# Assessing Hen Response to Ammonia and Thermal Comfort Combinations via Preference Test

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## Introduction

Maintaining comfortable environmental temperatures in modern laying-hen houses during wintertime is typically accomplished by using minimum ventilation rates. The winter minimum ventilation is designed to provide moisture control and maintain appropriate indoor air quality. As barns are adjusted to provide a lower stocking density or designed as alternative systems (e.g., cage-free housing), the house will hold considerably fewer birds. Fewer birds result in less animal heat in the house, as such in the winter there is often a struggle between thermal comfort and indoor air quality. Because the minimum ventilation rate for air quality control is likely greater than the ventilation desired to maintain thermal comfort, the house will encounter one of the conditions: a) a cooler environment with good indoor air quality, resulting from elevated ventilation with no or limited supplemental heating (to conserve energy); or 2) a comfortable environment with relatively poor indoor air quality, resulting from reduced ventilation rate.

The thermoneutral zone (TNZ) described by Sainsbury (2000) for hens covers a significant range from 12 to 24 °C, while Hy-Line Management Guide (2007) suggests a narrower range of 20 to 25 °C. In laying-hen housing, ammonia is a major contributor to air quality concerns (Carlile, 1984). Low ventilation rate will result in increased ammonia levels, which may affect the health and welfare of laying hens and the human occupants. In its guidelines for laying hen welfare, the United Egg Producers (UEP) has recommended the indoor ammonia concentration be less than 10 ppm and not exceed 25 ppm (UEP, 2002). Our preliminary lab study supports the literature findings that ammonia concentrations of 25 ppm or greater are aversive to hens (Kristensen et al., 2000).

The objective of this study was to determine if a low (<10 ppm) ammonia condition with a cool air temperature below the optimal performance range (18.3 °C; 65 °F) is preferred to an ammonia concentration known to be aversive (30 ppm) combined with thermoneutral (TN) air temperature (23.9 °C; 75 °F ). For this study, the cool temperature and low ammonia will be known as the "low" condition for the remainder of this report, whereas the TN temperature with high ammonia will be known as the "high" condition. A similar previous study with pigs found that pigs would choose ammonia comfort to a point. When the temperature dropped below the pigs' lower critical temperature, they switched their preference to thermal comfort (Jones et al., 1999). The 'cool' temperature in this study falls below optimal performance, and ideally it should be slightly below the lower critical temperature. Our hypothesis was that the hens would choose ammonia comfort over thermal comfort.

## Materials and Methods

The experiment was run with the environmental preference test chambers (EPTC) originally developed by Green and Xin (2008) and refined by the authors. The EPTC had four compartments (fig. 1). Two mixing boxes suspended above the compartments provide temperature-controlled air to the four compartments. In order to provide a cool temperature condition of 18.3 °C (65 °F), the air supply for the mixing box was given primary access to the room inlet and a 4100 Watt (14000 BTU/hr) portable air conditioning system was installed to supply air in the mixing box. In the warm air mixing box two heating fins were used together with small mixing fans running to ensure mixed air with more uniform warm air temperatures. The compartments had separate air supplies with airflow rates ranging from 9.5 to 15.7 $m^3$/hr to satisfy the airflow and temperature control needs of the 'low' and 'high' compartments. Ammonia (10%, balanced with air), as needed, could be injected into the airstreams of individual compartments to achieve the target ammonia levels in each compartment. The EPTC had doorways in the passages between compartments to limit air exchange between compartments. Although each compartment had doorways to both adjacent compartments, the EPTC was divided to provide two testing environments. The doorways between compartments 2 and 3 and between 1 and 4 were closed and sealed to prevent airflow. The clear solid acrylic doors were also covered with opaque plastic to provide a visual barrier. In this way, one bird could be tested in compartments 1 and 2 while a second bird could be tested simultaneously and independently in compartments 3 and 4. Each compartment was divided into an area for stimulus birds that remained in the compartment and an area for a test bird that was free to move between two compartments. An infrared (IR) detection system was used to determine in which compartments the test bird resided. The IR detectors took readings every 2 seconds.

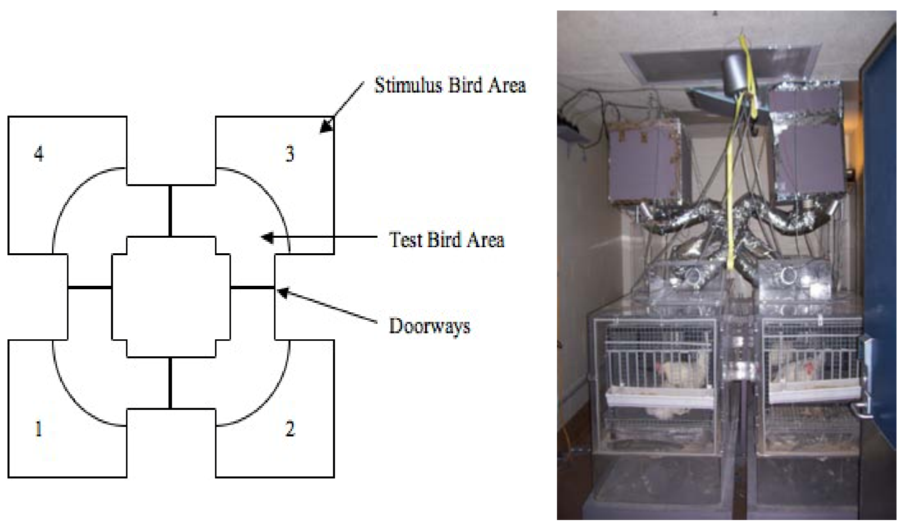


Figure 1. Top view of the EPTC with the four compartments labeled (left) and a picture view of compartments 1 and 2 of the EPTC with air tempering and mixing boxes (right).

This experiment was run with fifteen (15) Hy-Line W-36 hens at 26 to 35 weeks of age. The pullets were acclimated at 21°C and <5 ppm ammonia conditions for a few weeks prior to testing. The housing during acclimation consisted of two adjoining chambers with an identical door to the ones joining the EPTC compartments. The test bird had individual feeder and waterer in each compartment. Individual feeder in each compartment was weighed at the beginning and end of the treatment periods to determine the feed use. Water usage was handled in the same manner with a water bucket attached to the ceiling so that gravity could apply pressure for nipple drinkers in each compartment.

Two test birds were randomly selected every week for testing in the EPTC. The birds were first provided baseline condition of low ammonia with TN temperature (<5 ppm ammonia, 22.8°C). The birds were then subjected to the high and low treatment conditions. The high condition consisted of a temperature of 23.9°C (75°F) and 30 ppm ammonia, whereas the low condition consisted of <10 ppm ammonia with a cooler temperature of 18.3°C (65°F). The treatments were assigned in a randomized block. The birds were given at least 3 hours to acclimate to the EPTC and observed to confirm movement in each compartment. Data were taken for 3 d at baseline conditions, 2 d at the two test conditions, and 2 d with the conditions switched to the opposite compartments. When the conditions were switched, manure was removed, feed and water weighed and replenished, and eggs collected.

The amount of time spent in each compartment as well as the number of movements was calculated using data from the IR sensors. The sensor output was processed to create a summary data set with total time and percentage of occupation time in each compartment and feed and water utilized by the test bird in each compartment. The summary data set was analyzed using SAS PROC MIXED to determine if the first and second treatment applications were significantly different. As well the same command evaluated if the compartmental baseline preferences were related to the test condition compartmental choices. SAS PROC T-TEST was used to determine if the high and low conditions were significantly different. Effects were considered significant at $\alpha \leq 0.05$.

## Results

The first analysis was completed to determine if responses of the birds between the first and second applications of the conditions were significantly different. The difference in time of occupation between high condition and low condition was 16.4 (±8.6 S.E.) minutes per hour for the first application, and – 8.5 (±10.8 S.E.) minutes per hour for the second application. Although the difference was not significant at the 0.05 level due to the large variances, the probability ($p=0.09$) was close to the borderline, hence deserving further evaluation. The results below were further broken down into treatment application 1 and 2, but as none were significantly different, the average of the two applications is reported.

The second analysis evaluated if the baseline compartmental choice influenced the preference when environmental conditions were applied. When analyzed these values showed a strong relationship between compartmental choices at baseline and their compartmental choice when the test conditions were applied ($p=0.021$). The majority of the test birds (9 out of 15) showed the same compartmental preference in both treatment applications that they showed in the baseline period. One test bird preferred the low condition while two test birds preferred the high condition in both treatment applications.

In order to remove compartmental preference, t-test was used to determine if the difference in compartment occupation time between the low condition and the baseline was significantly different from zero. When this difference is significantly different from zero, a negative value means the high condition was preferred and a positive value means the low condition was preferred. Because the occupation data were normalized to a percentage of time (POT), the absolute differences of low minus baseline and high minus baseline are equal with opposite signs (equations 1 and 2). The average difference in POT between the low compartment and the same compartment at baseline was -1.7±6% (mean±SE, p=0.51). During the 16-hr light period (05:30 - 9:30 h) the difference was -3.8±7% (mean±SE, p=0.51). The results indicate that these birds did not make a clear choice between the two conditions provided. Figure 2 shows the overall mean and standard error of minutes per hour of occupation time spent by the 15 birds in the high and low treatment, feed and water usage. The difference in feed usage between the low condition and baseline was 4.08±7.0 g/d (p=0.49); and the difference in water usage between the low condition and baseline was 6.4 ±9.3 g/d (p=0.53).

$$POT_{low} + POT_{high} = 100\% = POT_{baseline\_low} + POT_{baseline\_high} \quad [1]$$

$$POT_{low} - POT_{baseline\_low} = -(POT_{high} - POT_{baseline\_high}) \quad [2]$$

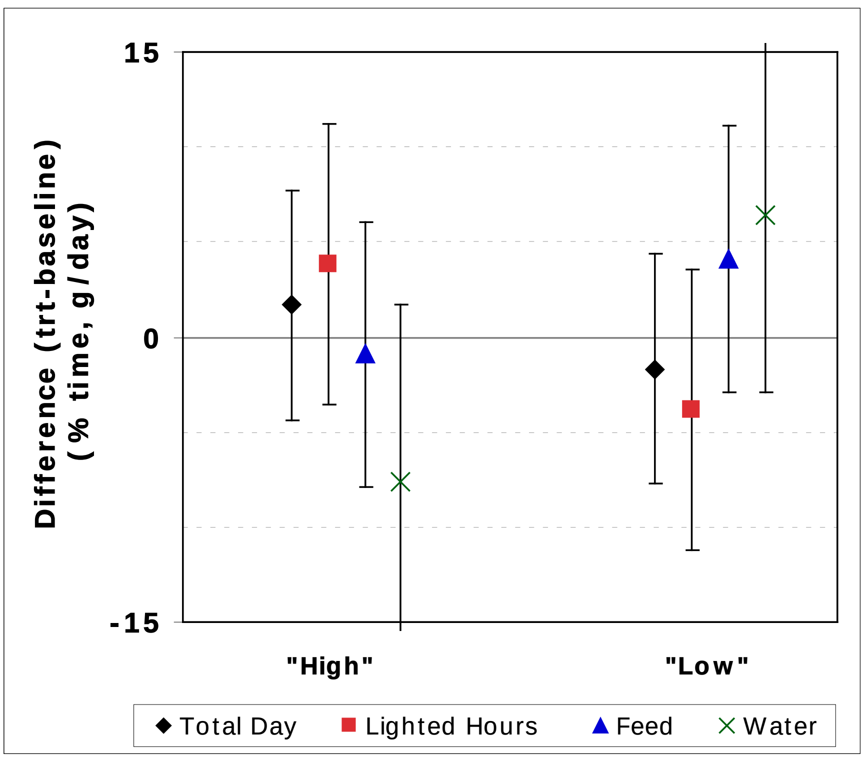


Figure 2. The total day, lighted hours (16 hr/d) difference in time spent in each treatment condition vs. the same compartments at baseline (n=15) with standard error, as well as the feed and water difference. The low environment was <10 ppm ammonia and 18.3°C, while the high condition was 30 ppm ammonia and 23.9°C.

Table 1. Percent of occupation time spent by the hen under baseline and low testing conditions as determined using the EPTC system (low condition = <10 ppm ammonia and 18.3°C); **Bold** values are >100 movements, shaded cells note where hens switched their compartmental preference from baseline to condition application periods.

| Test Bird | Trt * | Period Applied | Time in Low Compartment | | Number of Movements | | Baseline (corresponding to low) | | Difference (low-baseline) | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | total | *light* | total | *light* | total | *light* | total | *light* |
| 1 | 1H2L | 1 | 97.3% | *95.9%* | 44 | *44* | 83.2% | *89.8%* | 14.1% | *6.1%* |
| 1 | 2H1L | 2 | 1.4% | *1.4%* | 22 | *18* | 16.8% | *10.2%* | -15.4% | *-8.8%* |
| 2 | 3H4L | 1 | 0.9% | *1.4%* | 26 | *26* | 0.8% | *1.2%* | 0.1% | *0.2%* |
| 2 | 4H3L | 2 | 87.8% | *90.0%* | 24 | *18* | 99.2% | *98.8%* | -11.5% | *-8.8%* |
| 3 | 2H1L | 1 | 59.6% | *37.5%* | 52 | *52* | 57.1% | *38.9%* | 2.5% | *-1.4%* |
| 3 | 1H2L | 2 | 68.5% | *53.0%* | **147** | ***143*** | 42.9% | *61.1%* | 25.6% | *-8.1%* |
| 4 | 4H3L | 1 | 31.8% | *24.2%* | 43 | *42* | 26.3% | *35.6%* | 5.6% | *-11.4%* |
| 4 | 3H4L | 2 | 58.8% | *62.7%* | 96 | *95* | 73.7% | *64.4%* | -15.0% | *-1.8%* |
| 5 | 2H1L | 1 | 6.2% | *4.7%* | 21 | *18* | 4.5% | *7.9%* | 1.7% | *-3.2%* |
| 5 | 1H2L | 2 | 78.8% | *68.8%* | 42 | *42* | 95.5% | *92.1%* | -16.7% | *-23.3%* |
| 6 | 4H3L | 1 | 70.9% | *73.7%* | 59 | *59* | 61.3% | *73.5%* | 9.6% | *0.2%* |
| 6 | 3H4L | 2 | 25.2% | *-14.4%* | 41 | *41* | 38.7% | *26.5%* | -13.6% | *-40.8%* |
| 7 | 3H4L | 1 | 26.7% | *40.7%* | 91 | *91* | 26.5% | *38.6%* | 0.2% | *2.1%* |
| 7 | 4H3L | 2 | 95.9% | *93.9%* | 59 | *55* | 73.5% | *61.4%* | 22.4% | *32.5%* |
| 8 | 2H1L | 1 | 40.5% | *57.2%* | 94 | *88* | 74.1% | *77.7%* | -33.7% | *-20.4%* |
| 8 | 1H2L | 2 | 86.8% | *79.9%* | 40 | *38* | 25.9% | *22.3%* | 61.0% | *57.6%* |
| 9 | 4H3L | 1 | 22.3% | *3.9%* | 72 | *56* | 52.3% | *61.0%* | -30.0% | *-57.1%* |
| 9 | 3H4L | 2 | 81.6% | *96.2%* | 67 | *52* | 47.7% | *39.0%* | 33.8% | *57.2%* |
| 10 | 1H2L | 1 | 26.3% | *39.5%* | **105** | ***105*** | 29.6% | *27.1%* | -3.3% | *12.4%* |
| 10 | 2H1L | 2 | 33.7% | *-4.6%* | **180** | ***180*** | 70.4% | *72.9%* | -36.8% | *-77.5%* |
| 11 | 3H4L | 1 | 74.2% | *85.7%* | 61 | *43* | 30.3% | *46.7%* | 43.9% | *39.0%* |
| 11 | 4H3L | 2 | 5.3% | *7.8%* | 39 | *30* | 69.7% | *53.3%* | -64.4% | *-45.5%* |
| 12 | 1H2L | 1 | 88.8% | *91.3%* | 19 | *12* | 55.5% | *61.0%* | 33.3% | *30.3%* |
| 12 | 2H1L | 2 | 29.2% | *19.3%* | 33 | *26* | 44.5% | *39.0%* | -15.2% | *-19.6%* |
| 13 | 3H4L | 1 | 40.3% | *60.5%* | 55 | *49* | 41.7% | *32.3%* | -1.4% | *28.1%* |
| 13 | 4H3L | 2 | 16.7% | *24.2%* | **120** | ***116*** | 58.3% | *67.7%* | -41.6% | *-43.5%* |
| 14 | 2H1L | 1 | 4.7% | *7.2%* | 5 | *5* | 26.7% | *40.5%* | -22.0% | *-33.3%* |
| 14 | 1H2L | 2 | 94.7% | *92.4%* | 7 | *7* | 73.3% | *59.5%* | 21.4% | *32.9%* |
| 15 | 4H3L | 1 | 1.7% | *2.5%* | 5 | *3* | 0.8% | *1.2%* | 1.0% | *1.3%* |
| 15 | 3H4L | 2 | 91.5% | *88.2%* | 27 | *23* | 99.2% | *98.8%* | -7.7% | *-10.6%* |
| | **Mean** | | **48.3%** | ***46.2%*** | **57** | ***53*** | | | **-1.7%** | **-3.8%** |
| | **SE** | | **6.3%** | **6.9%** | **8** | **8** | | | **5.0%** | **5.9%** |

*** TRT notation 1H2L indicated compartment 1 is the "high" and compartment 2 is "low" condition**
**Table focuses on "low" condition. Time in compartment and corresponding baseline for the 'high' condition are (100-displayed, %). The difference of high minus baseline is - (low-baseline).**

Another measurement evaluated in this study was the number of movements each test bird made during the periods when the conditions were applied (table 1). The overall number of movements for the treatment application period (~48 hours) was 57±8 (mean±SE). The periods with very high numbers of movements (>100) are shaded in the table. There were four such periods. Three of them corresponded to the hen's switching of her preference from baseline to testing conditions. Each time a hen switched a compartmental preference from baseline to the treatment it was noted in the table as a bold number. There are seven such periods during this study. There seems to be an indication (by the large number of movements) that some birds were having a difficult time choosing a compartment.

**Discussion**

The primary issue addressed in this study was if the birds would choose thermal comfort or better air quality. The data suggested no clear preference, as evidenced by the time spent under test conditions, feed and water usage. The hens showed strong compartmental preferences that they developed during baseline conditions, which carried through as test environmental conditions were applied. Birds that did change compartmental preference with environmental conditions often had more frequent movements possibly indicating difficulty choosing.

This study does tell us that the birds will not avoid a cool condition down to 18.3°C (65°F) when combined with better air quality. This means that from the hens' comfort perspective, there is not a reason to avoid ventilation for improved indoor air quality in the winter as long as the indoor temperature can be maintained at or above 18.3°C (65°F).

**References**

Carlile, F. S. 1984. Ammonia in poultry houses: A literature review. World's Poult. Sci. J. 40:99-113.

Green, A.R. and H. Xin. 2008. Development of a novel environmental preference test system for laying hens and its initial application to assess hen aversion to atmospheric ammonia. ASABE Paper No. 084451. St. Joseph, Mich.: ASABE.

Hy-Line. 2007. Hy-Line Commercial Management Guide. W-36. 2007-2008. West Des Moines, IA.

Jones, JB, AJF Webster, CM Wathes. 1999. Trade-off between ammonia exposure and thermal comfort in pigs and the influence of social contact. Anim Sci. 68: 387-398.

Kristensen, H.H., L.R. Burgess, T.G.M. Demmers, and C.M. Wathes. 2000. The preferences of laying hens for different concentrations of atmospheric ammonia. Appl. Anim. Behav. Sci. 68: 307–318.

Sainsbury, D. 2000. *Poultry Health and Management*. London, UK.

United Egg Producers (UEP). 2002. United Egg Producers' Animal Husbandry Guidelines. Available at: http://www.uepcertified.com/program/guidelines/1999