

# Welfare Evaluation by Image Analysis of Laying Hens in Different Breeding Systems and Environmental Conditions

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**Abstract.** *This work aimed to evaluate aspects related to behavior and welfare of hens comparing a conventional housing system (battery cages) with another system using litter, lair and roost. In addition, the effects of environmental conditions (stress or comfort) on final egg quality and behavior responses (analyzed by precision techniques) were evaluated using two laying hen lines. Two groups of 20 birds (10 Hy-line W36 and 10 Hy-line Brown) at the beginning of the production phase were submitted to two environmental conditions (26°C with 60% RH or 35°C with 70% RH) and two housing systems (battery cages or litter) during two consecutive weeks. During the evaluation period, bird behaviors were recorded by video cameras. Eggs produced during the whole experimental period were analyzed according to these quality parameters: egg weight, shell thickness, specific gravity and Haugh unit. A significant ( $P < 0.05$ ) reduction in quality parameters was observed in eggs from laying hens raised under heat stress, mainly in those from the cage system. In relation to behavior, the housing system in litters proportioned the expression of all natural behaviors and those related to bird comfort, which permitted to improve welfare conditions. In the conventional system (laying cages), birds tried to manifest their natural behaviors in spite of lacking conditions for it. This impossibility for executing natural behaviors certainly promoted an increase of stress originated by this kind of housing system.*

**Keywords.** *Poultry welfare, Egg quality, Breeding systems, Image analysis*

## Introduction

In beneficence of laying hen welfare, a recent legislation is bounding the actual breeding system (battery cages) and its substitution through a set of changes in terms of building and handling aiming to adequate the laying hen lines to the new marked models.

Nowadays, laying hens are submitted to a very high confinement degree, which imposes severe restriction to natural animal activities. Normally, cages measuring 30 to 35 cm of width and 43 cm of height house 3 to 5 birds. Under this condition, birds can not stretch their wings or move without touching each other or stand up completely in cage (Singer, 1991).

Comparing studies about behavior of wild and domestic birds in environment controlled by human indicated that, in general, behavioral repertory of birds in non-confined environment is preserved, being observed changes only in frequency and intensity of behavioral traits (Craig, 1992).

It is well reported by studies carried out by Jensen and Toates (1993) that the highest problem of animals raised in confinement (cages) is the impossibility of natural behavior expression, which leads to frustration and development of anomalous behavior.

Nowadays, image analysis is a tool that has been considered very useful in studies about animal behavior. According to Xin et al. (1998), it is a system composed by a microcamera, image capture plate associated with a PC and a visual program for acquisition, processing and classification of animal images.

Leroy et al. (2003) developed a system for image analysis using a technique of on-line processing that quantify laying hen behavior for further comparisons with human view observation system. They concluded that both analysis systems might be used for evaluating bird behavior although human view observation system had resulted in a larger number of mistakes committed during total quantification of behaviors when compared to automated video system.

Production quality, which is being each more desirable by bird producers and consumers around the world, is being associated to animal welfare rules and is directly related to factors as hygiene, sanity and mainly animal health.

Thus, many studies have carried out aiming to test the effects of environment (temperature and relative humidity) on egg quality Mashaly et al. (2004). In Bennion and Warren (1933) trial with laying hen submitted to high temperatures, eggshell quality was affected by high temperature and values of egg constituent weight were reduced.

However, few researches were realized for verifying the effect of breeding system on egg quality parameters when laying hens are exposed to conditions of comfort or heat stress.

So, this work aimed to evaluate parameters related to animal behavior of laying hens raised in a litter system in substitution to a conventional system (cages), as well as the effects of environmental conditions on quality parameters of eggs produced in both systems.

### Material and Methods

A trial was conducted in a climate chamber at Animal Environment Research Nucleus (NUPEA), Department of Rural Engineering, ESALQ/USP. Forty 21 week-old laying hens, 20 of Hy-Line W36 (white eggs) and 20 of Hy-Line Brown (brown eggs), were evaluated.

For each environment condition, comfort or stress, 10 birds of each line were distributed in both breeding systems.

#### Treatments.

**Bird exposition to controlled environment (climate chamber):** Birds of each line and breeding system were exposed to two environmental conditions, one of thermal comfort and other of heat stress, according to following descriptions:

**Treatment A1** – Exposition to temperature of  $26^{\circ}\text{C} \pm 2^{\circ}\text{C}$  and  $60\% \pm 2\%$  of relative humidity (thermal comfort condition);

**Treatment A2** – Exposition to temperature of  $35^{\circ}\text{C} \pm 2^{\circ}\text{C}$  and  $70\% \pm 2\%$  of relative humidity (heat stress condition).

All birds were exposed to both treatments during 14 consecutive days in a controlled environment, according to Sevegnani et al. (2001). But, there was a gradual adaptation period, totalizing 21 days of exposition to controlled environment for each treatment.

**Breeding system:** Birds were submitted to two breeding systems for comparing their effect on bird welfare and production quality.

**Treatment C1** – Breeding system with litter and lair (alternative)

**Treatment C2** – Breeding system in cages (conventional)

#### Evaluated Parameters

**Climatic parameters:** Values of Temperature of dry bulb (Tdb), relative humidity (RH) were taken inside of climate chamber using data loggers. These loggers were installed at 1.70, 1.50 and 0.50 m of height from chamber floor. Readings were taken in intervals of 15 minutes during 24 h. Based on climate parameters, process enthalpy was calculated allowing a better characterization of comfort and heat stress intervals. Equation for enthalpy estimation was that described by Villa Nova (1999), cited by Barbosa Filho (2005):

$$H = 6.7 + 0.243 \times \text{Tdb} + \left\{ \frac{\text{RH}}{100} \times 10^{\frac{7.5 \times \text{Tdb}}{237.3 + \text{Tdb}}} \right\}$$

where: H = enthalpy (kcal/kg dry air); Tdb = environmental dry-bulb temperature ( $^{\circ}\text{C}$ ) and RH = relative humidity (%).

**Behavior parameters:** For behavior analyses, all birds were individually identified by a dorsal painting with a non-toxic ink, Rudkin and Stewart (2003), which allow following and analyzing behavior of each bird.

Images were taken using video cameras fixed to climate chamber's roof than were registered at each second during 1 hour in the morning (from 10:00 to 11:00 a.m.) and 2 hours in the afternoon (from 1:00 to 2:00h p.m. and from 4:00 to 5:00 p.m.) for two consecutive days within each environment condition and breeding system. Recorded images were managed by VIDEOCAP 5.1 software, being stored a data bank. Behavior data were not registered during the night.

After period of image capturing, they were analyzed by visual method, which means, without software for interpretation or image analysis. Thus, images were analyzed always by the same person for each poultry behavior and each day period. After, percentages of time for expression of each behavior were estimated, as well as duration of each behavior, in minutes and seconds.

Behavioral patterns were evaluated, according to Rudkin and Stewart (2003) taking into account activities developed by each bird:

**Eating** – characterized when bird was with the head inside the feeder.

**Drinking** – characterized when bird was pecking the Nipple drinker.

**Feather pecking** – non-aggressive behavior characterized when bird investigate its own feathers or other bird's feathers using beak.

**Dust-bathing** – common bird behavior characterized by a sequence of earth scratching and sprinkling sand over the own body, beside fast movements of shaking legs.

**Comfort movements** – are those movements considered when birds are within a condition of comfort and welfare, as wing beating and stretching and shaking legs.

**Scratching** – considered an intrinsic bird behavior characterized when birds exploring their territory using their beak and feet.

**Perching** – is associated to bird welfare due to be an original behavior and is characterized by the bird permanence on a place localized over the soil level.

**Aggressiveness** – behavior related to dominance establishment within the group or to stress, being characterized, in general, by fast and strong pecks on crest or other head locals.

**Sitting** – characterized when bird sits down on litter or other substrate.

**Looking for nest** – considered a pre-laying behavior, is easily observed and characterized by fast and frequent entrances in lair as a tentative to evaluate the area before laying eggs.

**Laying** – characterized when bird stays in its lair sitting down over eggs.

**Standing** – characterized when bird does not show any movement or apparently does not fit within any behavior described above.

#### **Egg quality parameters:**

**Egg weight** – for measuring total egg production during whole experimental period, eggs were weighted (g) in an analytic balance (Gehaka BG 2000 model).

**Specific gravity** – specific gravity was determined according to Voisey and Hunt (1974). Serial dilutions of saline solution were done from low to high concentrated solution. Eggs stayed in each saline solution until to fluctuate, when they were removed and the density was registered according to saline solution concentration.

**Eggshell thickness** – measured without removing inner membranes of eggshell using a precision micrometer of Mitutoyo Dial Thickness Gage model with scale of 0.01mm.

**Haugh unit**– after weighting, eggs were broken and its content (white + yolk) was placed on a glass plane graded clean surface for measuring albumen height (mm) by reading the value indicated by micrometer AMES S-6428 model. Egg weight (g) and albumen height (mm) values were used in equation proposed by Pardi (1977) for Haugh unit estimation:  $HU = 100 \log (h + 7.57 - 1.7W^{0.37})$ ; where: h = albumen height (mm) and W = egg weight (g).

**Experimental design** - Experimental design was a factorial 2 x 2 x 2, with 2 thermal conditions (Comfort and stress) x 2 laying hen lines (Hy-Line W36 and Hy-Line Brown) and 2 breeding systems (litter+lair and cages). Each bird was considered an experimental unit. Statistical analyses was performed by SAS (1992)® software and means were compared by Tukey test (P<0.05).

## **Results and Discussion**

### **Climate Parameters.**

**Enthalpy:** Figures 1 and 2 show Enthalpy (KJ / Kg dry air) found for comfort and stress conditions, as well as their respective limiting values.

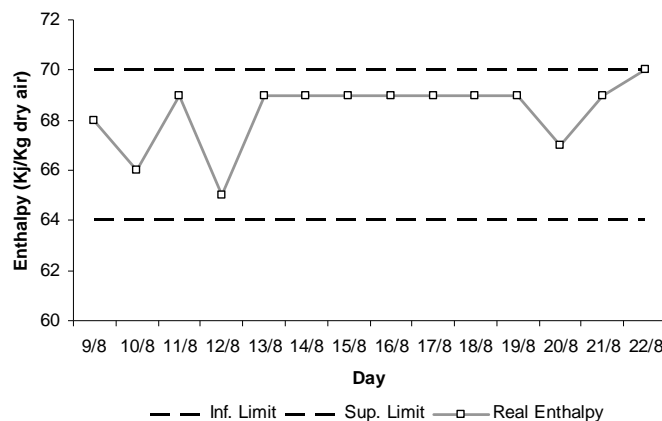


Figure 1. Values of enthalpy inside the climate chamber at comfort condition (Real Enthalpy) and ideal variation with the inferior and superior limits of bird comfort.

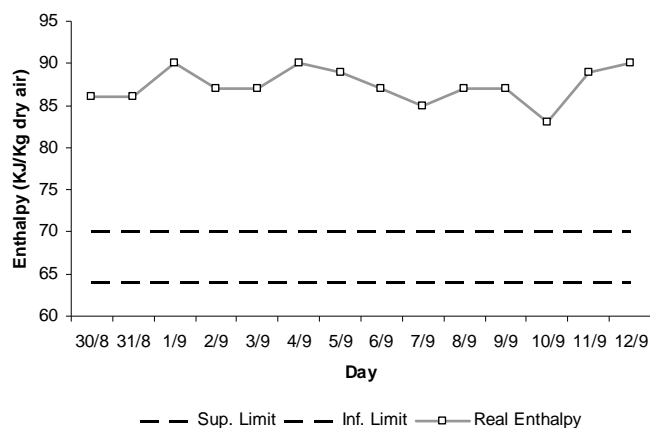


Figure 2. Values of enthalpy inside the climate chamber at stress condition (real enthalpy) and ideal variation with the inferior and superior limits of bird comfort.

Differences between both evaluated conditions might be visualized by Figures 1 and 2. In Figure 1, mean values of energy amount in dry air mass are within limits of comfort zone. Analyzing Figure 2, this same response was not found for stress condition, charactering enthalpy values higher than those of comfort index and, consequently, a situation of heat stress.

#### **Behavior Parameters.**

According to Johnson and Hogg (1996), bird monitoring has particular problems once some unpredictable behaviors might be suddenly executed by birds, which may difficult analyses. Comparing bird lines (Table 1), we found that Hy-Line Brown, during morning period and in relation to Hy-Line W36, spent more percentage of time eating, drinking, stretching, laying and dust-bathing. On the other hand, Hy-Line W36 spent more time perching and laying. During afternoon, the line difference occurred because of expression of behaviors as sitting and aggressiveness, being this last executed only by Hy-Line W36.

Another interesting result was the verification of laying during afternoon, as it had been verified during morning, was expressed only by Hy-Line W36 hens. Probably, this behavior is related to the better adaptation of this line to laying, which may be proved by percentage of time spent expressing this behavior in relation to another line.

#### **Expression of Behaviors in Litter System at Comfort Condition.**

In Table 1 are described bird behaviors at comfort condition. During evaluating of behaviors executed in litter/lair system at comfort condition (Table 1), the expression of the so-called "comfort behaviors" was evidenced by birds.

A factor that has great effect on bird behavior is that all animals follow a bio-rhythm, which is associated to photoperiod. In this study, the photoperiod was 16:8, in other words, 16 hours of light and 8 hours of darkness, being the light period from 8:00 to 24:00h.

The period of the day (morning or afternoon) was considered in behavior evaluation. At morning birds spent more time eating (around 30%), certainly because of the feeding time. Other behaviors that were more observed during the morning were stretching and laying, besides perching that had a percentage of 15% for Hy-Line W36. During afternoon, there was a slight reduction in time spent eating but, birds increased other behaviors, as feather pecking, perching, dust-bathing and standing, besides aggressiveness.

Table 1 also shows that in litter breeding system at comfort condition, lines did not show many differences in relation to those behaviors evaluated, independently of the period of the day.

Table 1. Values of average percentage of time spent for expressing behaviors at thermal comfort condition.

Behavior Pattern	Morning		Afternoon		Total	
	Hy-Line Brown	Hy-Line W36	Hy-Line Brown	Hy-Line W36	Hy-Line Brown	Hy-Line W36
Eating	32	28	23	20	27.5	24
Drinking	4	2	4	1	4	1.5
Stretching	29	11	23	14	26	12.5
Looking for nest	21	13	12	-	16.5	6.5
Laying	-	26	-	18	-	22
Comfort movements	1	1	1	1	1	1
Feather pecking	6	4	7	7	6.5	5.5
Perching	-	15	9	19	4.5	17
Dust-bathing	7	-	9	4	8	2
Aggressiveness	-	-	-	1	-	0.5
Sitting	-	-	8	12	4	6
Standing	-	-	4	3	2	1.5
Total	100	100	100	100	100	100

#### **Expression of Behaviors in Litter Breeding System at Stress Condition.**

Birds, at heat stress in relation to comfort condition, did not show or showed few behaviors associated to “comfort movements”. In general, at heat stress birds increase the expression of drinking, Sturkie (1967) and decrease abruptly feed intake, Payne (1967). Another interesting observation was the expression of behaviors as sitting and standing, certainly, because these behaviors are directly associated to animal physiology balance since heat production is reduced by avoiding movements.

In Table 2 bird behaviors at heat stress are described, considering observations realized in intervals of 1 hour during the morning (from 10:00 to 11:00 h) and 2 hours during the afternoon (from 13:00 to 14:00 h and from 16:00 to 17:00 h).

Table 2. Mean values, in percentage, of time spent for birds expressing their behaviors at heat stress condition.

Behavior Pattern	Morning		Afternoon		Total	
	Hy-Line Brown	Hy-Line W36	Hy-Line Brown	Hy-Line W36	Hy-Line Brown	Hy-Line W36
Eating	20	36	16	25	18	30.5
Drinking	8	9	6	6	7	7.5
Stretching	19	25	26	19	22.5	22
Looking for nest	-	-	-	25	-	12.5
Laying	-	-	-	-	-	-
Comfort movements	-	-	-	-	-	-
Feather pecking	3	4	5	5	4	4.5
Perching	24	-	19	4	21.5	2
Dust-bathing	-	-	-	-	-	-
Aggressiveness	-	-	-	-	-	-
Sitting	2	14	4	6	3	10
Standing	24	12	24	10	24	11
Total	100	100	100	100	100	100

Table 2 shows that Hy-Line W36 spent more time eating than Hy-Line Brown, independently of the evaluation period (morning or afternoon), indicating that birds of Hy-Line W36, even at heat stress condition, showed a better productive performance once they showed a high feed intake.

Another interesting result was the percentage of time spent standing (24%) by Hy-Line Brown hens, which evidenced the high heat stress resistance of this line.

In relation to perching, Hy-Line Brown hens reduced percentage of time perching at afternoon while Hy-Line W36 hens increased the expression of this behavior.

In relation to “comfort movements”, as dust-bathing and comfort movements, birds did not expressed them at heat stress condition, independently of the period of the day (morning or afternoon), confirming the stress effect provoked by high temperatures and relative humidity on expression of normal behaviors.

#### **Expression of Behaviors in Conventional Breeding System.**

Behavior analysis in conventional breeding system (cages) was hard to be done because of problems, as video camera position (fixed on the roof) and number of birds per cage (5 bird/cage), which prejudiced image analysis preventing a more detailed analysis.

Another problem was the impossibility of executing a complete individual evaluation of birds due to the distance of video cameras and to bird crowding in cages, beside the lack of effectiveness for distinguish birds by dorsal painting. Thus, at comfort condition during morning or afternoon Hy-Line Brown and Hy-Line W36 birds spent more time eating. Also in this condition, other behaviors were observed, as feather investigation and water drinking.

At heat stress condition, behaviors observed more frequently, mainly at afternoon, were sitting, standing and drinking. All of them are very common at heat stress condition. Feed intake, mainly of Hy-Line Brown hens, was greatly reduced (around 50%). Behaviors, such as comfort movements and lying, were not evaluated because was not possible to determine when they started or finished.

#### **Egg Quality Parameters.**

Analyzing egg quality parameters was possible to observe a reduction in egg quality when birds were submitted to heat stress. Table 3 shows the means of egg quality parameters found by variance analysis.

Table 3. Means of egg quality parameters.

			Quality Parameters			
			Weight	Specific Gravity	Haugh Unit	Eggshell thickness
Comfort	Line	W36	55.5a	1.0917a	102.0a	45.8a
		Brown	57.8a	1.0897a	105.4b	43.7b
	System	Litter	54.7a	1.0912a	104.0a	45.1a
		Cages	55.5a	1.0902a	103.4a	44.4a
Stress	Line	W36	51.8b	1.0772b	100.3a	38.3c
		Brown	52.7c	1.0775b	101.6a	36.8d
	System	Litter	53.5c	1.0777b	100.8c	38.3c
		Cages	51.0b	1.0770b	101.1c	36.8c

**Egg weight:** it was observed by statistical analysis that means of egg weight were significantly reduced ( $P<0.05$ ) when hens were submitted to heat stress. This result, difference between eggs laid at heat stress and those laid at comfort condition, was expected.

The quality parameter of **specific gravity** is closely associated to eggshell quality, Hamilton (1982). As it was found in this study, specific gravity increases with the increase of eggshell thickness.

The quality parameter of **Haugh Unit** also showed a significant ( $P<0.05$ ) reduction, which might had occurred as consequence of the high temperatures. This result is in accordance with reports showing the decrease of Haugh unit because of temperature increase.

For **eggshell thickness**, there was a significant ( $P<0.05$ ) reduction in means of values taken from eggs produced during heat stress, which might be a consequence of reduction in blood calcium balance. It is very well known that this balance is affected when bird are under high temperatures due to decrease of plasma calcium concentration, which prejudices eggshell formation (Hamilton, 1982).

### **Conclusion**

By analyzing bird behaviors, could be observed the expression of natural comfort behaviors in breeding system with litters that certainly allowed better conditions of bird welfare when it was compared to conventional breeding system in cages, where comfort behaviors were not observed.

Quality parameters of eggs were affected when birds were submitted to heat stress conditions, mainly in breeding system in cages, showing the importance of hen welfare aspects and the type of breeding system.

## References

- Barbosa filho, J.A.D. 2005. Welfare Evaluation by Image Analysis of Laying Hens in Different Houses Systems and Environmental Conditions. 2005. 123 p. Dissertation (MSc in Physics of Agricultural Environment) - Escola Superior de Agricultura "Luiz de Queiroz", Universidade de São Paulo, Piracicaba.
- Bennion, N.L.; Warren, D.C. 1933. Temperature and its effect on egg size in the domestic fowl. *Poultry Science*, 12: 69-82.
- Craig, J.V. 1992. Measuring social behavior in poultry. *Poultry Science*, 71: 650-657.
- Hamilton, R. M. G. 1982. Methods and factors that affect the measurement of egg shell quality. *Poultry Science*, 61: 2022-2039
- Jensen, P.; Toates, F.M. 1993. Who Needs "Behavioral Needs"? Motivational aspects of the needs of animals. *Applied Animal Behavior Science*, 37: 161-181.
- Johnson, N.; Hogg, D. 1996. Learning the distribution of object trajectories for event recognition. *Image and vision computing*, 14: 609-615.
- Leroy T.; Ceunen J.; Struelens E.; Janssen A.; Tuytens F.; De Baere K.; Zoons, J.; Sonck B.; Vranken E.; Berckmans D. 2003. Developing a quantitative computer vision method for on-line classification of poultry behavior in furnished cages, Proceedings... In: ASAE Meeting, Local: Las Vegas, USA (ASAE Paper, 034006).
- Mashaly, M.M.; Hendricks, G.L.; Kalama, M.A.; Gehad, A.E.; Abbas, A.O.; Patterson, P.H. 2004. Effect of heat stress on production parameters and immune responses of commercial laying hens. *Poultry Science*, 83: 889-894.
- Pardi, H.S. 1977. Influencia da comercialização na qualidade dos ovos de consumo. Rio de Janeiro, Dissertação (Mestrado) - Universidade Federal Fluminense, 73 p.
- Payne, G.C. 1967. Environmental temperature and egg production - The physiology of the domestic fowl, Edinburgh, p. 235-241.
- Rudkin, C.; Stewart, G.D. 2003. Behaviour of hens in cages – A pilot study using video tapes. A Report for the Rural Industries Research and Development Corporation (RIRDC), v. 40, n. 477, p. 102.
- Sevegnani, K.B.; Macari, M.; Nääs, I.A.; Moura, D.J.; Silva, I.J.O. 2001. Variação da temperatura média corporal de frangos de corte em terminação, submetidos às temperaturas de 24 e 32°C, em câmara climática. *Revista Brasileira de Ciência Avícola*, supl. 3, p. 58.
- SAS Institute. 1998. Statistical Analysis System (Software). Cary, 620p.
- Singer, P. 1991. Animal Liberation. p - 97,118.
- Sturkie, P.D. 1967. Fisiologia aviária, Zaragoza. Ed. Acribia, 607p.
- Voisey, P.W.; Hunt, J.R. 1974. Measurements of eggshell strength. *Textile Studies*. n. 5, p. 135-182.
- Xin, J.; Beck, H.; Halsey, L.; Fletcher, J.; Zazueta, F. 1998. Using digital cameras and the internet to identify plant insect and disease problems. In: International Conference on Computers in Agriculture, 17; Orlando, 1998. Proceedings... Orlando: ASAE, p. 327-329.