DIRECT VS INDIRECT BEHAVIOURAL OBSERVATIONS IN THREE ITALIAN CHICKEN BREEDS

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Original scientific paper

Abstract: Measuring beahaviour has already been a debated issue. The aim of this paper is to compare direct observations and observations by videotapes in three different Italian chicken breeds (Valdarnese Bianca, Bionda Piemontese, Robusta Maculata), in order to provide indications about the most suitable method in behavioural studies. The three breeds were reared in an experimental poultry house (10 birds=m^2, standard intensive broiler production density). Fifty birds/breeds (sex ratio 1:1) were housed in litter floor pens and fed the same diet (ME 11.8MJ=Kg, CP 18%). The comparison between direct and indirect observation showed significant differences for almost each behavioural category. Activity and resting behaviour were generally well visible in both types of observations while feeding and mainly interaction were better identified with the direct observation.

Key words: chicken behaviour, direct and indirect observations, Italian chicken breeds

Introduction

According to The State of the World's Animal Genetic Resources for Food and Agriculture, 20% of documented livestock breeds are at risk of extinction: 1500 of the 7600 breeds around the globe may be lost forever in the near future. Many indigenous breeds, some of which are threatened with extinction, have characteristics such as resilience to climatic stress and resistance to diseases and parasites, which make them well adapted to local conditions, and of great potential importance to the future livestock production (*FAO, 2013*). From these considerations it is necessary to preserve and encourage the use of indigenous breeds in rural and organic farms.

Valdarnese Bianca, Bionda Piemontese, Robusta Maculata are typical examples of Italian chicken breeds. Valdarnese Bianca can be considered the only

traditional Italian meat-type chicken breed, originates from the valleys between Florence and Arezzo in Tuscany, central Italy (*Marelli et al., 2006*). The average weight is 2.9–3.3 kg for cocks, 2.0–2.5 kg for hens. Hens lay an average of 135 eggs per year.

The Bionda Piemontese is a traditional dual-purpose breed of chicken originating by the Piemonte region in north western Italy, from which it takes its name. The average weight is 2.5-2.8 kg for cocks, 2-2.3 kg for hens. Hens lay 180–200 eggs per year.

The Robusta Maculata was created between 1959 and 1965 at the Stazione Sperimentale di Pollicoltura, an experimental chicken breeding centre, of Rovigo, in the Veneto Region. Robusta Maculata is a dual-purpose breed. Hens lay 140–160 eggs per year. In meat production, birds reach a weight of 1.9–2.0 kg in four months, and are usually slaughtered at 18 weeks.

The use of behavioural indicators has major advantages in animal welfare studies. Not only it is non-invasive, but it is also often non-intrusive (does not disturb the animal). Behaviour is also the result of all of the animal's own decision-making processes ‒ "the final common path" as *Sherrington (1906)* called it. It is also "the expression of the emotions" (*Darwin 1872*) ‒ the ultimate phenotype (*Dawkins, 2004*).

The choice of the tool used to record behavioural observations is very important and depends to sampling rules and data validity (*Altmann, 1974*). Observations may be direct or indirect. In the first case, the observer is in the field with animals. Several studies indicate that the presence of humans can affect their behaviour, during direct observations (*Götmark and Ahlund,1984; Davis and Balfour, 1992*). This problem may be overcome with indirect observations by using video recording systems. The major advantage of video recordings is the possibility of observing animals without the presence of humans (*Martin and Bateson, 1993*). Another advantage of video recordings is to provide a high degree of reproducibility when measuring observations, unlike real-time data collection, the video recordings can be reviewed several times; by using a time-lapse video recorder, it is even possible to record several days on one tape (*Gavinelli et al., 1994; Tosi et al., 2006*).

The aim of this paper is to define some behavioural characteristics of three Italian chicken breeds and to compare direct and indirect observations in order to provide indications about the most suitable method in behavioural studies. Moreover this study aims to identify which behaviours were visible in each behavioural category and what behaviours were better evaluated with direct and indirect observations.

Materials and Methods

Animals and husbandry. Three traditional Italian chicken breeds (10

birds=m^2) were compared: Valdarnese Bianca (VB), Bionda Piemontese (BP) and Robusta Maculata (RM). Table 1 shows breeds characteristics and performance (*Marelli et al., 2010*).

One day old chicks (mixed sex) have been grouped by breed in three pens. The birds of the three breeds were reared in standard condition at the same density (10 birds/m^2) in the same facility to avoid environmental effects on recorded behavioural data. The pens dimensions were 3m (length) x 2m (width) x 3m (height), located in the experimental facility of the faculty of Veterinary Medicine of Milan.

The pen floor was covered with wood shaving litter (10 cm deep) and every pen was equipped with one infra-red brooder. According to standard chick raising procedure the temperature was of 30°C at day one then reaching the rearing temperature of 21°C (after a decreasing of 2.5°C per week). Every pen was supplied with one feeding-tray and two drinking bells. The water and a standard broiler chicken pelleted diet were available *ad libitum*. The photoperiod was of 23:1 during the first week, then the light was only the natural one.

Table 1. Breeds characteristics and performance.

Breed name Feather colour	Mean weight at slaughter (both sexes; 80 d/age) Growth rate	Weight Rooster/Hen Slaughter age	Egg: Shell color Average weight N eggs/year Age at first egg
Bionda Piemontese	1186 g	2.5–2.8/2.0–2.3 Kg	pinkish
	slow	70 days	55–60 g
buff			180–200 eggs/year
			6–7 months
Valdarnese Bianca	1182 g	3.1–.5/2.5–3.0 Kg	white
	Slow	≥120 days	51.54 g
white			130 eggs/year
			7 months
Robusta maculata	1336 g	4.0–4.5/2.8–3.3 Kg	pinkish-medium brown
	average/slow	126 days	55–60 g
black and white mottled			140–160 eggs/year
			5–6 months

Behavioural observations. During preliminary observation the pricipal behaviours of the poultry ethogram were chosen and divided into five behavioural categories: feeding, activity, interaction, resting and comfort.

Indirect Observations (IO). Each pen was video recorded at 39 and 79 days of age for 24 hours continuously using a digital video recorder. The video tapes were analysed by scan sampling technique (*Martin and Bateson, 1993*). The number of birds performing these behaviours was counted at 10-min intervals.

Direct Observations (DO). The previous behavioural categories were direct observed by a person located near the pen. The behaviours of each group of birds were recorded using scan sampling (*Altmann, 1974; Martin and Bateson, 1993*).

Each pen was scanned at 5-min intervals for a period of 1 h, thus giving 12 records, and the number of birds performing each behaviour was registered. Observations were repeated two times daily (08.30 AM and 02.30 PM) at the age of 38 and 78 days. Before starting, the observer waited five minutes to allow the animals to get used to human presence.

Statistical analysis. The data collected are showed as means±SEM of frequencies spent performing each behaviour.

DO and IO behavioural categories were analysed by means of one-way ANOVA (SPSS vers.19) both for breeds and behavioural observations methods. To perform the analysis the data were grouped in six daily period (DP).

Results

After a preliminary observation a list of 14 states of each behavioural categories in DO was defined: feeding (eating, drinking), activity (scratching, running/walking), interaction (pica, fighting, running away), resting (standing, sitting/laying) and comfort (preening, stretching, wing flapping, swelling, dust bathing). Only six states in the IO: feeding (eating, drinking), activity (walking, standing), interaction (fight) and rest (sitting/laying).

Starting from the definitions of the each behaviour belonging to the main behavioural categories it was possible to define what behaviours are clearly visible through the used recording method (DO and/or IO).

Eating and drinking were defined respectively as pecking of food and swallowing and dipping the beak in the water and raising the beak thereafter (*Lolli et al., 2010*); these movements, that are categorised as feeding behaviour, were visible both in DO and IO.

Vice versa it was quite impossible to recognize animals performing comfort activity through IO.

The comparison between direct and observation through video recorder, showed significant differences for some behavioural categories (figure 1). Activity and rest behaviour didn't shown any differences between direct and indirect observations. Vice versa feed and interaction behaviours showed significant

differences between the two observation methods. The average number of animals direct observed in feeding behaviour were significant higher than the number registered with indirect observations (7,82 *vs* 6,5; P<0,0001). Interaction showed a high significant differences between observation methods (3,24 *vs* 1,19, direct *vs* indirect respectively; P<0,0001).

In figure 2 the different behaviours showed daily rhythms typical of the specie-specific ethogram.

Overwhelmingly, the greatest part of a poultry's day (63,5%) was spent resting. Birds spent more or less the same percentage of time in feed and active behaviours (16,3% and 17,2%), while the percentage of daily interaction were only 3%. Birds spent more time in rest behaviour in the first, fifth and sixth daily period (DP) while on the contrary, they showed less active behaviours. Instead activity, feeding and interactions decreased in these DP, while increased in the late afternoon and in the early morning like the natural one (figure 2).
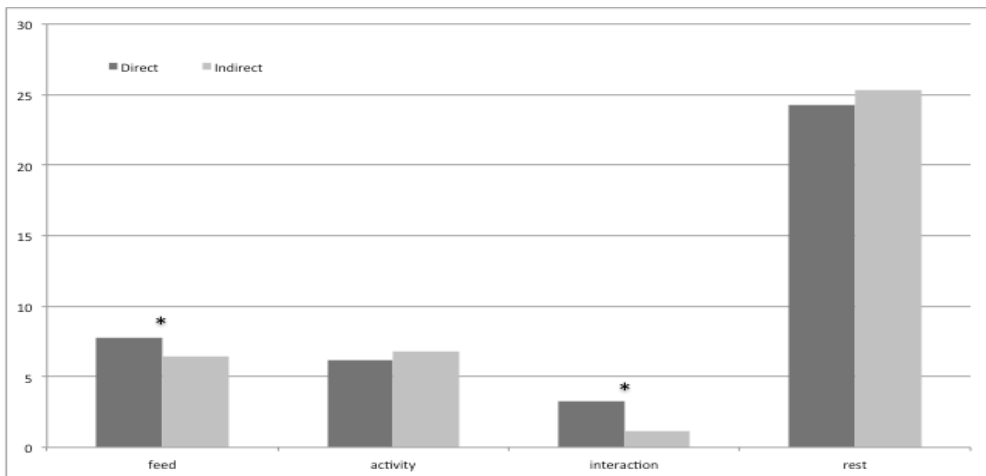

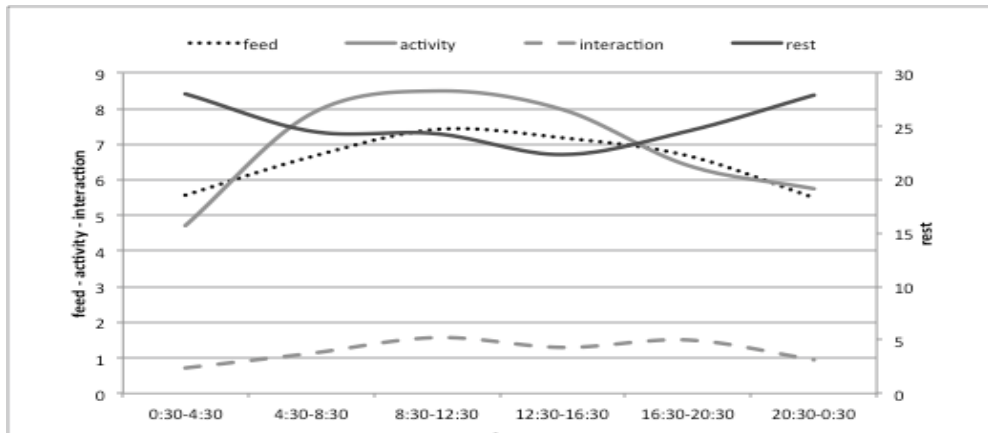**Figure 1. Direct *vs* Indirect observations (Mean freq±SEM)**

Figure 2. Ethogram and daily rhythm

The direct observations showed some significant differences between sessions (38 and 78 days of age). Feeding and explorative behaviour were higher in younger while comfort and interaction behaviour showed an increase in the second observational session (figure 3). Rest and activity didn't shown differences between sessions.
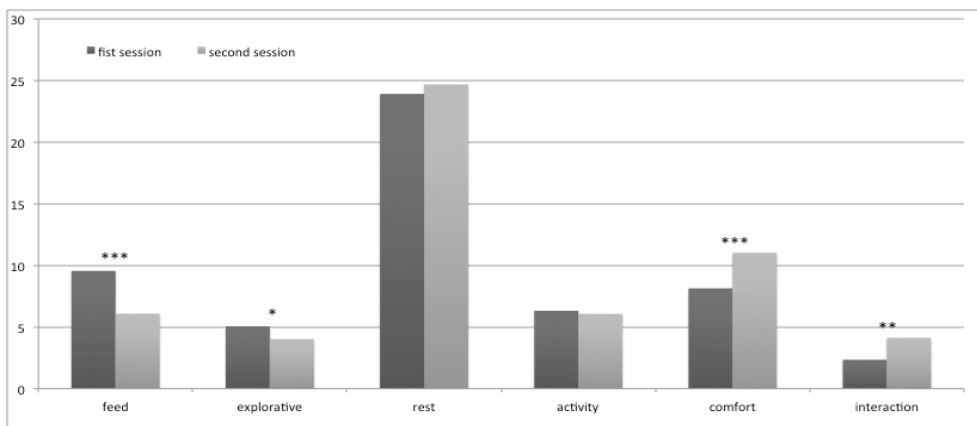

Figure 3. Effect of age on direct behavioural observations (Mean freq±SEM)

Breeds. After a general discussion about results, it necessary to analyse individual breeds. From a behavioural point of view, they showed different levels of the conservation of ancestral characteristics due to the different genetic selection (table 2).

Table 2. Number of birds of each breeds exhibiting a specific behavioural categories under the two methods of observations (mean +/- SEM)

Behaviour Categories	Valdarnese Bianca			Bianca Piemontese			Robusta Maculata		
	indirect	direct	P value	indirect	direct	P value	indirect	direct	P value
Feeding	6,59±0,24	7,65±0,81	0,119	5,18±0,18	7,29±0,5	<0,0001	7,7±0,24	8,52±0,45	0,18
Activity	6,98±0,23	4,48±0,373	<0,0001	7,08±0,37	7,31±0,6	0,80	6,46±0,23	6,79±0,48	0,58
Interaction	1,31±0,11	1,71±0,273	0,18	1,29±0,11	4,77±0,56	<0,0001	0,97±0,09	3,23±0,53	<0,0001
Resting	22,43±0,54	27,54±1,6	0,001	26,16±0,46	19,38±1,4	<0,0001	27,38±0,47	25,98±1,12	0,26

Valdarnese Bianca: analysis of the data in VB did not shown any statistical differences between the direct and indirect observations on feeding and interaction behaviour while highly significant differences in the activity and resting behaviour emerged.

Bionda Piemontese: all behaviours observed showed statistical differences between the two methods, except activity that had the same mean frequency in direct and indirect observations.

Robusta Maculata: there were not any statistical differences in feeding, activity and resting behaviour between methods, while the interactions showed a highly significant differences.

Indirect observations showed some differences among three breeds. Both mean frequency of feeding and resting behaviour were higher in RM than VB and BP. This breed spent more time in feeding and resting behaviour than in active and interaction behaviour.

Vice versa Bionda Piemontese and Valdarnese Bianca spent less time in resting and in feeding but registered high interaction and activity. VB showed less time spending resting (60,13%) than RM and BP (64,4% and 65,9%). VB showed +3% of activity and + 1,3% of interaction compared to RM.

Discussion

These breeds seemed to maintain a natural rhythm in feeding behaviour similar to their ancestors, in which food searching is usually concentrated at dawn and in the late afternoon. On the contrary, when birds were reared in intensive conditions with artificial lighting program, seems to loose specific rhythm (*Lolli et al., 2010*).

Results on the comparison between direct and indirect observations showed some interesting differences between behavioural patterns observed. These results are probably due to the fact that many behavioural patterns were not clearly visible on the screen due to the video resolution. Birds are small and move extremely rapidity, and for that reason it could be more difficult to see by video exactly what they are doing. Some behavioural categories, like feeding and interaction are better observed directly than indirectly. Observer can see if the animals really eat or drink investigating the position of the beck (up or down or inside drinkers/feeders), very difficult and not so clearly visible on the screen due probably to the little dimensions of birds. Interactions are not clear visible by video too. In this case, pica, fighting, running away and others agonistic behaviour, could be confused with activity behaviour due to their rapidity. Feeding and mainly interactions, are better distinguishable viewing details which may have been lost during video recording also due to the time-lapse fragmentation.

Under a behavioural point of view, the considered typical Italian breeds showed daily activity rhythms more similar to the natural one, according to others authors (*Bessei, 2006*).

Video recording used as an alternative to direct observations seems to be useful and practical. The low level of disturbance for observed animals, and a continuous record of behaviours are the main advantages of this tool. Indirect observations can be successfully adopted for studying less detailed behaviours, such as a period of activity and rest, and more generic behavioural categories, as shown by different studies (*McFarlane et al., 1988; Gottardo et al., 1997, Tosi et al., 2006*). In fact, our results showed difficulty in analysing some behavioural categories such as interaction and feeding behaviour, using videotapes. Direct observations seems to be the only way to exactly quantify the behavioural interactions between birds as already reported by several authors (*Wagner 2004; Hazelton and Grossman 2009a,b; Almeida and Grossman, 2012*).

Conclusion

The results of the present study seem to confirm that if the aim of behavioural observations is to investigate only the rhythms of activity and resting, indirect observations seem to be the best solution. First of all because animals are not disturbed by observers and not change their "normal" daily rhythm; additionally it possible to carry out longer uninterrupted observation periods in different environmental situations, like change in photoperiod or diet. The direct observation are almost mandatory in behavioural studies that aim to evaluate some behaviour more in detail.

In conclusion, the choice of the observation method is to be done according to the real aim of the research that the researcher want to perform.

Direktna i indirektna zapažanja ponašanja tri italijanske rase živine

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Rezime

Ocena ponašanja je već diskutovano pitanje. Cilj ovog rada je da se uporede direktna zapažanja, opservacije ponašanja i opservacije pomoću video kaseta kod tri različite italijanske rase (Valdarnese Bjanka, Bionda Piemontese Robusta, Maculata), kako bi se obezbedila indikacija o najpogodnijoj metodi.

Tri rase su gajene u eksperimentalnim objektima za živinu (10 grla/m^2, standardna gustina naseljenosti u intenzivnoj proizvodnji brojlera). Pedeset grla po rasi (odnos polova 1:1) je smešteno u podne bokseve i hranjeno istim obrokom (ME 11.8MJ = Kg, CP 18%). Poređenje između direktnog i indirektnog posmatranja pokazalo je značajne razlike za skoro svaku kategoriju ponašanja.

Aktivnost i odmor kao oblici ponašanja su generalno dobro vidljivi kod oba tipa posmatranja dok se hranjenje i interakcija uglavnom bolje identifikuju u direktnom posmatranju.

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