phase. The number of broken claws increased for all treatments, likely the result of handling, as opposed to the provision of additional space. The injuries to wings were greater for birds

in smaller group sizes, possibly indicating that birds in larger groups can better navigate the cage and crash into the cage walls or each other less frequently. Neck feather condition was better for birds in the lower stocking densities than for the greatest stocking density. Because neck feather pecking is a common behavior in social ordering, this may support the claim that birds in larger space are better able to avoid aggressive actions (including neck feather pecking). This may also indicate that the larger cages promote a more stable social group, and thus less neck feather pecking occurs. Because the data in this experiment were collected over a relatively short period of time, additional research is needed to verify the result and further explore the causation. Savory et al. (1999) reported that feather damage varied with group size and stocking density interactions, and was greater for large groups (20 birds) at higher stocking density (186 cm^2/bird) for bantams in a wire mesh cage. On the contrary, Moinard et al. (1998) reported that feather condition was independent of cage space allowance. The relationship between feather condition and space or group size is likely more complex than be summarized with the simple analyses applied in this and previous studies.

Conclusions

The study presented in this paper affirms the need to further understand consequences of adopting new housing practices, such as reducing stocking density, on environmental control. The results indicate a reduction in stocking density does not affect HMP on a bird mass basis. For example, a 30% reduction in stocking density reduces total heat production by 30% for birds of similar characteristics. Therefore, reducing the number of birds in a given house reduces the heat load, which may be beneficial in hot weather but have adverse effects in cold weather. Based on bird condition and production result, further research is

merited to quantify the impacts of varying stocking density and group size on management and bird health. The benefits did not clearly implicate a trend for stocking density, group size or combination.

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Table 4.1: Feed nutritional composition

Dietary Content	
ME (MJ/kg)	12.51
Crude protein (%)	17.80
Crude fat (%)	5.87
Crude fiber (%)	3.29
Calcium (%)	4.49
Total phosphorus (%)	0.72
Available phosphorus (%)	0.51
Sodium (%)	0.19
Lysine (%)	0.90
Methionine (%)	0.42
Methionine and Cystine (%)	0.76
Choline (mg/kg)	1348.24
Arginine (%)	1.14
Tryptophan (%)	0.18
Threonine (%)	0.68
Isoleucine (%)	0.78
Vitamin A (IU/kg)	7817.96
Vitamin D3 (ICU/kg)	3333.33
Vitamin E (IU/kg)	8.09
Linoleic Acid (%)	1.73
Xanthophyll (mg/kg)	8.87
Chloride (%)	0.31

Table 4.2: Statistical design and treatment allocation among the calorimeter chambers for each trial: stocking density (SD) in cm^2/bird (group size or GS in birds/cage). The English unit equivalents of the SD levels of 348, 387, 465, or 581 cm^2/bird are 54, 60, 72, or 90 in^2/bird.

Trial	Chamber 1	Chamber 2	Chamber 3	Chamber 4
1	348(16)	387(8)	581(16)	465(8)
2	581(8)	465(16)	348(8)	387(16)
3	387(8)	581(16)	465(16)	348(8)
4	465(8)	348(16)	387(16)	581(8)
5	465(16)	581(8)	387(8)	348(16)
6	387(16)	348(8)	465(8)	581(16)

Table 4.3: Sensible heat production (SHP), moisture production (MP), total heat production (THP), and respiratory quotient (RQ) of W-36 laying hens housed under varying levels of stocking density (SD) and group size (GS) under light or dark conditions and time-weighted average (TWA) daily means at 24C

Housing Regimen			BM	SHP, W/kg			MP, W/kg			THP, W/kg			RQ		
			n=144	n=3			n=3			n=3			n=3		
SD (cm^2)	GS		kg/hen	Light	Dark	TWA	Light	Dark	TWA	Light	Dark	TWA	Light	Dark	TWA
348	8	Mean	1.42	3.4	2.2	3.0	3.7	3.2	3.5	7.1	5.4	6.5	0.93	0.92	0.93
348	16	Mean	1.42	3.4	2.2	2.9	3.8	3.3	3.6	7.2	5.5	6.6	0.93	0.93	0.93
387	8	Mean	1.44	3.6	2.4	3.1	3.7	3.3	3.5	7.3	5.6	6.6	0.94	0.93	0.93
387	16	Mean	1.43	3.3	2.0	2.8	3.7	3.3	3.6	7.1	5.3	6.4	0.96	0.95	0.95
465	8	Mean	1.44	3.5	2.2	3.0	3.7	3.3	3.5	7.2	5.4	6.5	0.95	0.94	0.95
465	16	Mean	1.43	3.3	2.1	2.8	3.8	3.2	3.6	7.1	5.4	6.4	0.95	0.94	0.95
581	8	Mean	1.43	3.6	2.2	3.1	3.6	3.1	3.5	7.2	5.3	6.5	0.94	0.93	0.93
581	16	Mean	1.46	3.6	2.0	2.9	3.9	3.3	3.7	7.5	5.4	6.6	0.94	0.94	0.94
	Pooled	SE	0.01	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.02	0.02	0.02

BM = body mass

Table 4.4: Sensible heat production (SHP), moisture production (MP), total heat production (THP), and respiratory quotient (RQ) of W-36 laying hens housed under varying levels of stocking density (SD) and group size (GS) under light or dark conditions and time-weighted average (TWA) daily means at 32C

Housing Regimen			BM	SHP, W/kg			MP, W/kg			THP, W/kg			RQ		
			n=15	n=3			n=3			n=3			n=3		
SD (cm^2)	GS		kg/hen	Light	Dark	TWA	Light	Dark	TWA	Light	Dark	TWA	Light	Dark	TWA
348	8	Mean	1.43/1.34	1.3	0.2	1.0	5.0	5.0	4.9	6.2	5.2	5.8	0.88	0.84	0.87
348	16	Mean	1.44/1.31	1.3	-0.1	0.9	5.3	5.4	5.2	6.5	5.2	6.0	0.88	0.83	0.87
387	8	Mean	1.45/1.35	1.4	-0.3	1.0	5.3	5.4	5.3	6.6	5.2	6.1	0.89	0.84	0.87
387	16	Mean	1.47/1.40	1.2	0.0	0.9	5.1	5.1	5.1	6.4	5.2	5.8	0.92	0.86	0.90
465	8	Mean	1.37/1.35	1.1	-0.2	0.7	5.1	5.2	5.0	6.1	4.9	5.6	0.96	0.87	0.91
465	16	Mean	1.48/1.36	1.2	-0.2	0.8	5.2	5.2	5.2	6.4	5.0	5.8	0.92	0.88	0.92
581	8	Mean	1.40/1.32	1.5	0.0	1.0	4.9	5.1	4.9	6.4	5.1	5.8	0.90	0.85	0.88
581	16	Mean	1.50/1.38	1.3	0.0	1.0	5.3	5.2	5.2	6.7	5.2	6.1	0.89	0.85	0.87
	Pooled	SE	0.1/0.1	0.2	0.2	0.2	0.1	0.1	0.1	0.1	0.1	0.1	0.03	0.03	0.02

BM = body mass (begin of phase/end of phase, means of 5 tagged birds on central tier in each trial)

Table 4.5: Sensible heat production (SHP), moisture production (MP), total heat production (THP), and respiratory quotient (RQ) of W-36 laying hens housed under varying levels of stocking density (SD) and group size (GS) under light or dark conditions and time-weighted average (TWA) daily means at 35C

Housing Regimen			BM	SHP, W/kg			MP, W/kg			THP, W/kg			RQ		
			n=15/n=144	n=3			n=3			n=3			n=3		
SD (cm^2)	GS		kg/hen	Light	Dark	TWA	Light	Dark	TWA	Light	Dark	TWA	Light	Dark	TWA
348	8	Mean	1.34/1.27	-0.3	-0.7	-0.5	6.3	5.5	6.0	5.9	4.7	5.5	0.84	0.81	0.83
348	16	Mean	1.31/1.27	-0.6	-0.9	-0.7	6.6	5.7	6.3	6.1	4.7	5.6	0.84	0.81	0.82
387	8	Mean	1.35/1.29	-0.8	-1.3	-1.0	6.7	6.1	6.5	5.9	4.8	5.5	0.86	0.83	0.85
387	16	Mean	1.40/1.31	-0.3	-0.8	-0.5	6.2	5.4	5.9	5.9	4.7	5.4	0.87	0.87	0.87
465	8	Mean	1.35/1.28	-0.3	-0.6	-0.5	6.2	5.5	5.9	5.8	4.9	5.5	0.83	0.79	0.82
465	16	Mean	1.36/1.30	-0.4	-0.9	-0.6	6.6	5.7	6.2	6.1	4.8	5.6	0.88	0.84	0.86
581	8	Mean	1.32/1.29	-0.3	-0.5	-0.4	6.3	5.5	6.0	6.1	5.0	5.7	0.83	0.79	0.82
581	16	Mean	1.38/1.31	-0.5	-1.0	-0.7	6.7	6.0	6.4	6.1	4.9	5.7	0.86	0.83	0.85
	Pooled	SE	0.1/0.01	0.2	0.2	0.2	0.1	0.1	0.1	0.2	0.2	0.2	0.02	0.04	0.03

BM = body mass (begin of phase/end of phase, means of 5 tagged birds on central tier in each trial)

Table 4.6: Feed disappearance, feed conversion, and manure production for hens housed under varying stocking densities (SD) and group sizes (GS) at 24C, 32C, and 35C.

Housing Regimen			Feed Disappearance, g/(hen-day)			Feed Conversion, g feed/g egg			Manure Production, d.b., g/(hen-day)		
SD (cm^2/hen)	GS		24	32	35	24	32	35	24	32	35
348	8	Mean	95	68	43	1.97	1.62	1.46	59	44	30
348	16	Mean	94	67	46	1.98	1.77	1.75	54	42	34
387	8	Mean	99	70	45	1.98	1.62	1.38	61	48	32
387	16	Mean	97	69	47	2.06	1.64	1.49	62	48	35
465	8	Mean	98	68	45	1.96	1.77	1.70	51	45	31
465	16	Mean	98	71	46	1.96	1.80	1.64	60	43	34
581	8	Mean	95	67	43	1.96	1.71	1.44	47	35	30
581	16	Mean	102	73	46	2.17	1.82	1.52	61	45	31
	Pooled	SE	1	2	2	0.06	0.04	0.06	2	2	2

d.b.=dry basis; Feed conversion=(g feed)/(g usable egg output)

Table 4.7: Egg production, percent good eggs, egg mass, and total usable output for hens housed under varying stocking densities (SD) and group sizes (GS) at 24C, 32C, and 35C.

Housing Regimen			Egg Production, egg/(hen-day)			Good Eggs, % of total			Egg Mass, g/egg			Total Usable Output, g/day		
SD (cm^2/hen)	GS		24	32	35	24	32	35	24	32	35	24	32	35
348	8	Mean	0.82	0.79	0.74	0.97	0.87	0.70	59	58	57	49	36	20
348	16	Mean	0.78	0.74	0.72	0.97	0.81	0.71	60	59	58	44	32	19
387	8	Mean	0.83	0.79	0.80	0.98	0.87	0.75	60	58	60	46	38	23
387	16	Mean	0.80	0.79	0.78	0.97	0.84	0.64	59	57	57	48	36	21
465	8	Mean	0.83	0.77	0.73	0.98	0.84	0.69	60	58	59	48	31	19
465	16	Mean	0.84	0.77	0.76	0.97	0.86	0.70	59	58	58	49	35	19
581	8	Mean	0.82	0.76	0.75	0.99	0.86	0.72	59	58	58	49	37	22
581	16	Mean	0.79	0.75	0.73	0.95	0.91	0.77	59	58	57	43	35	22
	Pooled	SE	0.02	0.01	0.02	0.01	0.03	0.04	0.3	0.4	2	1	2	1

Total Usable Output=(Production)*(% Good Eggs)*(Egg Mass)*Number of Hens

Figure 4.1: Environmentally controlled calorimeter chambers used in this study at the Iowa State University LEAP Laboratory

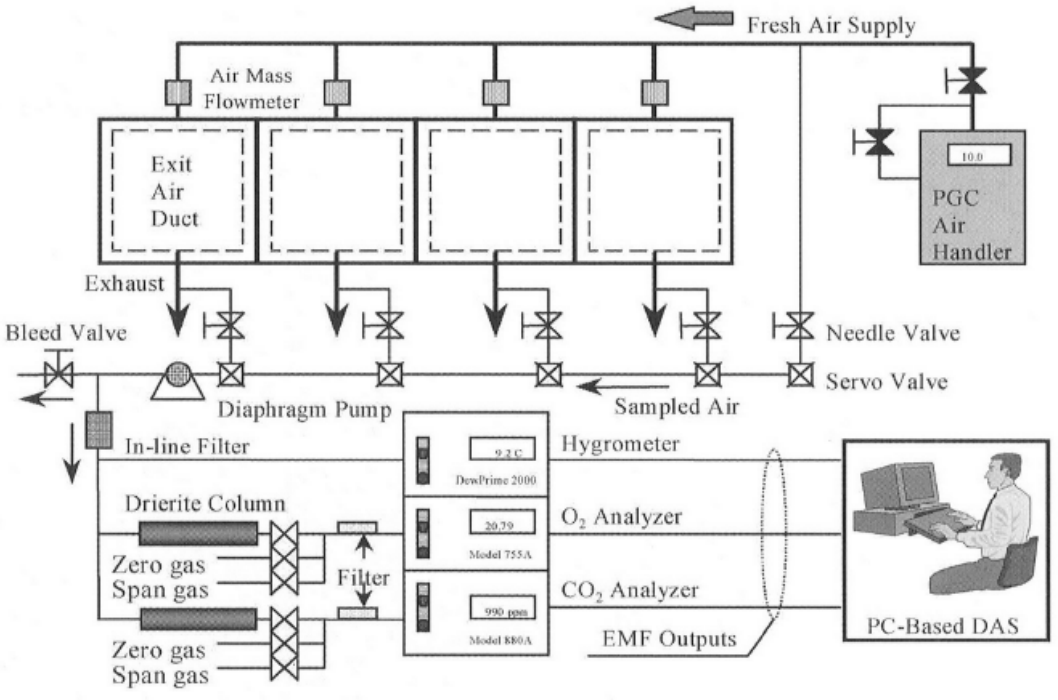

Figure 4.2: A schematic representation of the indirect calorimeter system used in the present study.

Figure 4.3: Gas and dew-point analyzers for measuring bioenergetic response of laying hens

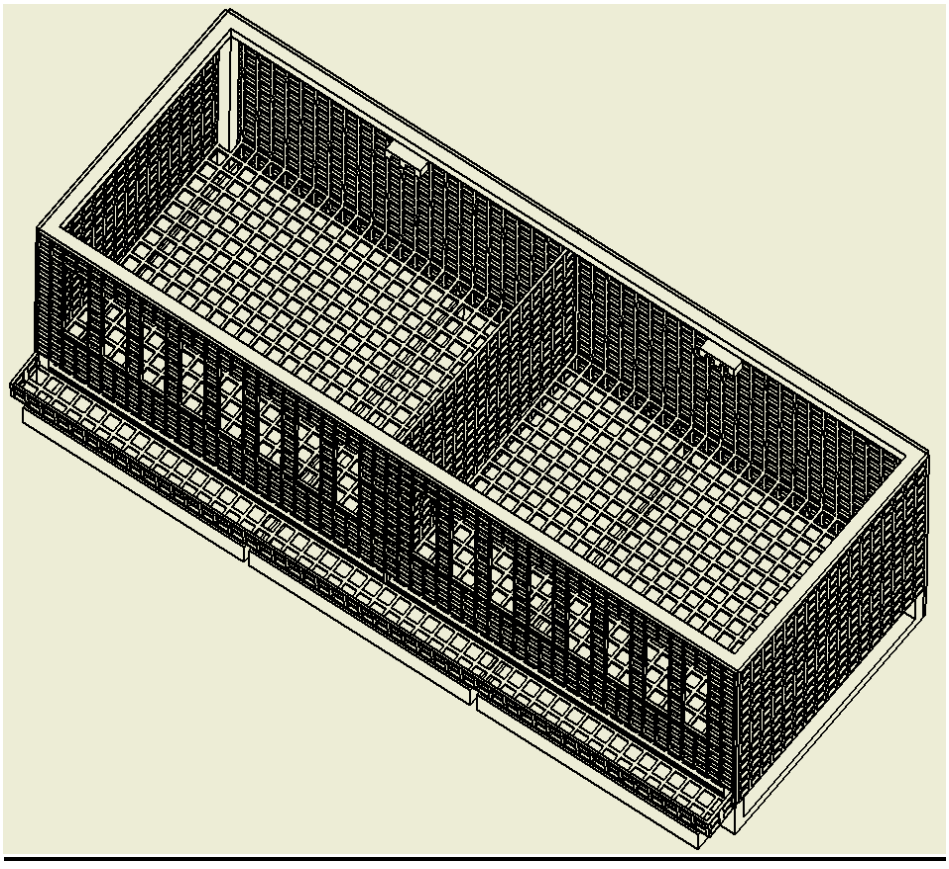

Figure 4.4: A schematic representation of one cage tier with varied stocking density and group size (cage depths vary space and dividers vary group)

Figure 4.5: View of hen cages inside the calorimeter chamber during trial

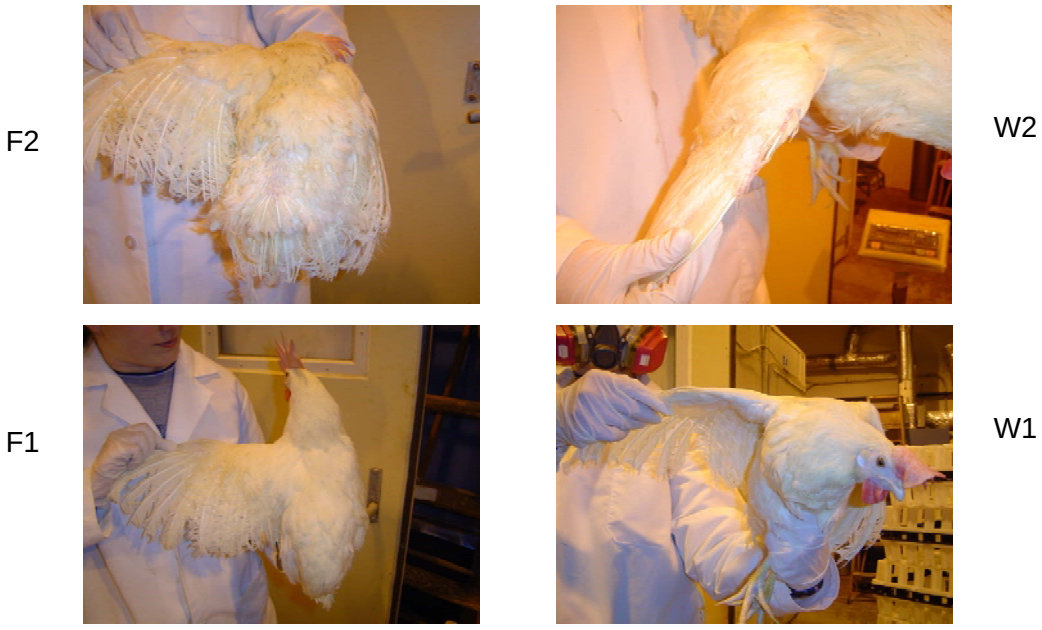

Figure 4.6: Score examples for feather (left) and wing condition (right), hens scored for moderate coverage (F2, upper left) or good coverage (F1, lower left) and wing injured (W2, upper right) or not injured (W1, lower right). NOTE: birds with a feather score of poor coverage (F3) were not used in experiment.

Figure 4.7: Score examples for neck feather condition, hens scored from best coverage (N1, upper left) to worst coverage (N3, lower right)

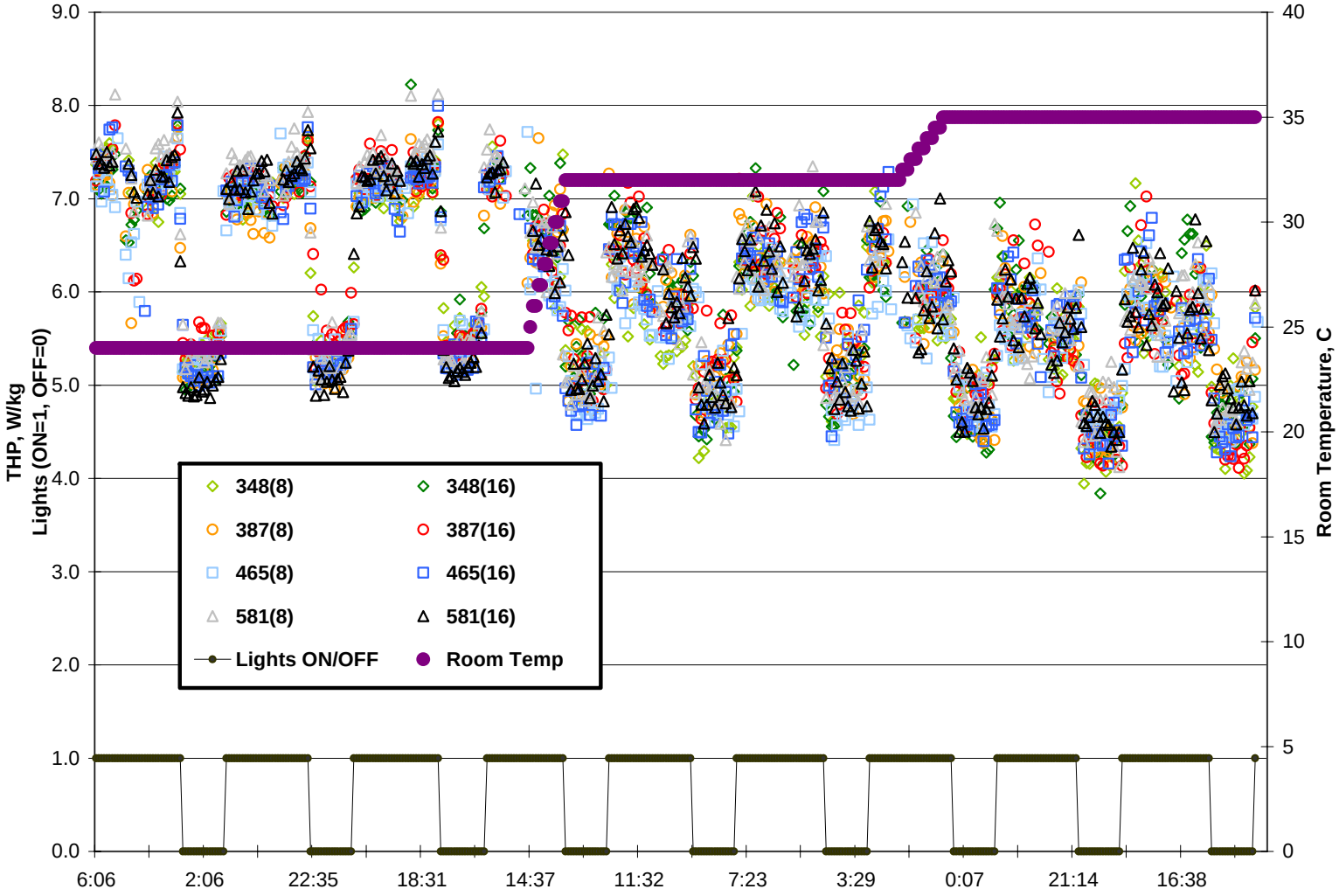

Figure 4.8: Total heat production (THP) of W-36 laying hens housed in different stocking densities and group sizes.

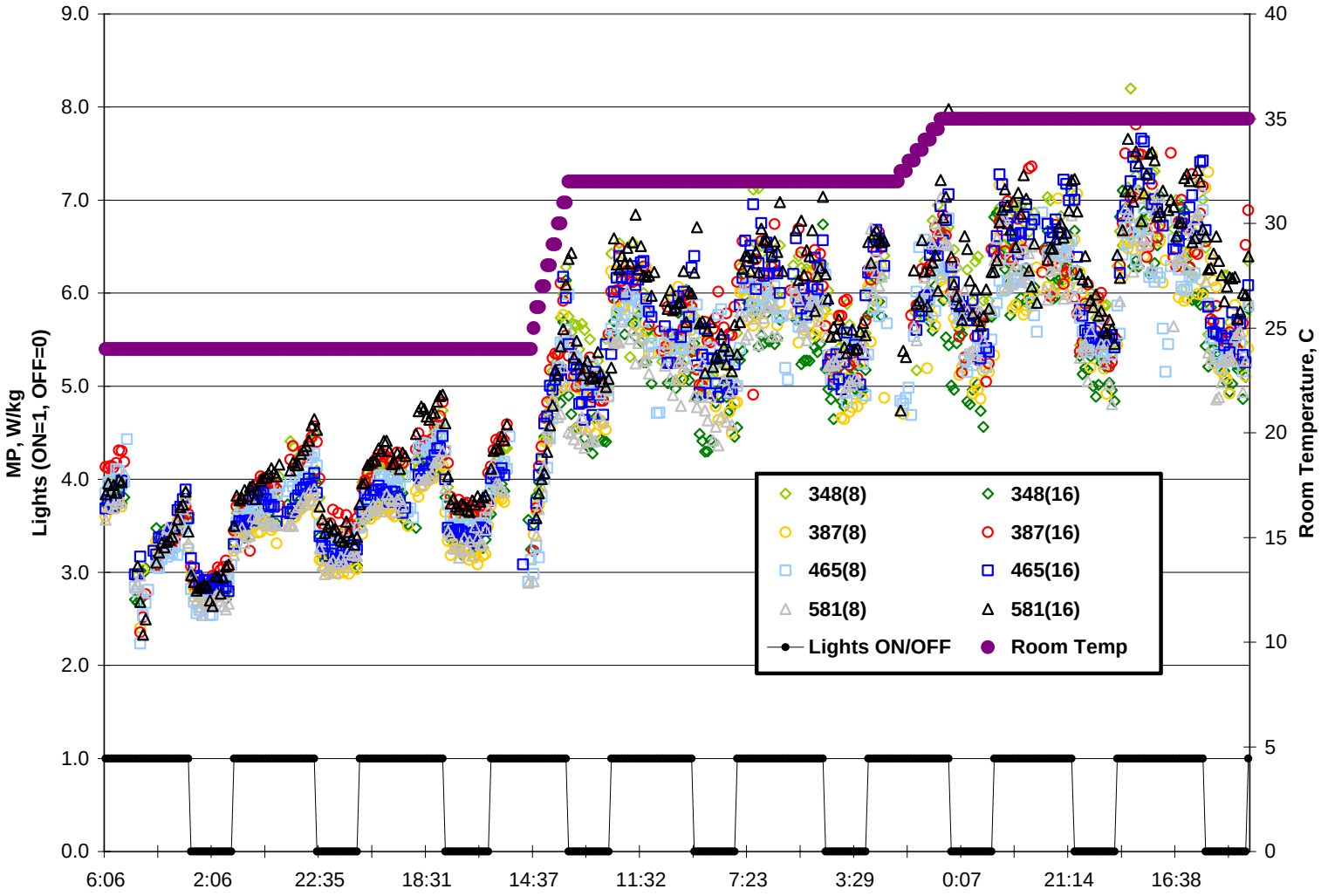

Figure 4.9: Moisture production (MP) of W-36 laying hens housed in different stocking densities and group sizes.

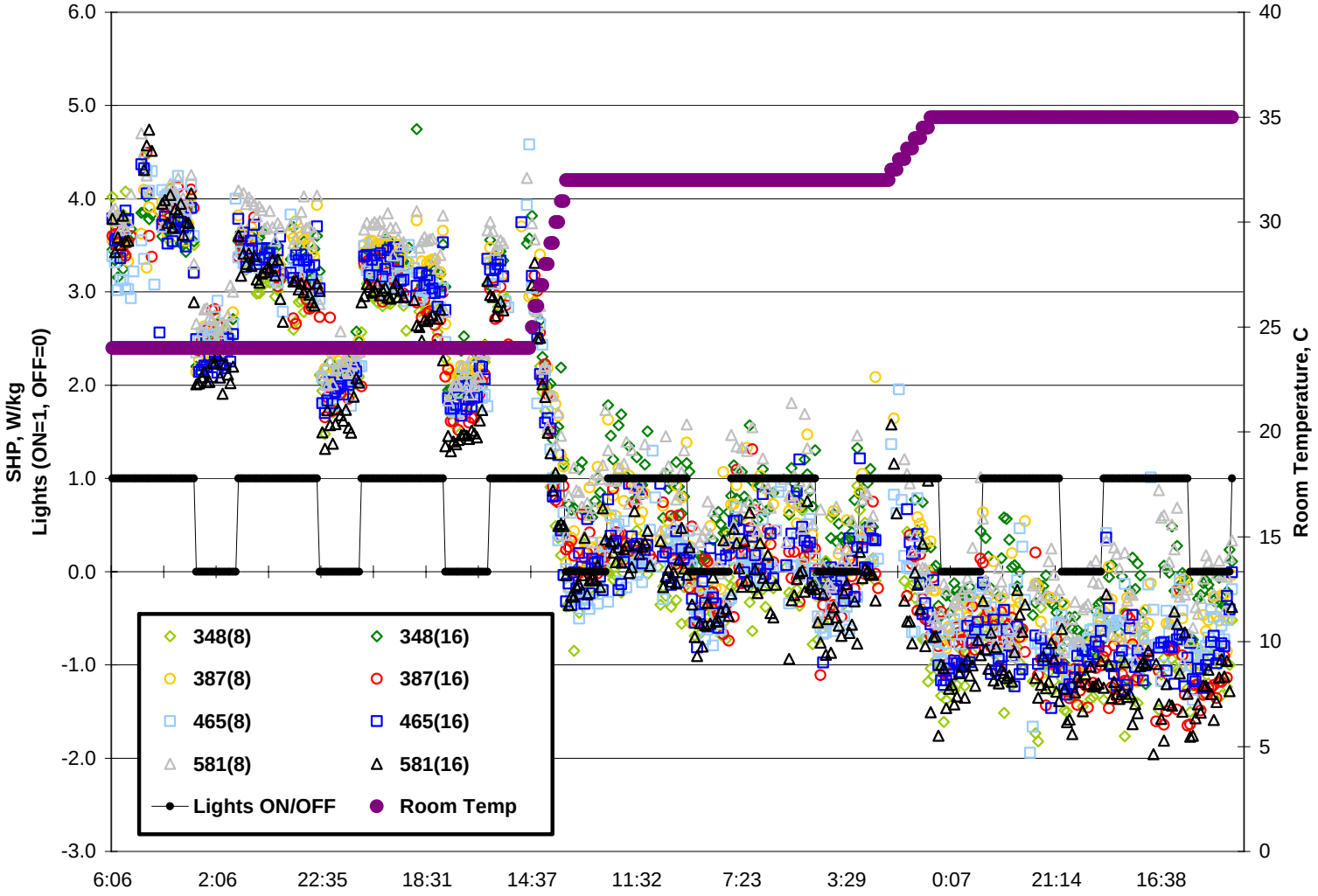

Figure 4.10: Sensible heat production (SHP) of W-36 laying hens housed in different stocking densities and group sizes.

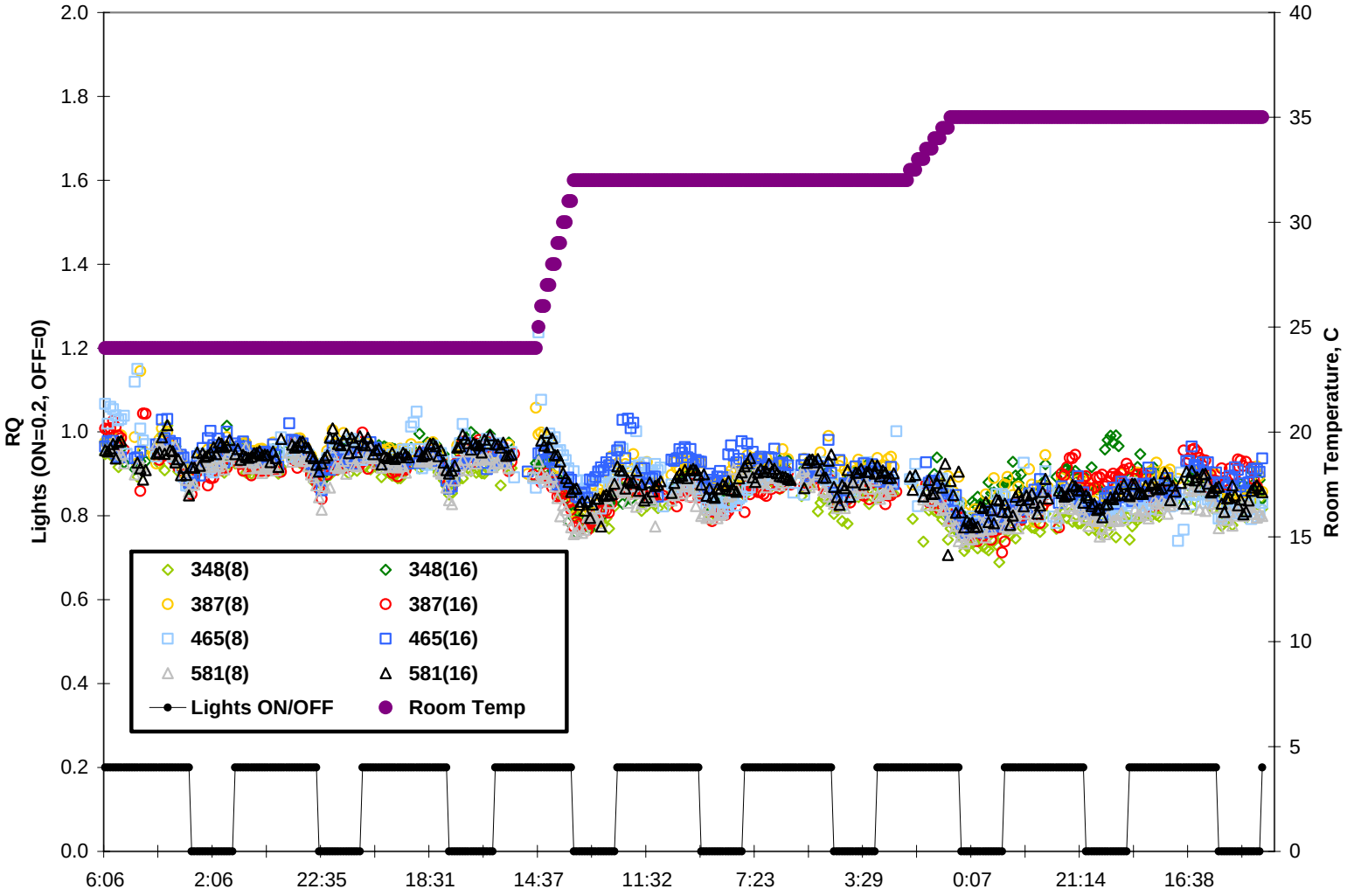

Figure 4.11: Respiratory quotient (RQ) of W-36 laying hens housed in different stocking densities and group sizes.

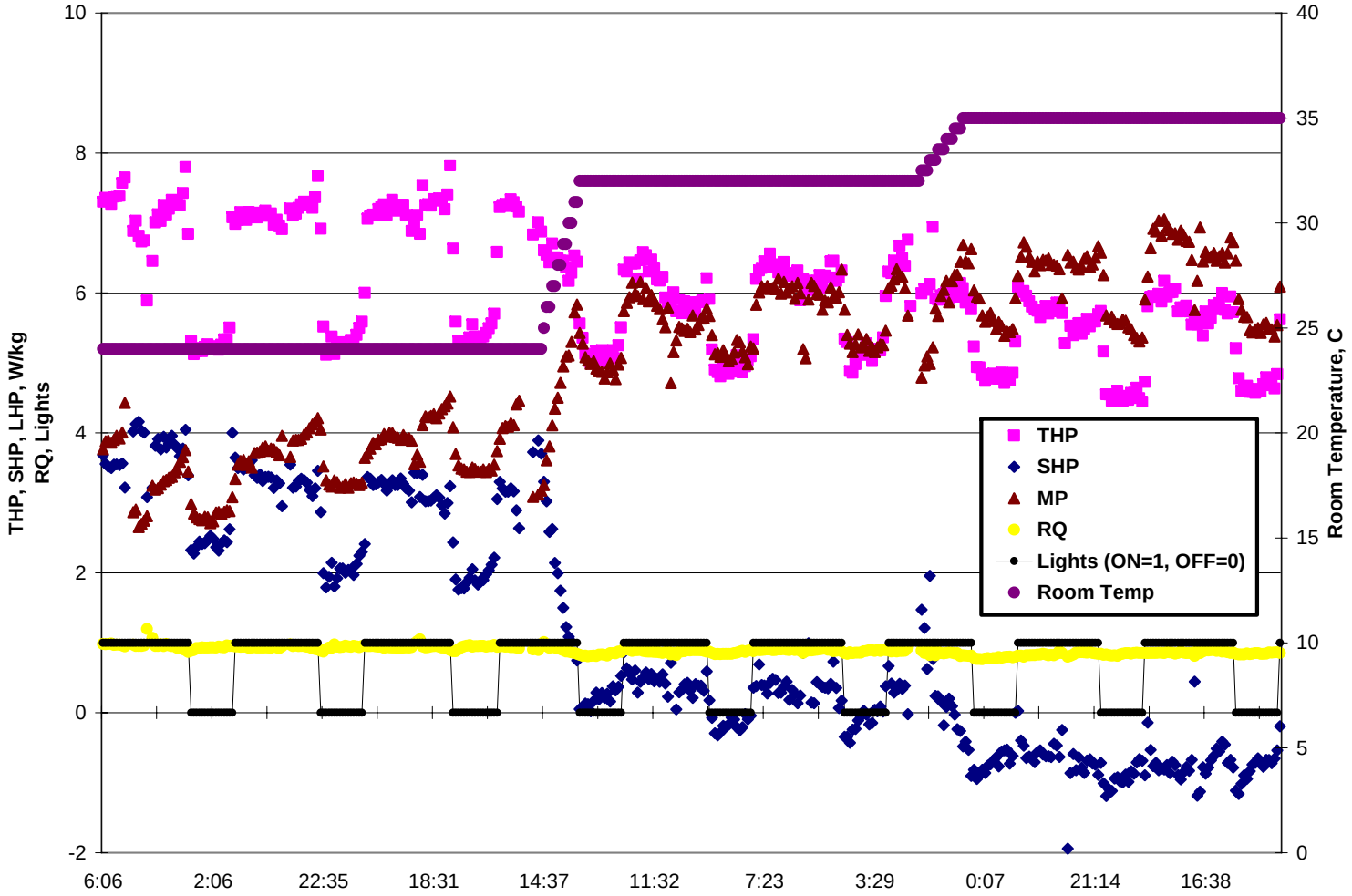

Figure 4.12: Mean plot of heat and moisture production for W-36 laying hens at 24C, 32C, and 35C. THP=total heat production, SHP=sensible heat production, MP=moisture production, RQ=respiratory quotient

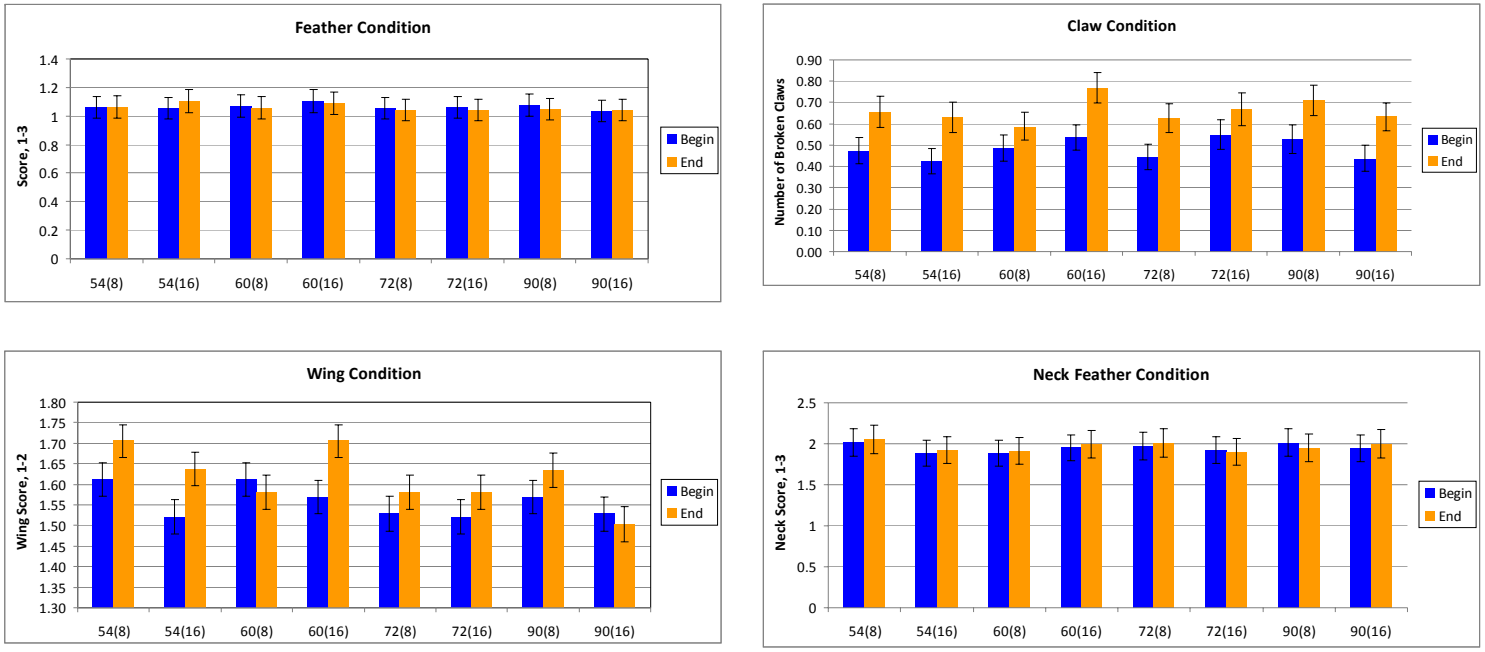

Figure 4.13: Bird condition scores at begin and end of experiment (prior to acclimation and after 35C), (n=144). Refer to Figures 6 and 7 for description of scoring method.

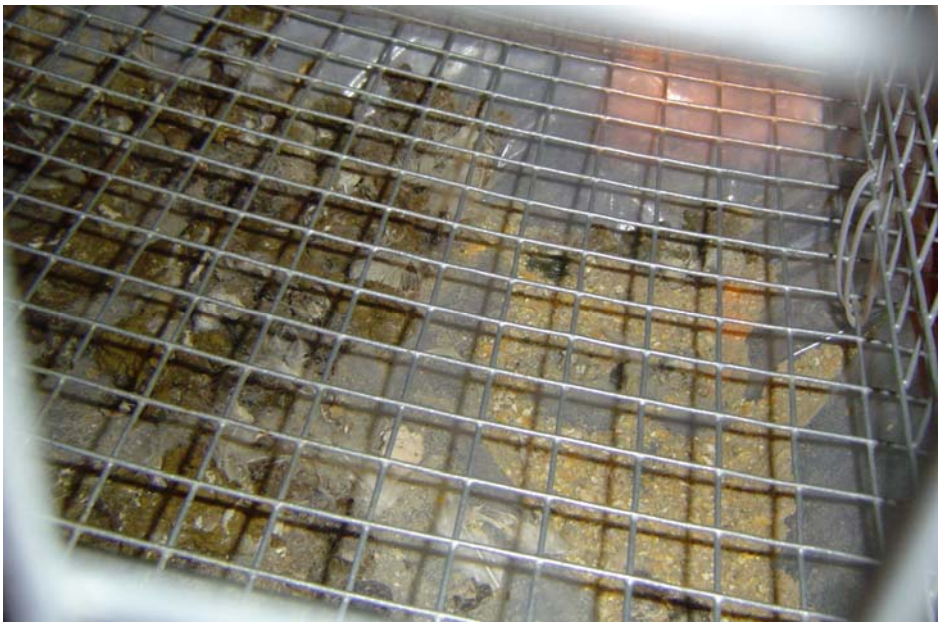

Figure 4.14: Manure accumulation away from feeder. Note feed wastage near feeder.

Appendix A

Equations for calculation of heat and moisture production by indirect calorimetry

(Xin and Harmon, 1996; Chepete, 2002)

$THP = 16.18O_2 + 5.02CO_2\left(\frac{W}{kg}\right) =$ total heat production (Brouwer, 1965)

$O_2 = V_i\left(X_i - \alpha X_o\right)\times 10^{-6}\left(\frac{mL}{s\cdot kg}\right) =$ oxygen consumption rate

$CO_2 = V_i\left(\alpha Y_o - Y_i\right)\times 10^{-6}\left(\frac{mL}{s\cdot kg}\right) =$ carbon dioxide production rate

$V_i\left(\frac{mL}{s\cdot kg}\right) =$ inlet air flowrate at STPD (standard temp and pressure, 20C, 101.325 kPa, dry basis)

$X_i, X_o\left(ppm\right) =$ O_2 concentration at inlet, outlet

$Y_i, Y_o\left(ppm\right) =$ CO_2 concentration at inlet, outlet

$\alpha = \frac{V_o}{V_i} = \frac{1-\left(X_i + Y_i\right)\times 10^{-6}}{1-\left(X_o + Y_o\right)\times 10^{-6}} =$ correction factor for outlet flowrate (McLean, 1972)

$V_{STPD} = \frac{V_{STP}\left(101.325 - P_w\right)}{101.325}\left(\frac{mL}{s\cdot kg}\right)$

$CO_2 = V_i\left(\alpha Y_o - Y_i\right)\times 10^{-6}\left(\frac{mL}{s\cdot kg}\right)$

$P_w = 0.61078e^{\left(\frac{17.2693882\cdot t_{dp}}{t_{dp}+237.30}\right)}\left(kPa\right)$ (Weiss, 1977)

$e = 2.7182818 =$ base of natural logarithms

$MP = V_i\rho\left(\alpha W_o - W_i\right)\cdot\frac{3600}{1000}\left(\frac{gH_2O}{kg\cdot h}\right) =$ moisture production rate

$\rho = 1.293\left(\frac{g}{L}\right) =$ density of air at STPD

$W_i, W_o = 0.62198\left(\frac{P_w}{P - P_w}\right)\left(\frac{gH_2O}{gdryair}\right) =$ humidity ratio of inlet, outlet air (Weiss, 1977)

$$LHP = MP\left(\frac{h_{fg}}{3600}\right)\left(\frac{W}{kg}\right) = \text{latent heat production}$$

$$h_{fg} = 2427\left(\frac{kJ}{kg}\right) = \text{latent heat of vaporization for mean between } T_{bird} \text{ and 21C}$$

$$SHP = THP - LHP\left(\frac{W}{kg}\right) = \text{sensible heat production}$$

$$RQ = \frac{CO_2}{O_2} = \text{respiratory quotient}$$

Chapter 5

Effects of Stocking Density and Group Size on Laying Hens: Part II – Microenvironment and Thermoregulatory Responses under Thermoneutral and Heat Challenging Conditions

A manuscript prepared for submission to Transactions of the ASAE

A.R. Green and H. Xin

Abstract. *Sectors of the US cage layer industry have begun adopting practices of reduced stocking density (i.e., increased cage floor space) and varying group sizes. This study was conducted with 24 groups of 48 W-36 laying hens (39 to 46 weeks old) to assess the effects of cage floor space or stocking density (SD) (348, 387, 465, or 581 cm^2/bird; 54, 60, 72, or 90 in^2/bird) and group size (GS) (8 or 16 birds/cage) on the microenvironment and ability of the hens to cope with heat challenging conditions. Data were collected at thermoneutral (24C or 76F) and warm conditions (32C or 90F and 35C or 95F). On average, temperatures at bird level were 2.9C, 1.4C, and 0.3C, respectively, above the 24C, 32C and 35C room temperature (P<0.0001, P=0.0001, P=0.01, respectively). No differences in core body temperature (CBT) of the hens were observed among the treatment regimens at 24C. In general, mean CBT increased with heat exposure duration (P<0.0001) but leveled off after the 32C phase. At 32C, CBT was higher for GS of 16 versus 8 (42.3 vs. 42.1C, P=0.05); higher for SD of 348 and 387 cm^2/bird than for 465 or 581 cm^2/bird (42.4 and 42.2C vs. 41.9 and 42.1C, respectively, P=0.009); and higher for the second day of the three-day exposure*

at 32C (41.9, 42.2, and 42.1C, respectively, P=0.0007). Bird body mass decreased as heat challenge duration increased (P<0.0001), but no differences were observed amongst the treatments. No mortalities were observed during the thermoneutral period, and the mortality rate increased with heat challenge duration. Minor differences were observed for production variables, including more broken eggs as heat challenge duration increased. The results suggest that decreasing stocking density offers no clear benefit for coping with heat challenge of 32C or 35C; and attest the importance of considering (bird-level) microenvironment in the building environmental control.

Keywords: *cage, layer, heat stress, welfare, core body temperature, production*

Introduction

Heat stress is a concern for animal production agriculture, including egg production. Consequences of heat stress include reduced production performance, impaired immune function and elevated mortality of the animal (Payne, 1966). Heat stress results from the inability of the hen to thermoregulate and thus to maintain homeostasis under increased ambient temperatures and humidity. The hen's core body temperature (CBT) begins to increase when heat dissipation to the environment by conduction, convection, radiation, evaporative losses (panting), and excretion is no longer effective (Bell, 2002).

Core body temperature has been measured by various methods, ranging from manual rectal probe to telemetric, implanted transmitters. Remote, continuous recording of CTB has proven valuable in numerous studies of poultry (Hamrita et al., 1998; Mitchell et al., 2000;

Brown-Brandl et al., 2001; Yanagi et al., 2002; Tao and Xin, 2003a,b). The upper lethal CBT for laying hens is approximately 47C (Bell, 2002).

Generally, the most effective way to alleviate heat stress is through ventilation. Control of the ventilation system within commercial layer houses is typically based upon a few temperature sensors distributed throughout the main aisles, representing the average macroenvironmental temperature. The conditions experienced at bird level, or the microenvironment, may vary. Under heat challenging conditions, this difference could be especially critical to consider.

Stocking density (SD) has been the topic of ongoing debates in the US. Sectors of the US cage layer industry have begun adopting the practice of reduced SD. The United Egg Producers (UEP) recommends cage space allowance between 432 and 555 cm^2/bird (67 and 86 in^2/bird) for white and brown varieties (UEP, 2006), with the upper end of the range intended for larger birds; and McDonald's requires a minimum of 465 cm^2/bird (72 in^2/bird) from its egg suppliers (McDonald's, 2006). However, for the few producers who are not UEP members and do not contract with McDonald's or a similar buyer, compliance with these recommendations are voluntary, and some farms stock as densely as 310 cm^2/bird (48 in^2/bird).

Many unknowns remain regarding the impacts of altering SD. It has been suggested that increased space may offer a benefit to hens during warm weather, when temperatures rise within commercial houses. The objective of this study was to quantify the impact of varying space allowance or SD and group size (GS) of laying hen housing on the macro- and micro-environment gradient, hen CBT, and production responses under heat challenging conditions.

Materials and Methods

This study was conducted using environmentally-controlled calorimeter chambers at Iowa State University Livestock Environment and Physiology Laboratory (ISU LEAP). Hen cages were constructed of 2.54 cm (1 in) square wire mesh attached to a frame of 2.54 cm (1 in) square steel tubing. The cages were assembled in a three-tier arrangement, similar to that of a commercial house. Each tier housed 16 birds, for a total of 48 hens per chamber per trial. All cages had equal feeder openings (one per bird at spacing of 7.62 cm or 3 in/bird) and drinker access (2 nipple drinkers on one port per 8 birds). Each cage had a sloped floor (approximately 8 degrees) and egg collection area beneath the feeder (Chapter 4, Figure 4). Manure trays were located beneath each cage tier, and manure was removed every 3 days.

Treatment combinations were based upon four levels of SD (348, 387, 465, or 581 cm^2/bird; 54, 60, 72, or 90 in^2/bird) and two levels of GS (8 or 16 birds/cage). The variation in SD was achieved by varying only the depth of the cages while maintaining constant feeder space. Group size was varied by addition of a removable section of wire mesh placed at the center of each tier, thus separating the tier into two groups of 8 birds or removing the divider to achieve one group of 16 birds. Once assigned to a cage, birds remained in the same cage for the duration of the trial.

Hens for this study were acquired from a commercial egg production facility in central Iowa. Prior to the study, the hens were housed in cages 51 by 61 cm (20 by 24 in), in groups of 8 at SD of 389 cm^2/bird (60 in^2/bird), under thermoneutral conditions. Feed during the trials was provided by the commercial facility to maintain consistency. The hens were randomly selected as needed for each trial from two houses of Hy-Line W-36 birds, and ranged in age from 39 to 46 weeks. Prior to the start of the data collection, the hens were

individually weighed and randomly assigned to cages. Twenty-four (24) groups of 48 hens were used in this study. Each group was allowed at least 2 days of acclimation under thermoneutral conditions (24C or 76F).

Following acclimation, temperature and production data were collected for 3 days at thermoneutral conditions (24C or 76F), immediately followed by 3 days at 32C or 90F, and finally by additional 3 days at 35C or 95F to simulate heat challenge conditions. Temperature was increased gradually over 6 h during each phase change. All hens were allowed *ad-lib* access to feed and water for the duration of the experiment. Feed was added, eggs collected, and drinkers checked once per day. During heat challenge conditions, birds were observed and inspected twice daily, and mortalities were collected and documented.

One cage in each chamber was selected as a monitoring cage, located on the middle tier leftmost cage when divided. Five random birds in this cage were tagged for individual identification. All birds were individually weighed at the start and end of each trial. Additionally, the five tagged birds were weighed as a group every 3 days (at the end of each phase) for the duration of the trial. Egg production and total egg weight was documented daily. Feed disappearance was documented between each phase of the trial.

A temperature logger (H08-032-08, Hobo Pro, Onset Computer Company, www.onsetcomp.com) was placed inside the monitoring cage and another was hung in the room at the same level as the monitoring cage. The loggers were programmed to collect temperature every 5 min and were downloaded at the end of each trial.

On the afternoon of the third thermoneutral collection day, an ingestible telemetry CBT sensor (1.3 cm dia. by 2.7 cm L) was orally administered to one of the five tagged hens in the monitoring cage of each chamber. The antenna for the CBT sensor was placed at the

top center of the rear wall of the monitoring cage (Figure 1). All four antennas were connected to a receiver unit (model 4000, HQI Technology, Inc., Palmeto, FL) located outside the chamber that was connected to a PC for data acquisition (Figure 2). This CBT monitoring system has been previously applied in other experiments (Brown-Brandl et al., 2001; Yanagi et al., 2002; Tao and Xin, 2003a,b). The system was configured to sample and save every 15s for this experiment. At the end of each trial, each bird was euthanized and sensor retrieved to assess sensor integrity (Figure 3).

Treatment (SD and GS) combinations were assigned to chambers in a randomized incomplete block arrangement (Table 1). Three replicates of each treatment combination were completed during six trials between January and May 2007.

Macro- and micro-environment temperature data were summarized into daily time weighted means, as well as 30 min averages. A composite treatment mean was calculated for the 30 min averages, and a comparative summary plot was developed. The average difference between micro-environment compared with the room environment by phase was calculated collectively using data for all treatments. Daily means were organized for statistical comparison.

The CBT data were processed by filtering the outliers, using a technique similar to Green et al. (2005). Any temperature out of the normal range of a laying hen (40.6-41.7C; Bell, 2002) was discarded. Additionally, any temperature change greater than 0.3C in one sampling period (a change the sensor would be incapable of detecting) was also discarded. The remaining data were summarized into hourly means for developing comparative plots. The hourly means were used to generate daily time weighted average (TWA) and average by

photoperiod values for each treatment regimen, and organized for statistical comparison. The hourly means were also used to calculate average CBT rise above baseline CBT.

Each CBT, bird body mass, production, and mortality data set was summarized and analyzed with SAS PROC MIXED for main effects of SD, GS, chamber, trial, and interaction between SD and GS. Significant effects were separated and compared using LSMEANS and PDIFF. Calculations were completed and comparisons were made for average CBT, average body mass, average daily feed disappearance, egg production, percentage of broken eggs, and average daily mortalities. An additional analysis was completed for CBT, bird body mass, and mortality including the main effect of temperature phase. Micro- and macro-environmental analysis included the main effect of location (cage or room), and were analyzed separately for temperature phases individually, as well as collectively and including main effect of pphase. Treatment effects were considered significant at $\alpha \leq 0.05$.

Results

Figure 4 displays the mean micro-environment (cage temperature) and macro-environment (room temperature) over the trial duration. On overall average, air temperature was significantly higher within the cage (at bird level) than within the aisle (at room level) for all phases, namely, 2.9C, 1.4C, and 0.3C, respectively, above the 24C, 32C, and 35C room temperatures ($P<0.0001$, $P=0.0001$, and $P=0.01$). During the thermoneutral period, the highest SD yielded the highest bird-level temperature rise and the lowest SD yielded the lowest temperature rise ($P=0.01$). The difference between the highest and lowest bird-level

temperature was 0.2C. Group size of 16 yielded a higher temperature at bird level than GS of 8 (P=0.01). No differences were observed for SD or GS during heat challenge conditions.

Table 2 summarizes bird body mass for each phase, separated by treatment regimens. Bird body mass decreased as heat challenge duration increased (P<0.0001), but no differences were observed amongst the treatments.

Table 3 summarizes feed disappearance, egg production, and rate of broken eggs. Feed disappearance was lower at 24C for birds housed at 348 cm^2/bird than at 387, 465, or 581 cm^2/bird (P=0.01, 0.02, and 0.006, respectively); more broken eggs overall as heat challenge duration increased, and more broken eggs at 24C for GS of 16 versus 8 (P=0.03). No differences were observed for egg production amongst treatments.

Table 4 summarizes daily mean mortalities per chamber, separated by treatment regimens. No mortalities were observed during the thermoneutral period, and the mortality rate increased with heat challenge duration. The highest mortalities were observed on the first day of 35C (1.2 birds/chamber or 2.5%, P<0.0001), but there was no clear advantage amongst the treatments.

Table 5 summarizes daily mean CBT separated by phase and treatment regimen. CBT analyses for photoperiod yielded no additional information. Figure 5 depicts mean CBT response and room temperature over the trial duration, with the inserted table highlighting the CBT rise relative to the respective baseline. No differences were observed for CBT at 24C. In general, mean CBT increased with heat challenge duration (P<0.0001) but leveled off after the 32C phase. At 32C, CBT was greatest for GS of 16 versus 8 (42.3 *vs.* 42.1, P=0.05); greater for SD of 348 and 387 cm^2/bird than for SD of 465 or 581 cm^2/bird (42.4 and 42.2C *vs.* 41.9 and 42.1C, respectively, P=0.009); and greater for the second day of

the three-day exposure to 32C (41.9, 42.2, and 42.1C, respectively, P=0.0007). At 35C, CBT was greatest for the regimen of 387 cm^2/bird with GS 16 (42.9C) and lowest for the regimen of 387 cm^2/bird with GS of 8 (42.0C).

Figure 6 depicts CBT responses to micro-environmental temperature, with an inserted table summarizing the slope of lines fit to the data.

Discussion

Micro-environmental temperatures in all tiers were elevated above room temperatures. The elevation was greatest for the highest SD and lowest for the lowest SD. It may be important to consider that the cage temperatures are warmer than the aisle sensors used as feedback for house ventilation system control. It appears that crowding cages increases the bird-level temperature during thermoneutral conditions, although the magnitude of the increase (0.2C) is not likely to have a measurable impact under thermoneutral conditions. During heat challenge conditions, this increase is not significant, and poses no additional threat to bird well-being.

The CBT sensors were all in acceptable condition upon recovery. The epoxy that protects the sensor circuitry was intact, but the outer silicon covering was missing. All sensors were located in the gizzard and none in the crop, as reported to occur occasionally in previous studies (Yanagi et al., 2002).

CBT increased as room temperature increased, and leveled off after the 32C phase, as birds adapted to the warm environment. A positive correlation was observed between all CBT responses and the micro-environmental temperature. Interestingly, the treatment of

348cm^2/bird with a GS of 8 had the lowest slope, but the hens also had the highest baseline CBT.

Feed disappearance includes feed wastage by the birds, not formally quantified, though not observed to be excessive. Difference observed during thermoneutral phase for the highest SD may have resulted from the inability to sham dustbathe (and in the process spill feed into the tray) or may have resulted from competition at the feeder, or a combination of the two. This was not confirmed in this analysis, but it is likely that the restriction of the smaller space allowance prevented the birds from engaging in the same behaviors as birds with more space.

Bird body mass decreased, egg production rates declined, and the percentage of broken eggs increased as the heat challenge duration increased. Mortality increased as the heat challenge duration increased. All of these results were expected, based on information available (Mashaly et al., 2004). There were only minor effects of the treatments, if any, for these variables. This is a critical observation for advantages of one treatment over another. While the conditions and CBT responses may have varied slightly, the ability of the hens to ultimately cope with the heat did not vary by treatment, and varying the space allowance and group size did not offer an advantage for coping with short-term heat challenge. The true benefit of reduced stocking density may lie in the ability to provide more comfortable conditions during periods of warm weather, in which case the hen will have less severe conditions to cope with.

Conclusions

The results of this study imply that decreasing stocking density offers no clear benefit for coping with heat challenge of 32C and 35C, on the basis of physiological responses of the hens and impact on egg production. The results also highlight the importance of including micro-environment in considerations of ventilation control schemes.

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Table 5.1: Statistical design and treatment allocation among the calorimeter chambers for each trial: stocking density (SD) in cm^2/bird (group size or GS in birds/cage). The English unit equivalents of the SD levels of 348, 387, 465, or 581 cm^2/bird are 54, 60, 72, or 90 in^2/bird.

Trial	Chamber 1	Chamber 2	Chamber 3	Chamber 4
1	348(16)	387(8)	581(16)	465(8)
2	581(8)	465(16)	348(8)	387(16)
3	387(8)	581(16)	465(16)	348(8)
4	465(8)	348(16)	387(16)	581(8)
5	465(16)	581(8)	387(8)	348(16)
6	387(16)	348(8)	465(8)	581(16)

Table 5.2: Bird body mass (BM) for hens housed under varying levels of stocking density (SD) and group size (GS) at 24C, 32C, and 35C air temperatures.

Housing Regimen			BM, kg/hen			
SD (cm^2/hen)	GS		Pre-24	Post-24	Post-32	Post-35
			n=144	n=15	n=15	n=144
348	8	Mean	1.42	1.43	1.34	1.27
348	16	Mean	1.42	1.44	1.31	1.27
387	8	Mean	1.44	1.45	1.35	1.29
387	16	Mean	1.43	1.47	1.40	1.31
465	8	Mean	1.44	1.37	1.35	1.28
465	16	Mean	1.43	1.48	1.36	1.30
581	8	Mean	1.43	1.40	1.32	1.29
581	16	Mean	1.46	1.49	1.38	1.31
	Pooled	SE	0.01	0.09	0.09	0.01

SD = floor area; GS = group size

Table 5.3: Feed disappearance, egg production, and broken eggs for hens housed under varying levels of stocking density (SD) and group size (GS) at 24C, 32C, and 35C air temperatures.

Housing Regimen			Feed Disappearance, g/(hen-day)			Egg Production, egg/(hen-day)			Broken Eggs, % of total		
SD (cm^2/hen)	GS		24	32	35	24	32	35	24	32	35
348	8	Mean	95	68	43	0.82	0.79	0.74	3	13	30
348	16	Mean	94	67	46	0.78	0.74	0.72	3	19	29
387	8	Mean	99	70	45	0.83	0.79	0.80	2	13	25
387	16	Mean	97	69	47	0.80	0.79	0.78	3	16	36
465	8	Mean	98	68	45	0.83	0.77	0.73	2	16	30
465	16	Mean	98	71	46	0.84	0.77	0.76	3	14	25
581	8	Mean	95	67	43	0.82	0.76	0.75	1	13	28
581	16	Mean	102	73	46	0.79	0.75	0.73	5	9	23
	Pooled	SE	1	2	2	0.02	0.01	0.02	1	3	4

SD = floor area; GS = group size

Table 5.4: Daily mean mortalities for hens housed under varying levels of stocking density (SD) and group size (GS) at 24C, 32C, and 35C air temperatures.

Housing Regimen			Mortalities, % of flock						
SD (cm^2/hen)	GS		24C	32C D1	32C D2	32C D3	35C D1	35C D2	35C D3
348	8	Mean	0.0	0.0	0.0	0.0	1.5	1.5	0.8
348	16	Mean	0.0	0.6	0.0	0.0	1.9	0.6	0.6
387	8	Mean	0.0	0.6	0.0	0.0	4.2	0.0	0.6
387	16	Mean	0.0	0.8	0.0	0.0	2.1	0.0	0.8
465	8	Mean	0.0	0.8	0.0	0.0	0.6	0.0	4.2
465	16	Mean	0.0	0.0	0.0	0.0	3.8	0.8	0.8
581	8	Mean	0.0	0.8	0.8	0.0	3.5	0.0	0.0
581	16	Mean	0.0	0.0	0.0	0.0	1.9	0.6	0.0
	Pooled	SE	0.6	0.6	0.6	0.6	0.6	0.6	0.6

SD = floor area; GS = group size; D=day

Table 5.5: Daily mean core body temperature for hens housed under varying levels of stocking density (SD) and group size (GS) at 24C, 32C, and 35C air temperatures.

Housing Regimen			Mean Daily Core Body Temperature, C						
SD (cm^2)	GS		Baseline, 24C	32C D1	32C D2	32C D3	35C D1	35C D2	35C D3
348	8	Mean	40.3	42.0	42.4	42.2	42.8	42.6	42.6
348	16	Mean	40.5	42.6	42.9	42.4	42.7	42.4	42.8
387	8	Mean	40.8	42.1	42.5	42.2	42.1	41.9	42.0
387	16	Mean	39.9	41.9	42.4	42.2	42.9	42.8	43.0
465	8	Mean	40.5	41.4	41.9	41.9	42.2	42.7	42.8
465	16	Mean	41.2	41.9	42.2	42.1	42.1	42.3	42.1
581	8	Mean	40.4	41.8	42.1	41.9	42.4	42.4	42.5
581	16	Mean	39.6	41.4	42.6	42.3	42.4	42.6	42.0
	Pooled	SE	0.3	0.3	0.3	0.3	0.2	0.2	0.2

SD = floor area; GS = group size, D=day

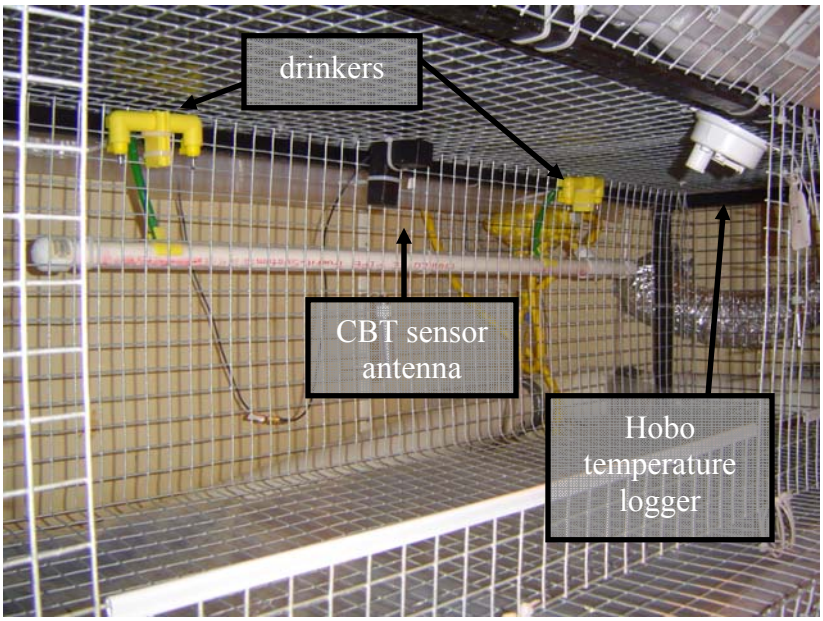


Figure 5.1: Inside of middle cage tier with group size 16, Hobo temperature logger inside cage, core body temperature (CorTemp^{TM}) antenna, drinker, cage divider at center when present.

Figure 5.2: Core body temperature (CorTemp^{TM}) base unit and hosting PC.

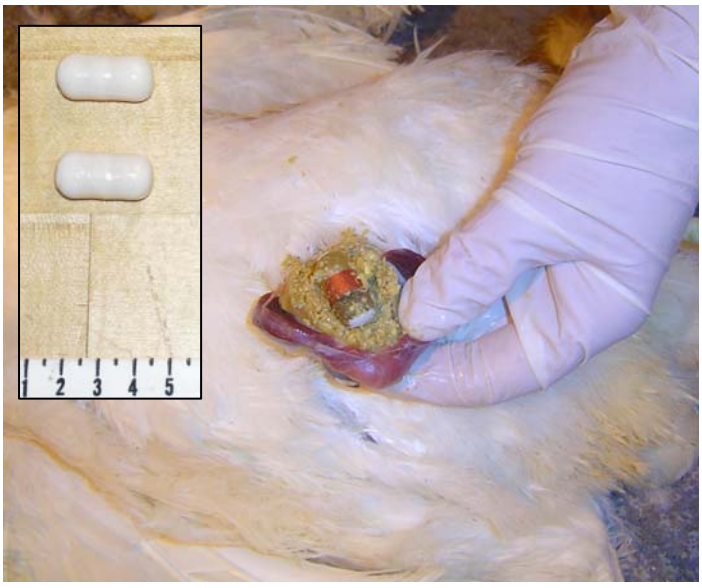
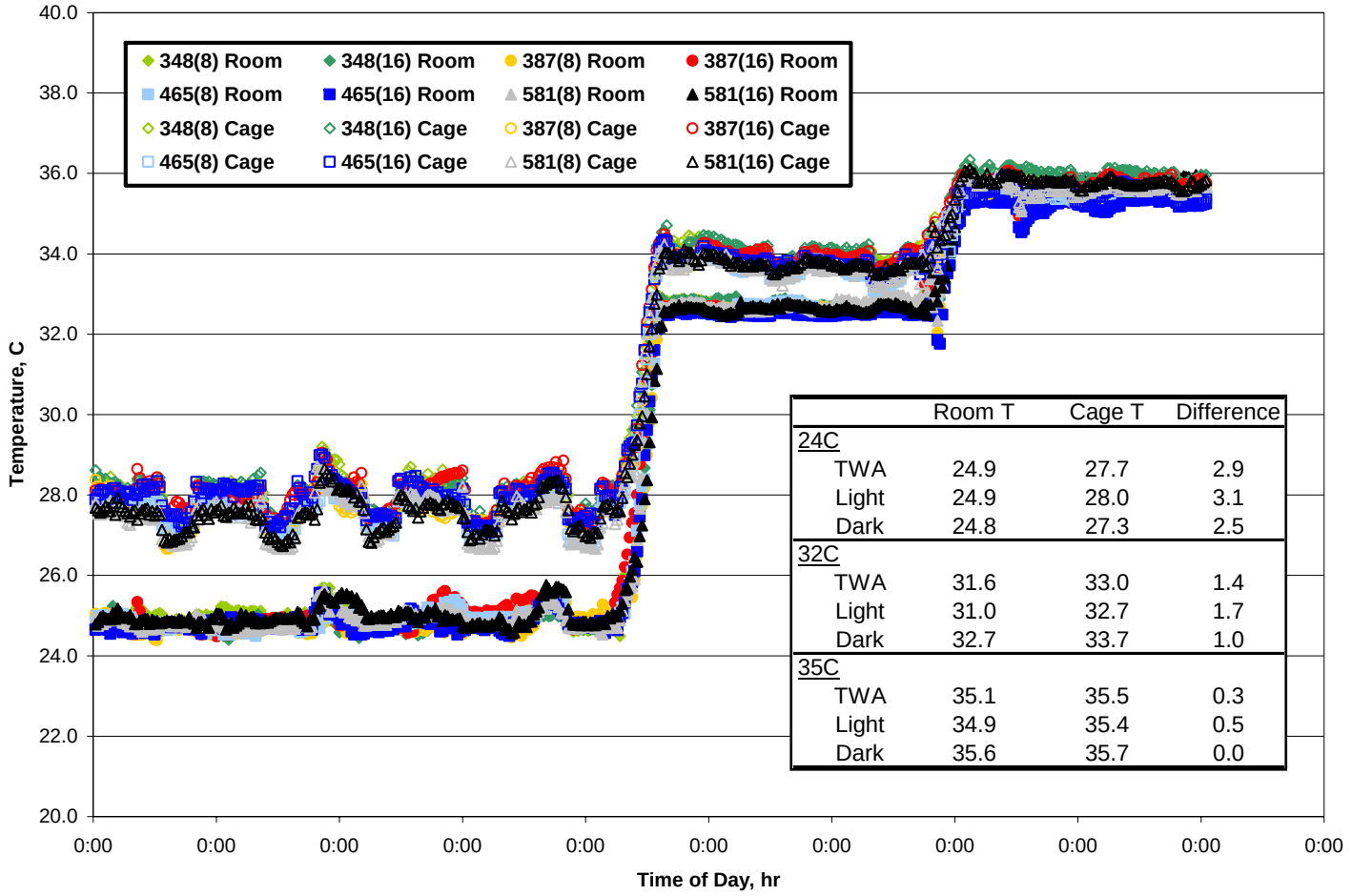
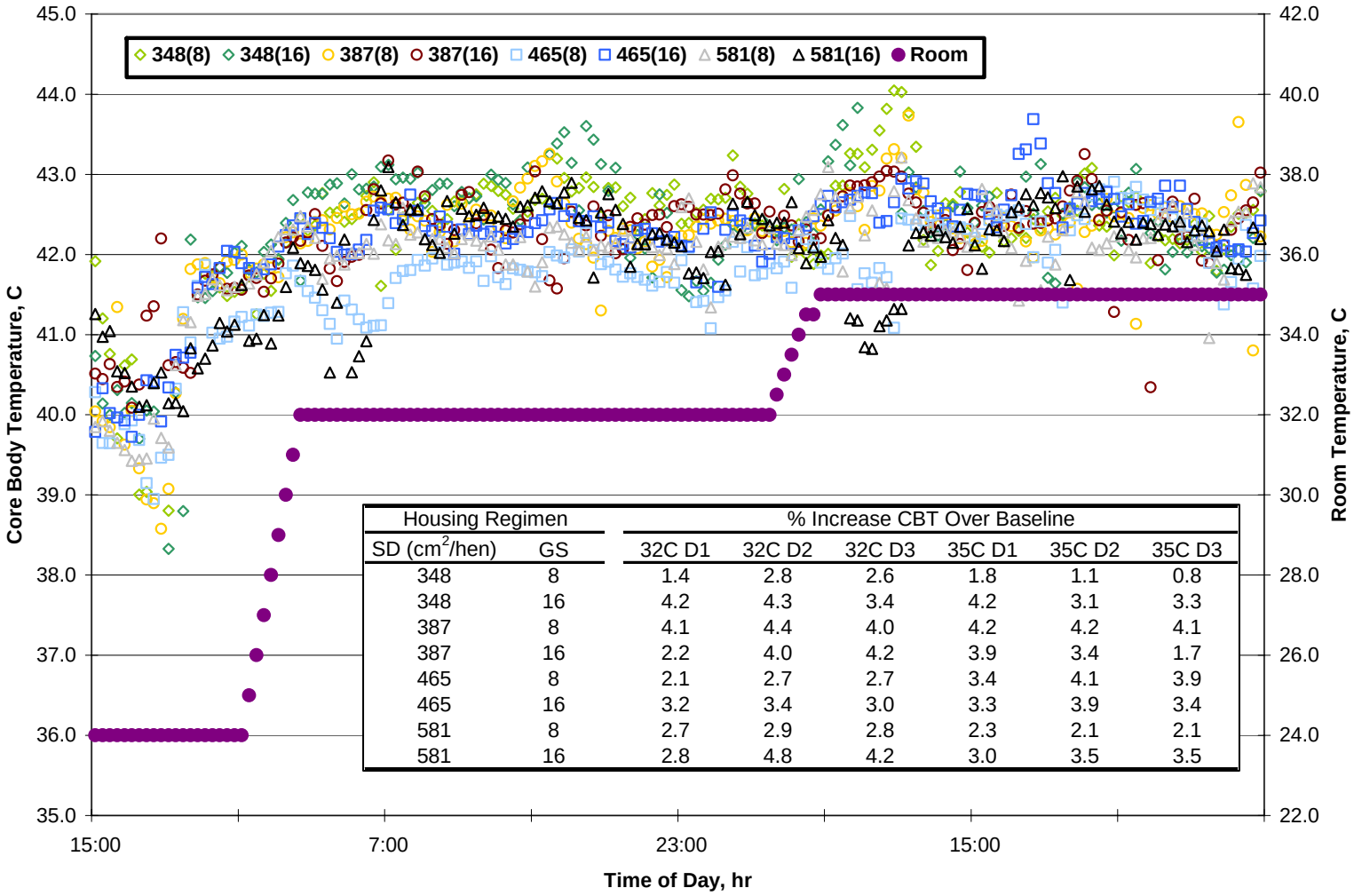


Figure 5.3: New core body temperature sensor (top left, cm scale) and recovery of used sensor.



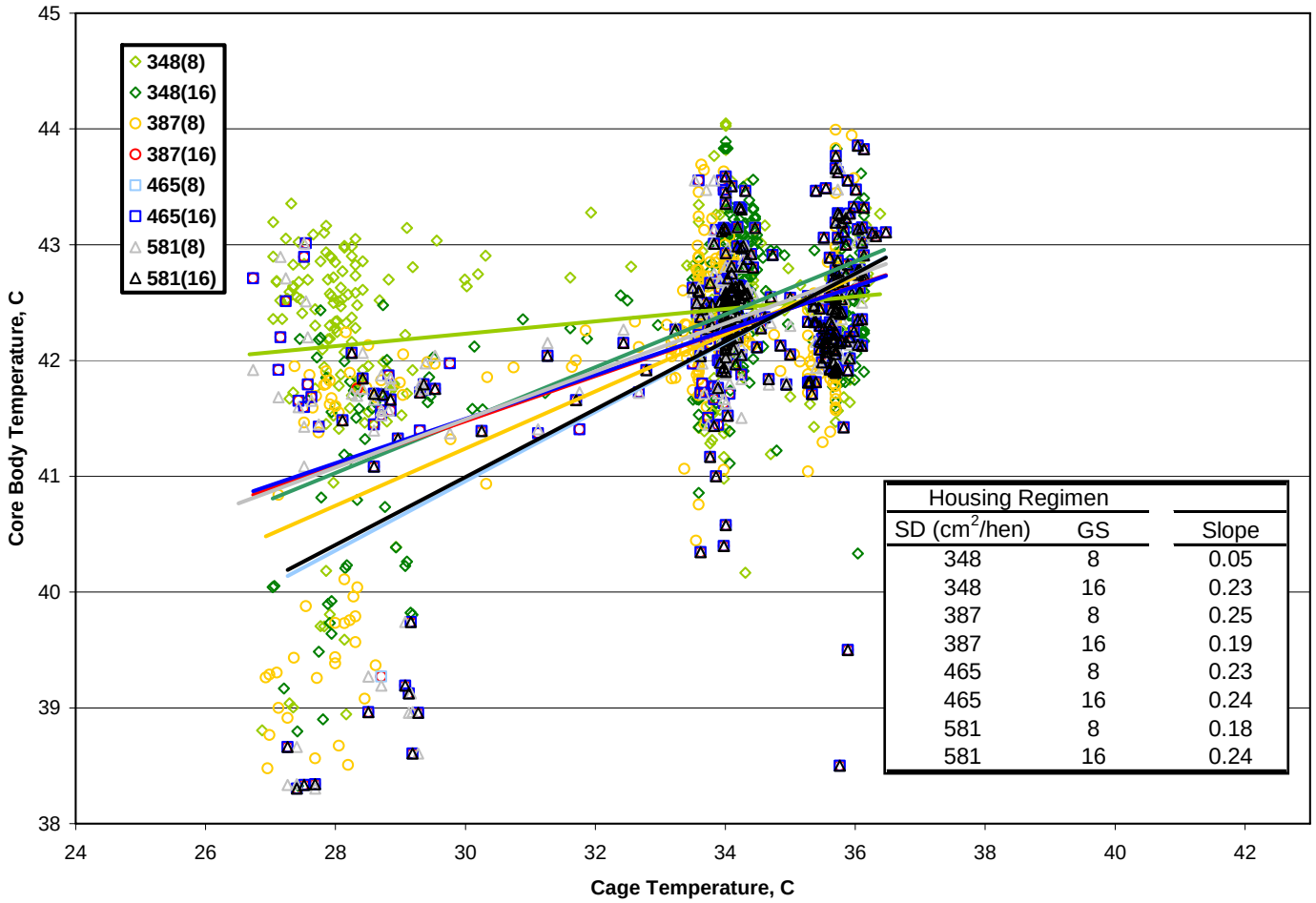
	Room T	Cage T	Difference
24C			
TWA	24.9	27.7	2.9
Light	24.9	28.0	3.1
Dark	24.8	27.3	2.5
32C			
TWA	31.6	33.0	1.4
Light	31.0	32.7	1.7
Dark	32.7	33.7	1.0
35C			
TWA	35.1	35.5	0.3
Light	34.9	35.4	0.5
Dark	35.6	35.7	0.0

Figure 5.4: Mean micro-environment (cage) and macro-environment (room) temperatures, separated by treatments (plot, n=3) and combined into overall mean (insert table, n=8).



Housing Regimen		% Increase CBT Over Baseline					
SD (cm^2/hen)	GS	32C D1	32C D2	32C D3	35C D1	35C D2	35C D3
348	8	1.4	2.8	2.6	1.8	1.1	0.8
348	16	4.2	4.3	3.4	4.2	3.1	3.3
387	8	4.1	4.4	4.0	4.2	4.2	4.1
387	16	2.2	4.0	4.2	3.9	3.4	1.7
465	8	2.1	2.7	2.7	3.4	4.1	3.9
465	16	3.2	3.4	3.0	3.3	3.9	3.4
581	8	2.7	2.9	2.8	2.3	2.1	2.1
581	16	2.8	4.8	4.2	3.0	3.5	3.5

Figure 5.5: Mean hourly core body temperature (n=3).



Housing Regimen		
SD (cm^2/hen)	GS	Slope
348	8	0.05
348	16	0.23
387	8	0.25
387	16	0.19
465	8	0.23
465	16	0.24
581	8	0.18
581	16	0.24

Figure 5.6: Core body temperature versus micro-environmental temperature (plot) and summary of slopes of fitted lines (insert table).

Chapter 6

Development of a Novel Environment Preference Test System for Laying Hens and Its Initial Application to Assess Hen Aversion to Atmospheric Ammonia

A manuscript prepared for submission to Transactions of the ASAE

Abstract. *An environmental preference test chamber (EPTC) was designed, constructed, and utilized in an initial test for response of laying hens to atmospheric ammonia. The EPTC features four interconnected, individually ventilated clear acrylic compartments. Each compartment contained a wire-mesh cage that is divided into two sections, one section used for a test bird to navigate between the compartments and the other section used for three stimulus birds to reside in each compartment. The EPTC was designed to assess individual bird preferences without isolation effects. Alternatively, the section dividers may be removed to assess group preferences. An initial experiment was conducted with six test hens to assess aversion to atmospheric ammonia. Each hen was trained to navigate the inter-compartment door prior to the experiment. Following one day of acclimation to the chamber, data were collected for 2 days at ambient conditions (baseline) and 3 days with ammoniated compartments (25 ppm versus <10 ppm). Hen location (compartment) was documented and compared for baseline and treatment periods. All hens learned to navigate the chamber within 10 h; 4 of the 6 hens learned within 2 h. No preference for fresh versus polluted air was observed with regard to occupancy of environments or number of entries into each environment; further investigation is warranted to determine if this finding is a lack of aversion or other phenomenon. The EPTC developed in this study will also enable future*

users to examine preference responses of hens to other environmental conditions, such as thermal comfort vs. air quality.

Keywords: ventilation, air quality, aversion, behavior

Introduction

In recent years, the perceptions of laying hens regarding the environment in which they are housed have become an important factor for determining housing conditions and establishing husbandry guidelines, especially in the European Union (EFSA, 2005). Preference and motivation testing offer methods for assessing perceptions (Dawkins, 1999). Previous studies have implemented test arrangements ranging from simple choice tests (Sanotra et al., 1995) to varying cost motivational tests (Cooper and Appleby, 1997; Cooper and Appleby, 2003; Olsson and Keeling, 2002) to operant condition tests with key pecking (Faure, 1994) to approach-avoidance tests (Webster and Fletcher, 2004) to interconnected compartments (Albentosa and Cooper, 2005). These studies have reported preferences for environmental parameters such as perches, nest boxes (Freire et al., 1996; Freire et al., 1997), dustbaths (van Liere, 1990; Sanotra et al., 1995), lighting (Davis et al., 1999; Prescott and Wathes, 2002), cage size and feeder space (Faure, 1986), as well as design and construction of cage furnishing. Preference and motivation studies have been used to demonstrate strong motivations for perches and nest boxes (Cooper and Appleby, 1997; Olsson and Keeling, 2002), which consequently led to changes in regulations for housing laying hens in the European Union (EFSA, 2005).

Atmospheric ammonia is a common air pollutant in laying hen housing with potential health implications (Faddoul and Ringrose, 1950; Anderson et al., 1964; Sato et al., 1973) and reduced egg quality and production (Cotterrill and Nordsog, 1954; Charles and Payne, 1966). Ammonia concentrations at ventilation exhaust from commercial egg laying facilities have been reported to range from 3 to 50 ppm for varying housing systems, environmental control systems, and weather conditions (Wathes, 1998; Liang et al., 2005); this may not be reflective of concentrations experienced at bird level, but demonstrates a potentially large variation. Limited information was found regarding hen preferences for air quality. Only one study was found using a chamber for testing environmental conditions, reporting that hens find atmospheric ammonia concentrations greater than 25 ppm highly aversive (Kristensen et al., 2000). Another test was found for gas atmospheres using approach-avoidance for stun gases (Webster and Fletcher, 2004).

The objectives of this work were: 1) to design and construct an environmental preference test chamber (EPTC) which provides the ability to monitor individual or group behavior of birds, to supply varied environmental parameters, and electronic monitoring of bird location within the chamber; and 2) to conduct an initial experiment to assess the performance of the EPTC and delineate aversion or preference response of laying hen to two levels of atmospheric ammonia.

Materials and Methods

Environmental Preference Test Chamber. The EPTC consists of four interconnected compartments, each accessible to two adjacent compartments with a hanging door mounted in a connection passageway. The compartments were constructed with clear acrylic panel (6

mm or 1/4 in) and house a wire-mesh cage divided into two sections, one for three stimulus birds to reside and the other for one test bird with access to the passageways (Figure 1). Stimulus birds (3 in each cage, 12 total) provide a group setting to avoid effects of isolation in preference tests. The test chamber allows 729 cm^2 (113 in^2) within each cage for the test bird, and 1097 cm^2 (170 in^2) per bird for the stimulus birds (Figure 2). Each hanging door assembly (four total) consisted of three connection pieces, one mounted to each adjoining compartment and one containing the hanging door to connect the two (Figure 3). The connection pieces and hanging door (20 by 34 cm or 7.75 by 13.25 in, W by H, suspended at top by two u-bolts 6 mm or 1/4 in diam.) were constructed of clear acrylic panel (6 mm or 1/4 in and 3 mm or 1/8 in, respectively).

The cages are raised above the compartment floor, and manure falls into a removable tray suspended beneath each cage. Each compartment provides handling access to stimulus hens via a wire mesh door fitted into one side wall of each cage and complementary hinged wall in the compartment. Access to test bird area is provided through the top of each compartment by removal of the inlet plenum box, connected by latches at the sides.

Ventilation

Air is supplied to each compartment by individual fans mounted inside one of two insulated mixing boxes. Fresh air is drawn into each mixing box near the ceiling. Two electric fin heaters (120V, Vulcan 0SF1510-350A, Cat. No. 3HM48, Grainger, Kansas City, MO) were suspended in the center of the mixing box, powered by a variable voltage supply, and connected with a temperature limit switch for safety. This allows for control of supply air temperature, if desired. Two small mixing fans (12V DC mini-fan, Model 2730240,

Radio Shack, Ames, IA) were located in the opposite corners near the top of the box and oriented diagonally to enhance mixing within the box (Figure 4). Flanges (10 cm or 4 in diam., aluminum, RS-100, Maurice Franklin Louver Co., Georgetown, SC, with wire mesh removed) were mounted on the outside of the box, and 10 cm (4 in) semi-transparent flexible hose (UFD.020 Thermo Polyurethane Flexible Duct, Item No. 48667, United States Plastic Corporation, Lima, OH) connects the appropriate mixing box to flanges affixed to the inlet plenum of each corresponding compartment. Ventilation supply fans (Delta FFB0412SHN, Cat. No. TGS10-12FAN, RaQware, Shreveport, LA) were located at the inlet side of the flexible hose, mounted to the mixing box wall. For the ammoniated compartments, compressed NH_3 is injected into the supply duct approximately 5 cm (2 in) beyond the supply fan (Figure 5).

Each compartment features a ventilation inlet plenum with an array of 61 holes (19 mm or 3/4 in diam. in an area 47 cm by 47 cm or 18.5 by 18.5 in) oriented above the test bird area (Figure 6). The entire cage and manure collection assembly is elevated to allow exhaust through an array of 61 holes (2.54 cm or 1 in diam. in an area 67 cm by 67 cm or 27 in by 27 in) in the floor of the compartment. The exhaust air passes through a ventilation exhaust plenum with a 15 cm (6 in) opening and then into to the room (Figure 7). A flange (15 cm or 6 in diam., aluminum, RS-100, Maurice Franklin Louver Co., Georgetown, SC) was fitted to each exhaust port to attach filter material for exhaust air.

Ventilation supply to each compartment was checked for uniformity prior to the experiment by assessing velocity in a cross-section of the supply hose to each compartment. Ventilation was estimated by calculating the average air speed (measured by a hand-held airflow transducer) along a horizontal and vertical plane through the hose and multiplying by

the cross-sectional area. Ventilation ranged from 9.3 to 10.5 m^3/h or 5.5 to 6.2 CFM, approximately 19 ACH per compartment.

Control Systems

A Campbell CR10 logger (model CR10, Campbell Scientific, Inc., Logan, UT) was configured to received data and implement feedback to control the ammonia concentration within each compartment (Figure 8). One air sampling line was located along the wire mesh divider within each compartment, with a stainless steel microfilter (5 μm pores, Cat. No. 48222-02, MicroSolv Technology Corporation, Eatontown, NJ) at tubing inlet. An additional sample port was located near the ceiling of the room for sampling ambient air. The sampling lines were connected to one of four solenoid valves (Burkert, model # 456655, Wirrel, UK) controlled by relay switches (SDM-CD16AC, 16 Channel AC/DC Controller, Campbell Scientific, Inc., Logan, UT) for switching to sample air for each compartment. A pump (Gast Linear SPP-6GAS-101, Cat. No. 79610, Cole-Parmer, Vernon Hills, IL) was connected between solenoid valves and air analyzer, with flow meters (0.5-5 LPM, RMA-26-SSV, Cat. No. 116273-30 , Dwyer, Michigan City, IN) to control sampling rate, supply rate, and bypass for excess flow. Samples were analyzed for atmospheric ammonia concentration by a photoacoustic infrared gas detector (Chillgard RT NH_3, Mine Safety Appliances Company, Pittsburgh, PA). Temperature and relative humidity were monitored within each compartment by sensors (HMP35C, Temperature and relative humidity probe, Campbell Scientific, Inc., Logan, UT) located along the divider within each compartment.

Ammonia was supplied to each compartment with compressed 10% NH_3 balanced in N_2. The supply was controlled by individual mass flow controllers (0-100 sccm, FMA5508,

Omega Engineering, Inc., Stamford, CT). Voltage supply to the mass flow controllers for feedback control was provided by voltage divider PC boards connected to additional channels on the relay board for the solenoid valves. An additional solenoid valve was located in the NH_3 supply line to shut off flow in the event of a power failure.

Tracking System

The EPTC was equipped with detection sensors to determine the location of the test hen within the chamber. Three IR emitter-detector pairs (5 mm, 890 nm, OP291A LED and OP555A Phototransistor, Cat No. 365-1057-ND and 365-1077ND, Digi-Key, Thief River Falls, MN) were mounted within each test bird area (Figure 9). Emitters were mounted into PVC for protection of wires and placed above the feeder. Detectors were also mounted into PVC and placed along the divider. Therefore, a hen standing in the test area will be blocking at least one pair. Sensors were connected to a PC board, powered with 2.5V, and the output voltage connected to the CR10 multiplexer (AM416 Multiplexer, Campbell Scientific, Inc., Logan, UT). The EPTC also incorporates digital video monitoring and recording. One camera was located above each test bird section, and recorded continuously for the duration of the trial.

Animal Husbandry

Feed was provided to stimulus birds by a trough near the access door and to the test bird by a container located in the corner of each compartment. Nipple drinkers were located along the wire mesh divider in each compartment. Throughout the entire process, birds were allowed ad lib food and water access.

Lighting within each compartment was checked for uniformity. The best placement for the light was located at the center of the chamber, in the gap created by the four compartments, facing upward into the supply hoses (which acted as a light diffuser).

Hens for this study were acquired from a commercial farm, and were previously used in an experiment assessing thermal environmental conditions under varying housing arrangements. At the time of preference data collection, hens ranged in age from 70 to 76 weeks old (Hy-Line W-36 White Leghorn). All the experimental hens were acclimated under 21C and <5 ppm ammonia environment for several weeks prior to placement in the EPTC. During this time, hens were housed in a holding cage fitted with an identical passageway and connected to an adjacent cage for training purposes. Test birds and most stimulus birds were housed in the same room with visual and vocal, but not physical, contact. Additionally, four stimulus birds (one for each compartment) were housed with and trained with the test birds.

Training Birds

Prior to the experiment, birds were trained to navigate the acrylic hanging door. Two holding cages were joined by a door fitting as described in the husbandry section. Initially, all birds were housed in the same holding cages. Twelve birds were selected from the holding area based on condition and appearance (likely dominant birds but not verified) and placed into the larger of the two cages. The hanging door was fastened open for two days. Several birds thoroughly explored both cages, and the door was returned to its hanging position. No additional incentive was offered, and within a few days, some birds had learned

to navigate the door with ease. All other birds learned the task from these birds within a few weeks. All 12 birds learned to navigate the door, though only six were ultimately used in the initial experiment.

Experimental Design

Test birds were randomly selected from the trained birds, and assigned to the test chamber. Treatments were assigned to compartments in a randomized complete block arrangement, according to the treatment scheme outlined in Table 1. For the initial experiment, two treatments, 25 ppm NH_3 and <10 ppm NH_3, were applied to each of two compartments simultaneously. Once the trial began, the test bird was given at least 1 day to acclimate to the test chamber, under thermoneutral conditions (21C) with comfortable ammonia (<2 ppm). During this period, the test bird was observed to demonstrate its navigation of the chamber by moving into and out of each compartment at least one time. Following the acclimation period, bird behavior was collected for 2 days at comfortable conditions and 3 days with ammonia treatment imposed. On the morning of the transition day between baseline and treatment, manure was removed, eggs were collected, and feed was replenished in all compartments. Following this, ammonia injection rate was increased hourly over 5 hours to achieve 25 ppm.

Analysis. Total time in each compartment was analyzed with data collected by the automated tracking system. An algorithm (using the IR sensor output) was developed to create a summary of location by time and calculate time spent in each condition. The processing algorithm was verified with video for one 24 h period.

Location information was summarized into compartment occupancy for each day of each trial. Summaries were completed for complete 3 day baseline and treatment periods, as well as third (and final) day only of baseline and treatment periods. Data summaries were compared in SAS PROC MIXED for effects of treatment, compartment, phase, and hen. An analysis was also completed using treatment and baseline differences, with the effect of phase removed. Effects were considered significant for $\alpha=0.05$.

Results

The EPTC design was completed (Figure 10), and the chamber was constructed (Figure 11). An initial experiment was conducted to assess hen preferences for fresh versus polluted air (Figure 12). Figure 13 demonstrates IR sensor output and corresponding hen location for a sample data set over several hours. Table 2 presents tracking system accuracy as compared to video analysis for hen occupancy and number of movements into each compartment. Differences in hen occupancy between the two methods were 0.0, 0.2, 0.1, and 0.0 h (for compartment 1, 2, 3, and 4, respectively). Total compartment entries calculated were 179 with the tracker and 242 with the video. Table 3 displays hen occupancy and number of moves into each compartment for complete 3 day baseline and treatment phases. There was no compartment effect for occupancy or number of entries for baseline or treatment phases. Table 4 gives occupancy and number of move into each treatment during the treatment phase only. There was no treatment effect for occupancy or number of entries (11.6 *vs.* 12.5 h or 45 *vs*. 47 entries for ammonia <10ppm *vs.* 25ppm, respectively). In general, the number of moves tended to be greatest on the first day and declined for each consecutive day of the trial, with a slight increase on the first treatment day with subsequent

decline. Figure 14 presents a sample of environmental conditions, compartment temperature and atmospheric ammonia concentration, for one complete trial, including 3 day baseline and 3 day treatment phase.

Discussion

The EPTC described in this study is different from previous chambers (Kristensen et al., 2000; Webster and Fletcher, 2004) because it allows for collection of individual behavior without effects of social isolation. It would be expected that the stimulus birds may affect hen choices, but this effect should be included in the compartment effect (since stimulus birds were always located in the same compartment) and would be overcome by proper randomization and replication. Another benefit is that the divider can be removed if group behavior is desirable, or to supplement individual behavior results.

Prior experience affects subsequent choice (Dawkins, 1976; Bradshaw, 1992). In a no-cost versus cost preference test, access to six areas was offered from a central empty wire mesh cage. Addition of the cost (squeezing between two vertical rods at the door) resulted in decreased frequency of movements, but did not decrease the time spent scratching and pecking, indicating an ethological need (Bradshaw, 1992). A similar approach can be implemented by varying the weighting of the door in the EPTC.

The original design of the EPTC included allowance for testing synergistic effects, such as varying temperature and atmospheric ammonia levels simultaneously, which should be explored in future experiments, as discussed below. There are many other additional applications of the chamber, ranging from air quality to environmental enrichment to nutrition.

Few problems were encountered in the initial application of the EPTC. The most critical challenge resulted from cross-contamination of NH_3 into fresh air compartments from ammoniated compartments. This likely resulted from the variation in ventilation rates from compartment to compartment, which were slightly different at the start of the trials (18-19 ACH) but likely varied as dust accumulated on the exhaust filters. Feedback could be added to the voltage supply to the supply fans to adjust ventilation rate, or adjustable dampers could be added.

The tracking system correctly identified the majority of entries into compartments (179 of 242 entries or 74%). Quick entries and exits were not recorded due to the sampling rate of the sensors (5 s), but the sampling rate can be reduced in future studies if capture of these quick passages is critical. Because the duration of these entries was short, the results for compartment occupancy were only slightly impacted by the failure to indentify quick moves (maximum of 4.7% difference for one compartment). This likely became less important as trials progressed because the number of moves tended to decrease as the trial progressed, with the most on the first day presumably due to chamber exploration. Further validation of the tracking system performance should be completed using more than one day of data and multiple birds. Optimization of the algorithm used to assess hen location might further reduce occupancy error.

The lack of observed aversion to atmospheric ammonia contradicts results reported by Kristensen et al. (2000) and could have resulted from several factors. It is possible that the hens did not find the concentrations in this study aversive. The age of the hens and previous exposure to atmospheric ammonia may have reduced their ability to detect it or may have increased their tolerance level. Mature hens (70 to 76 weeks old) were used in this

experiment, and were 40 weeks old when acquired from high-rise houses on a commercial farm during winter. It is possible that genetics could also affect perceptions. It is also possible that the hens became desensitized to the ammoniated compartments after initial exposure; though this is not likely based on the results of a neurological study quantifying the nerve responses to short-duration ammonia exposure (McKeegan et al., 2002). The previous study used a brown variety, whereas this study used a white leghorn. Another possibility is that the hens' desire to remain with a particular social group or interact with all social groups outweighed desire for fresh air. Because of individual bird to bird variability, a sample size of six birds may be insufficient to reveal an actual aversion. It also must be considered that the hens were unable to associate compartment with environment, and therefore did not recognize the choice offered. One previous study reportedly overcame this obstacle by placing color markers within compartments (Abeyesinghe et al., 2001). These options should be further explored before making conclusions based on the results of this experiment.

Further analysis of the data collected may yield more insight to the perceptions of the hens in this experiment. Occupancy data may be extracted for photoperiod. None of the hens moved during the dark period, heavily weighing occupancy toward the night compartment. Compartment usage during light period may yield different results than total occupancy. In addition, further analysis should include correlations of behaviors with location and environment. An ethogram should include the following behaviors and postures: eating, drinking, sitting, standing, traveling, preening, interacting with conspecifics, other, and unknown/out of view. Behavior and occupancy data may be supplemented with location of egg-lay and quantification of feed disappearance, feed wasteage, and manure dispersal in each compartment.

Assessment of hen environmental perceptions should not be over-simplified or over-generalized. Limitations of preference testing an interpretation of results are acknowledged (Duncan and Dawkins, 1977; Hughes, 1977). It must be considered that some preferences may be non-exclusive, prefer to do a certain behavior in one space and a different behavior in another space (Nicol, 1986). It has also been observed that preferences do not always correlate with functionality. For example, hens were shown to prefer open-sided cages over solid-sided cages (Elston et al., 2000a), even though no behavioral differences were observed within the two types (Elston et al., 2000b). A thorough exploration of methods should be implemented before conclusions are drawn.

Conclusions

An environmental preference test chamber (EPTC) for laying hens was successfully designed and constructed. The chamber consists of four interconnected compartments with an area for a test bird to navigate between the compartments and an area in each compartment for a group of three birds to reside. The EPTC incorporated automated environmental control for atmospheric ammonia concentration and an automated tracking system for location of the test bird. An initial experiment was conducted using the EPTC to assess aversion to atmospheric ammonia. The automated tracking system yielded less than 5% error for compartment occupancy, but failed to identify quick moves through compartments due to sensor sampling rate. The occupancy results revealed no preference for any compartment or treatment. Further investigation regarding hen usage of the compartment and correlations with behavior should be completed.

Acknowledgements

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Table 6.1: Statistical design and treatment allocation, atmospheric ammonia concentrations less than 10 ppm (A<10 ppm) or approximately 25 ppm (A=25), for final 3 days of each trial. Preceding 3 day baseline data collected with fresh air.

Trial	Compartment 1	Compartment 2	Compartment 3	Compartment 4
1	A<10	A<10	A=25	A=25
2	A<10	A=25	A<10	A=25
3	A=25	A=25	A<10	A<10
4	A=25	A<10	A=25	A<10
5	A=25	A<10	A<10	A=25
6	A<10	A=25	A=25	A<10

Table 6.2: Occupancy and number of entries into preference chamber compartments for 24 h as calculated by automated tracking system and video analysis.

Compartment	Cumulative Occupancy				Compartment Entries	
	Tracker, h	Video, h	Difference, h	Difference, %	Tracker	Video
1	2.4	2.5	0.0	0.9	48	61
2	4.2	4.0	0.2	4.7	50	61
3	10.6	10.8	0.1	1.3	38	60
4	5.9	6.0	0.0	0.5	43	60
				TOTAL	179	242

Table 6.3: Mean daily occupancy and number of entries into preference chamber compartments during 3 day baseline and 3 day treatment phases (n=6).

Compartment	Occupancy, h		Occupancy, %		Compartment Entries	
	Baseline	Treatment	Baseline	Treatment	Baseline	Treatment
1	5.5	5.8	22.7	24.1	23	22
2	7.9	8.8	32.6	37.0	27	26
3	6.3	5.5	26.1	23.0	19	14
4	4.6	3.8	18.6	15.9	23	20
SE	1.0	1.0	5.0	5.0	6	6

Table 6.4: Mean daily occupancy and number of entries into preference chamber treatments for final 3 days of preference trials with ammonia <10 ppm NH_3 or ammonia controlled at 25 ppm NH_3 (n=6).

Treatment	Occupancy, h	Occupancy, %	Compartment Entries
A<10	11.6	48.0	45
A25	12.5	52.0	47
SE	2.0	10.0	12

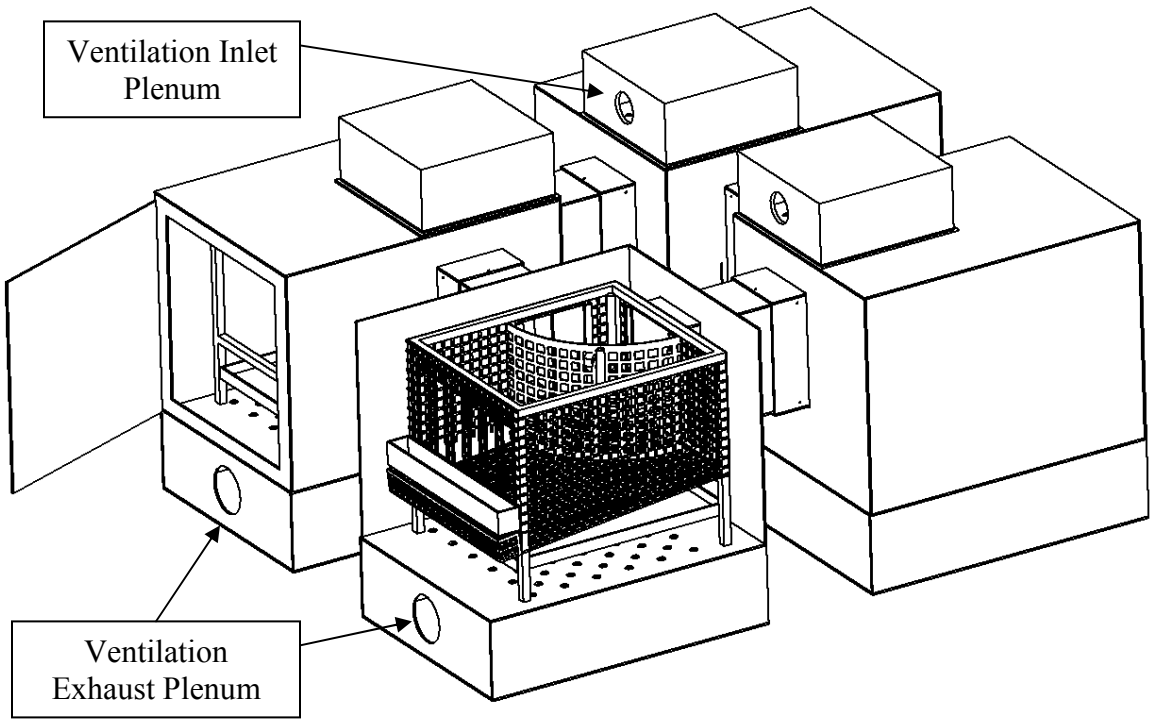

Figure 6.1: Schematic of environmental preference test chamber.

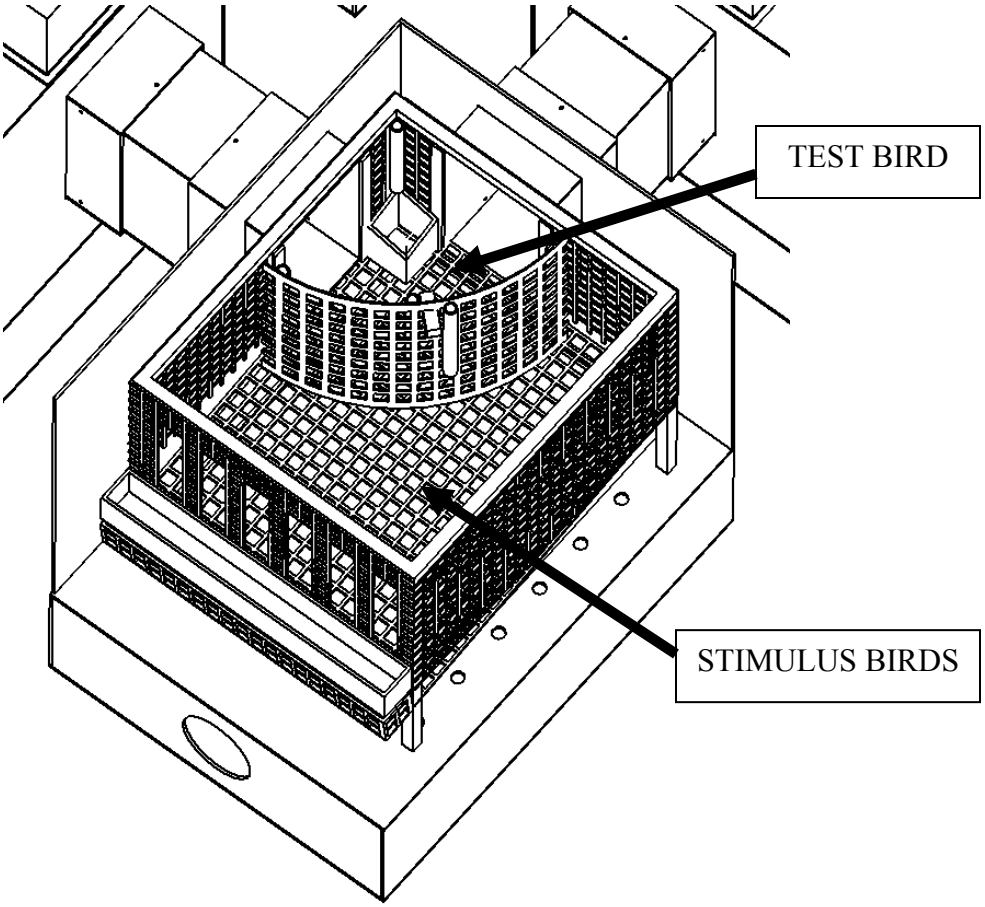

Figure 6.2: Schematic of one compartment housing one cage.

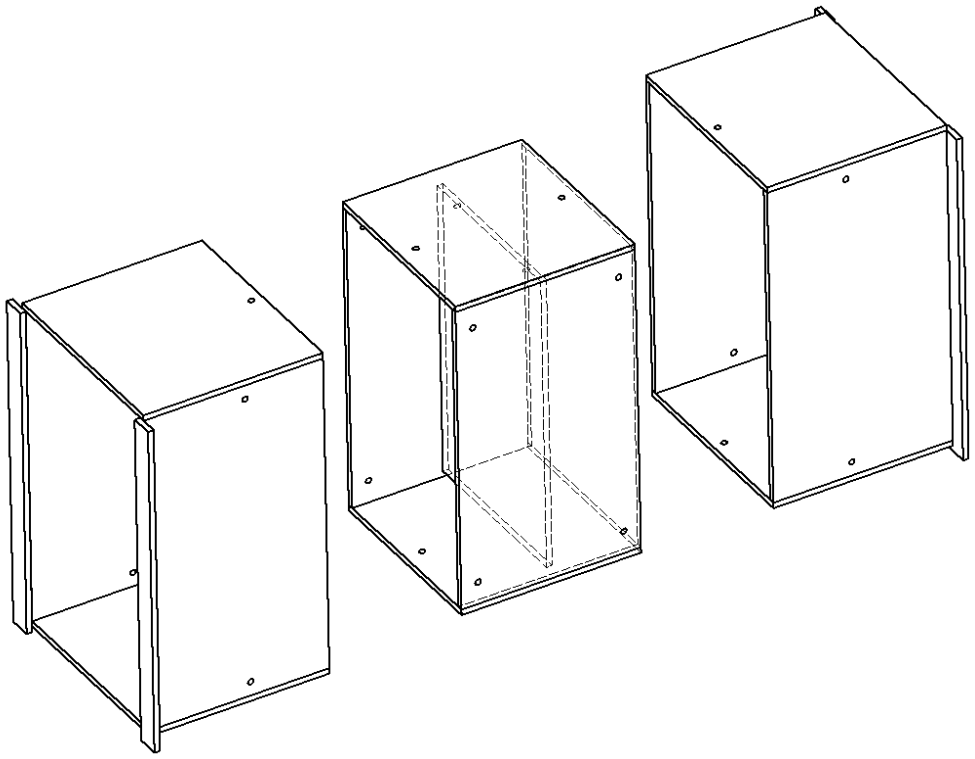

Figure 6.3: Schematic of door assembly for connection between cages

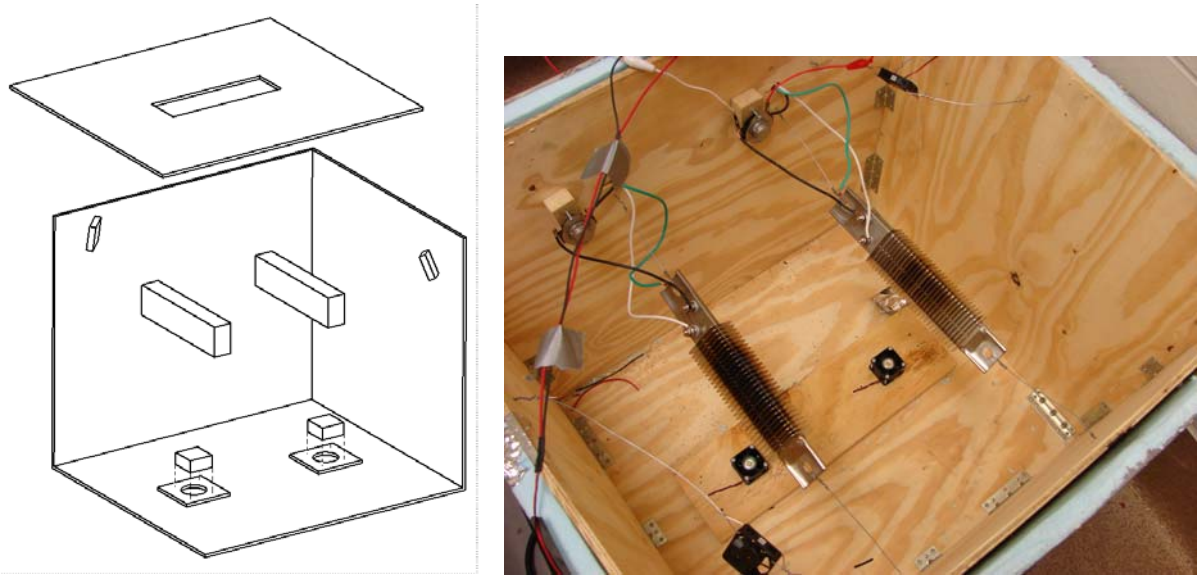

Figure 6.4: Interior view (schematic, left, and photographical, right) of mixing box showing ventilation supply fans, fin heaters, mixing fans, and temperature limit switches

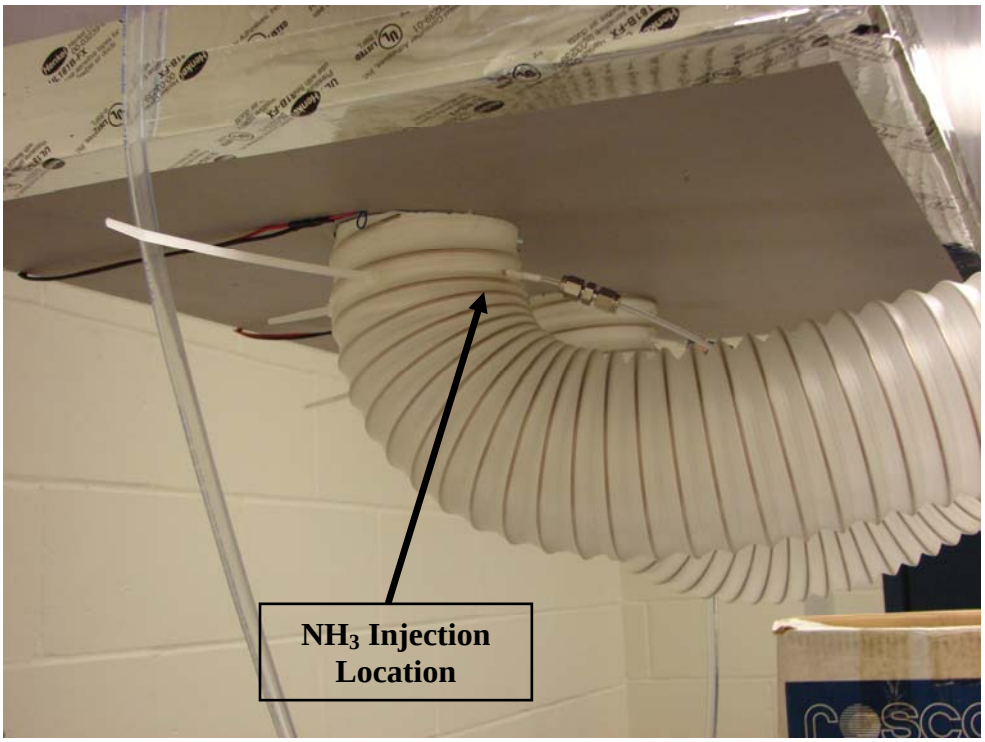

Figure 6.5: Ammonia injection into ventilation supply hose

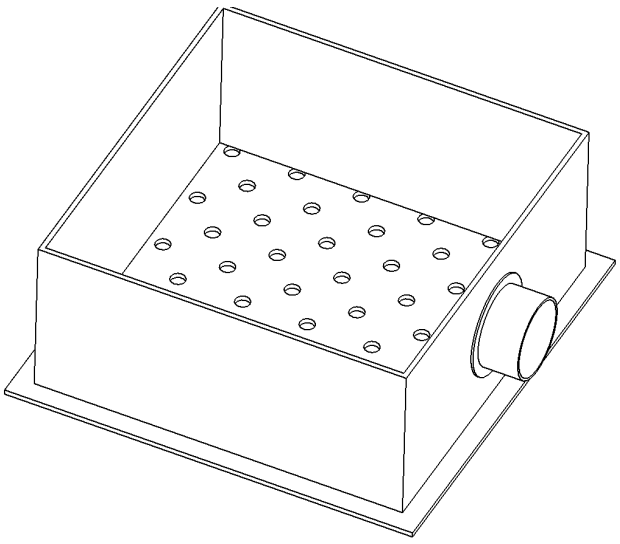

Figure 6.6: Schematic of inlet plenum

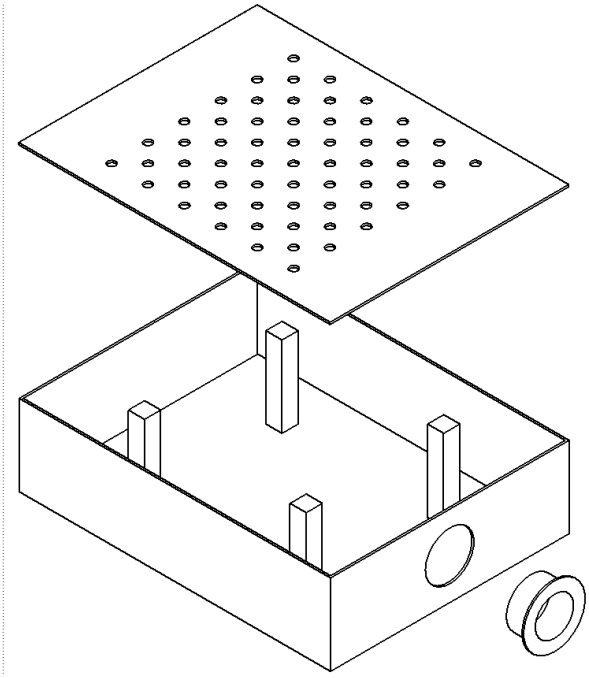

Figure 6.7: Schematic of exhaust plenum

Figure 6.8: Instrumentation for control system

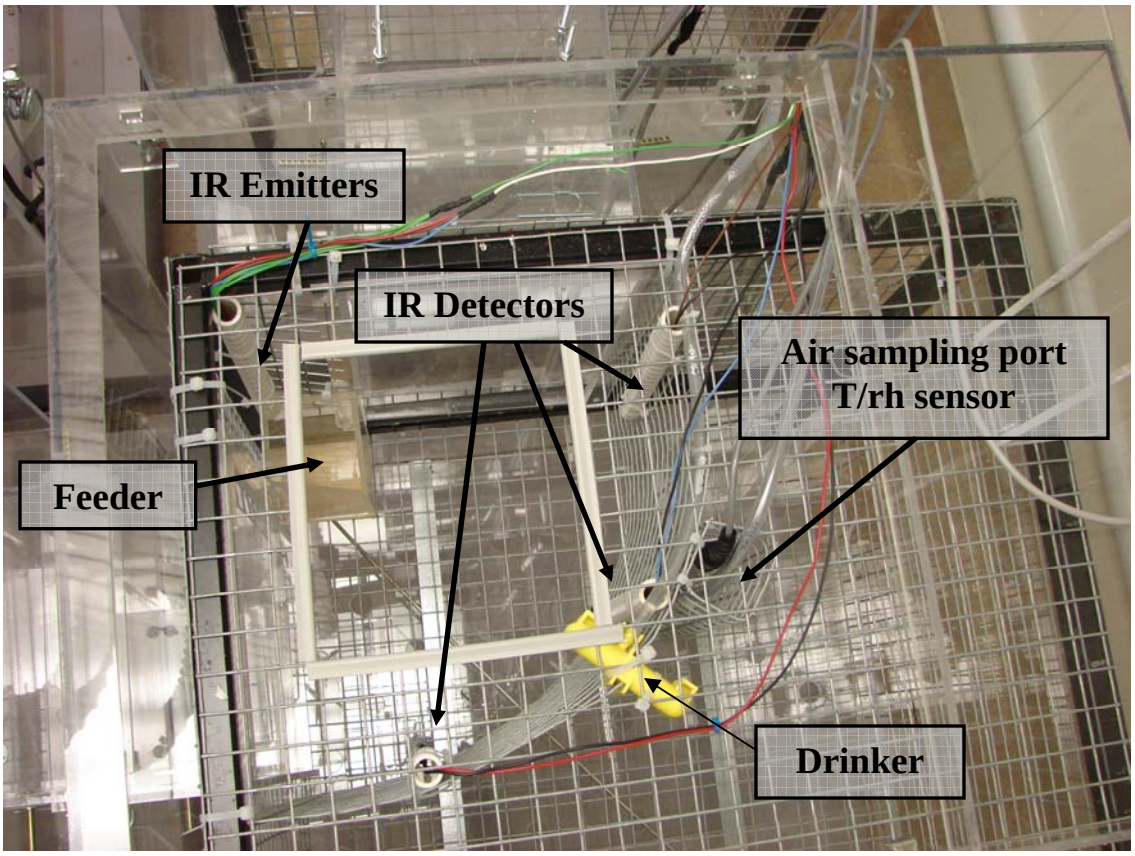

Figure 6.9: Top view of test hen area in one compartment.

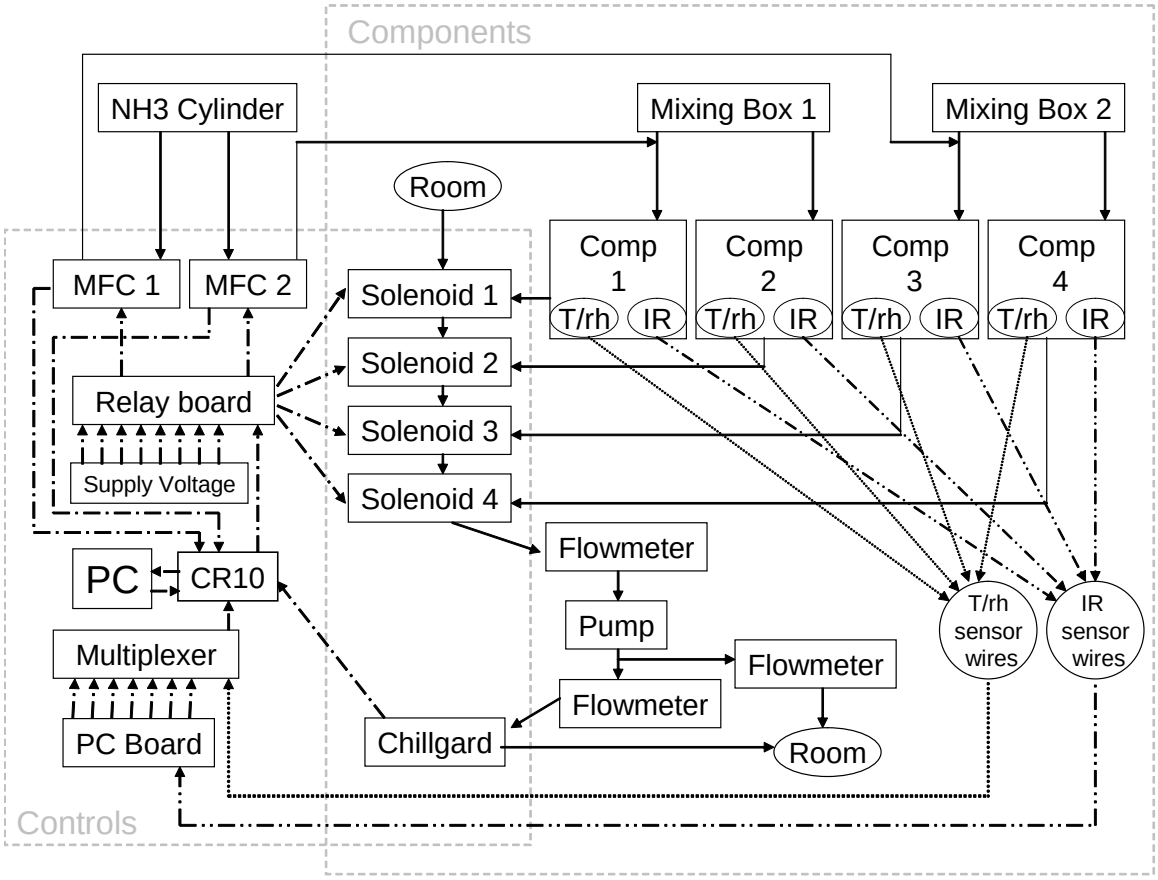

Figure 6.10: Block diagram of complete environmental preference test chamber system.

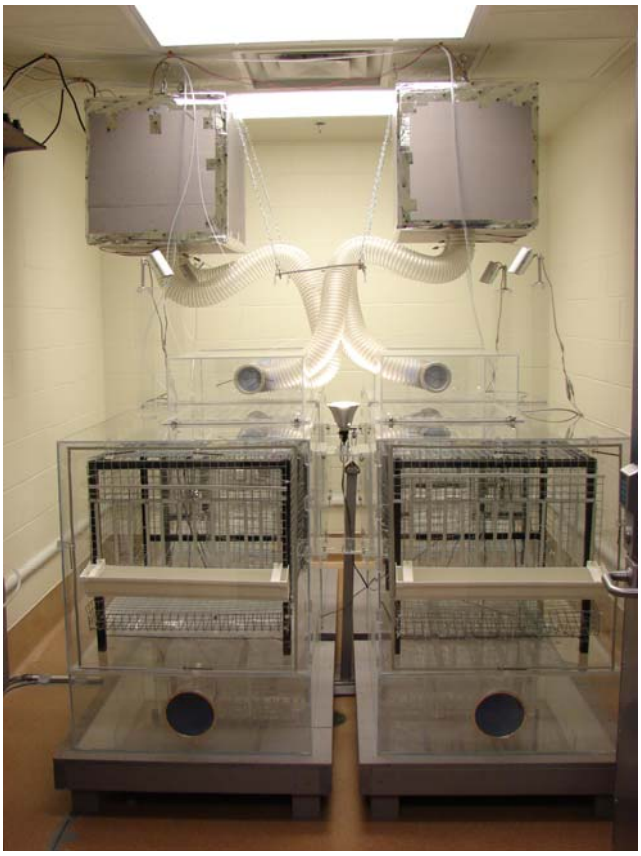

Figure 6.11: Photo of complete environmental preference test chamber

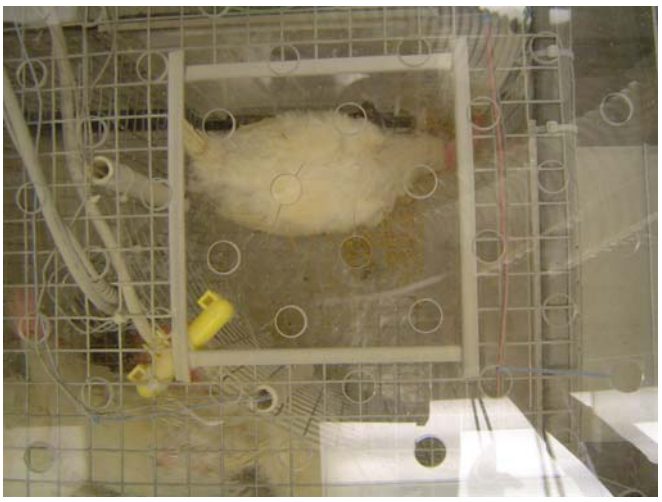

Figure 6.12: Top view of test hen inside environmental preference test chamber, with stimulus birds visible on other side of divider

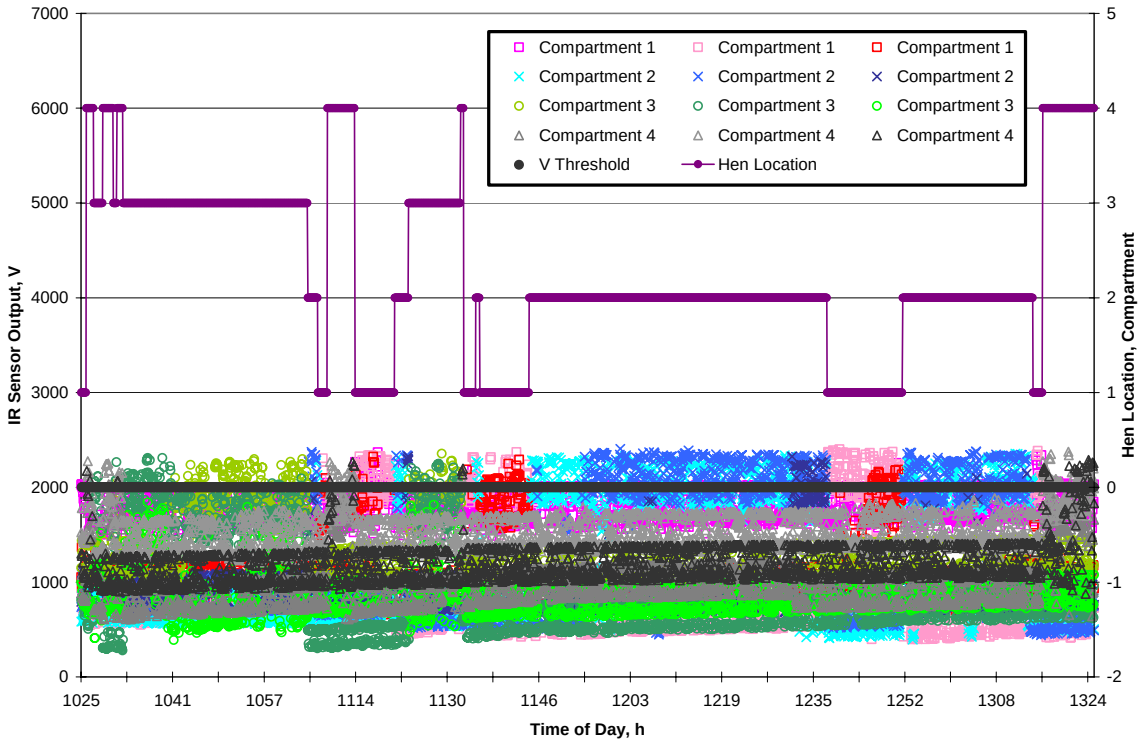

Figure 6.13: Sample comparison of IR sensor output versus verified hen location within laying hen environmental preference chamber, with marked voltage threshold for data processing algorithm.

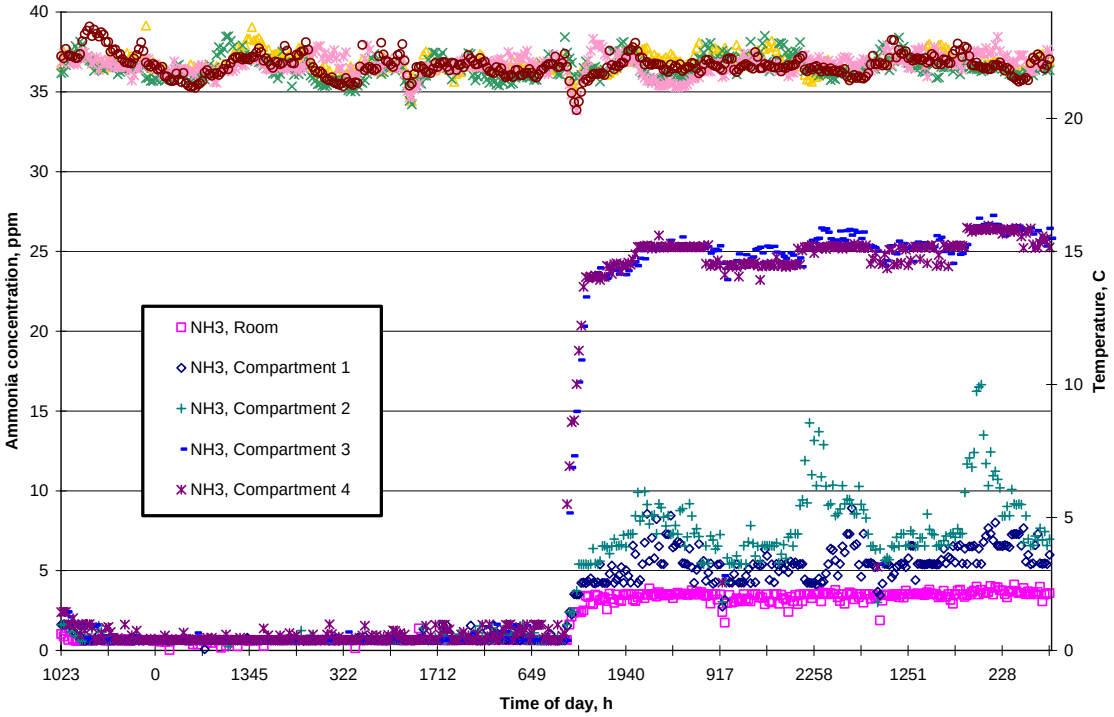

Figure 6.14: Sample compartment environments (during one complete trial) for baseline and treatment phases (3 day each) within laying hen environmental preference chamber.

Chapter 7

Summary and Implications

This dissertation attempts to supplement the existing knowledge base concerning laying-hen housing. Where possible, a systematic assessment approach was used in the comprehensive literature review, and combined field monitoring and controlled-environment laboratory studies.

The following is a summary of the studies conducted and the findings:

1. A comprehensive review of literature on current and emerging housing systems for laying hen production revealed positives and negatives for each housing system, and an initial attempt was made to summarize the comparison with a general ranking score for various areas of housing considerations. Traditional cages and free range systems yielded the most extreme rankings (both good and bad in almost equal prevalence), but the overall scores did not vary greatly for the housing systems. Equal importance rankings were applied to all parameters, which may or may not be appropriate. There are many unknowns and contradictions in the literature for enriched cages and cage-free barns that may affect the overall rankings. The literature review highlighted many areas of inadequate information in the literature for comparing housing systems and anticipating consequences of altering housing schemes. Studies described in the remainder of this dissertation were conducted to address some of these research areas.
2. A field observational study was conducted to assess environmental conditions (T, RH, CO_2, and NH_3) and bird health status in winter and summer for three

types of laying-hen housing in Iowa: a) cage-free floor-raised (FR), b) caged high-rise (HR), and c) caged manure-belt (MB). Differences in environmental conditions and/or pathogen frequency were observed among all three housing types during summer and winter conditions. During winter, NH_3 levels were much higher in the FR housing systems than in HR or MB systems. Air temperature in the FR houses also fluctuated more, following the outside temperature. The results also indicate seasonal differences among housing systems for prevalence of bacterial foodborne pathogens, but the results do not conclusively show that one system yields lower pathogen frequencies than another. Further studies should include multiple representations of each housing type, encompassing different management and housing configurations to better delineate the cause-effect relationships. Future studies should also consider collecting environmental, physiological, and production data collected periodically over an extended period of time (e.g., one year). One important finding in cage-type housing affects temperature control. The results indicate that it may be prudent to periodically monitor the cage interior temperature, and adjust the temperature setpoint, when necessary, to reflect the microenvironment that the birds are experiencing. Alternatively, consideration should be given to locating the thermostat temperature sensors near the bird microenvironment.

3. A series of controlled laboratory trials were conducted to quantify the bioenergetics (heat and moisture production or HMP) and thermoregulatory responses of W-36 laying hens to varying space allowances (348, 387, 465, or

581 cm^2/bird; 54, 60, 72, or 90 in^2/bird) and group sizes (8 or 16 birds/cage) in traditional cage houses under thermoneutral (24C) and heat challenging conditions (32 or 35C). This study affirms the need to further understand consequences of adopting new housing practices, such as reducing stocking density, on environmental control. Specifically, the results indicate a reduction in stocking density does not affect HMP on a bird mass basis. Therefore, reducing the number of birds in a given house would reduce the heat load, which may be beneficial in hot weather but could have adverse effects in cold weather. Based on bird condition and production results, further research is merited to quantify the impacts of varying stocking density and group size on management and bird health.

4. In the same study concerning the impact of stocking density and group size on laying hens kept in cages, the results imply that decreasing stocking density offers no clear benefit for coping with heat challenge of 32C and 35C on the basis of physiological responses of the hens and on economic impacts of production. The results also highlight the importance of including micro-environment in ventilation control schemes, because the temperatures within cages were higher than room temperatures for thermoneutral conditions.
5. An environmental preference test chamber (EPTC) system to assess responses of laying hens to different environmental factors was designed, constructed and tested. The EPTC consisted of four interconnected compartments with an area for a test bird to navigate between the compartments and an area in each compartment for a group of three birds to reside. The EPTC incorporated

automated environmental control for atmospheric ammonia concentration and an automated tracking system for location of the test bird. The automated tracking system yielded less than 5% error for compartment occupancy, but failed to identify quick moves through compartments due to sensor sampling rate. An introductory test on aversion responses of laying hen to atmospheric ammonia using the newly developed EPTC was carried out. The occupancy results revealed no preference for any compartment or treatment. Further investigation regarding hen usage of the compartment and correlations with behavior should be completed.

The results of the research presented provide science-based data regarding the impacts of different husbandry practices on housing environment and hen responses. These results may be considered by the egg industry and regulatory agencies in making more informed, science-based decisions toward modifying production practices. They also contribute to clarification of uncertainties that arise in engineering design for environmental control of laying-hen houses when conditions deviate from those under which the design data had been collected for the current handbooks (i.e., change in stocking density). The preference testing chamber system introduces a new tool that will aid future studies assessing hen perceptions of environment.