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## Detection and comparison of time patterns of behaviours of two broiler breeder genotypes fed ad libitum and two levels of feed restriction

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### Abstract

Time structure of behavioural patterns of broiler breeders were investigated to assess comparative behavioural complexity using “Theme”. The behaviour of an experimental dwarf heavy broiler breeder selected for better viability and reproductive traits at the partial expenses of growth (E) was compared to a standard heavy broiler breeder (S). Both were either fed ad libitum (A), feed restricted at 55% of A from 6 to 15 weeks of age (I), or feed restricted as in commercial practice to match a standard growth curve (R) in a 2 genotypes × 3 diets factorial design with 8 pens of 14 hens per treatment. The fine mash feed contained 10 MJ ME/kg. In each pen, three hens were coloured-marked and video recorded twice from 6 to 13 weeks. Four to 5 h after feed distribution, 10 min files (288) were coded by focal sampling and 1 min files (107) of one hen per pen were coded in detail in the morning and afternoon sessions. In the 10 mins files, while E rested more often and longer on average than S, the total number of changes of states recorded per hour were 222 for E versus 184 for S. When both genotypes were feed restricted, resting was replaced by more frequent stepping and standing bouts, and eating events by pecking at the empty feeder and at the litter. The overall number of time structured T-patterns detected by Theme were more frequent per hour of behaviour in genotype E (142) compared to S (115) and in the feed restricted hens (151) compared to the ad libitum fed hens

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(83). Genotype and diet modulation of activity followed different paths and interactions were not significant. In the 1 min detailed files, most of the pecks were included in T-patterns and their mean lengths were consistent (feed = 145 ms for E and 161 ms for S; litter = 174 ms; empty feeder = 193 ms) whatever the feed restriction levels. Feed restriction did not “disorganize” the behaviour of broiler breeder hens and “hyperactivity” of feed restricted hens was mainly due to transitions between various activities. A transfer of feeding activities toward foraging and spot-pecking and a reduction of the number and duration of resting bouts were the major observed changes. No specific sequences of behaviour attributable to feed restriction could be consistently identified. A more detailed analysis of resting in heavy genotypes might bring new insights on adaptability of broiler breeders to ad libitum feeding.

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## 1. Introduction

In commercial practice, the growth curve of fast growing broiler breeders is controlled so that it is similar to that of slow growing chicken lines to preserve their liveability and reproductive fitness. The increasing gap between the amount of feed eaten ad libitum and restricted raises ethical concern requiring scientific quantification of alternatives including genetic selection to decrease the need for feed restriction (Mench, 2002).

Partial feed restriction has been explored with variable success (Yu et al., 1992; Hocking, 1993b; Bruggeman et al., 1999). Recently, an experimental dwarf heavy broiler breeder type selected for better viability and reproductive traits at the partial expense of growth (E) is capable of being fed ad libitum with relatively little effect on reproductive performance (Heck et al., 2004). The time-budget of these E hens showed a consistent trend to rest longer than a standard heavy broiler breeder (S) fed ad libitum or restricted (Puterflam et al., 2005). However, in that experiment E- and S-hens ate different feeds and the E genotype was not tested with a restricted feed allowance.

In the present experiment reproductive performances of the E and S breeders were further compared at three levels of feed allowances: ad libitum (A), intermediate feed restriction from 6 to 15 weeks of age (I) and constant feed restriction as in commercial practice (R). The production results confirmed that tolerance to ad libitum feeding a diluted diet in broiler breeders depends on the genotype and that the dw gene has a favourable effect on the reproductive responses to full feeding. Intermediate feeding strategies such as moderate feed allowances from 6 to 15 weeks of age, did not reduce the adult body weight compared to ad libitum fed hens and had a limited effect on egg abnormalities but improved laying rate compared with ad libitum feeding. A severe feed restriction, as commercially practiced, was the only way to maintain reproductive fitness in the standard S line (Bruggeman et al., 2005).

The behaviour of the two types of breeders receiving the three diets was studied by focal sampling during the critical period from 6 to 15 weeks of age. It is the purpose of the present publication to present the results of an analysis of sequences of behaviour. As recently illustrated by Rutherford et al. (2003) by fractal analysis, the quantification of the severity of stressors requires novel methods to analyse the complexity of behavioural

patterns. Theme, an algorithm developed by Magnusson (2000) is able to detect hidden time related patterns (T-patterns) of behaviour, that when quantified may indicate the degree of structuring of the behaviour. The method has been applied successfully to various types of data from sport (Borrie et al., 2002) and to detailed analysis of feed pecking in chickens (Martaresche et al., 2000). Theme was applied in the present work, to the focal sampling files of E and S breeder hens fed the three feed allowances at two time scales: 10 min files coded at normal speed for which the basal unit is 0.1 s and accuracy depends on human observer's speed, and 1 min files at slow motion speed for detailed observations of the small-scale behaviour of hens at intervals of one image (0.04 s).

The three objectives of the research were (1) to compare E and S broiler breeder hens under three feeding conditions to try to identify behavioural markers such as sequences of events that might be used for selection of birds with specific responses to ad libitum or restricted feed allowances, (2) to assess the comparative welfare of birds of both genotypes and of each feeding regime (diet), and (3) to evaluate the potential use of Theme for the study of behavioural complexity in birds.

## 2. Animals, materials and methods

### 2.1. Animals and housing

A detailed description of the experiment and production results was published (Bruggeman et al., 2005). Briefly, 672-day-old female chicks were delivered by Hubbard (Chateaubourg, France). There were 336 chicks of conventional standard broiler breeders Hubbard (S), and 336 chicks of an experimental dwarf heavy broiler breeder type selected for better viability and reproductive traits at the partial expenses of growth (E). A third of the chicks of each genotype (SA and EA) were fed ad libitum. Another third (SR and ER) were feed restricted from the age of 2 weeks in order to match a reference body weight curve provided by the breeder (average feed restriction from 6 to 15 weeks of age: 29% of SA for SR and 45% of EA for ER). The last third (SI and EI) received an intermediate feed restriction program from 6 to 15 weeks of age only, at 55% of the feed intake of the corresponding ad libitum fed groups. The six experimental treatments (SA, SI, SR, EA, EI, ER) were replicated 8 times (8 pens of 14 chicks) according to a randomised block design in a poultry shed of 48 pens (1.7 m × 1.7 m). A low energy (10 MJ/kg) diet containing 373 g wheat bran/kg from 2 to 20 weeks and 320 g wheat bran/kg during the laying phase was fed as a fine meal (particles >2 mm: 5–10%; particles <0.5 mm: 30–35%). Feed distributed was weighed every day for each pen in order to minimize the accumulation of feed refusal in the feeder of ad lib fed hens. Fresh feed was distributed daily at 10 am, 1.5 h after lights were switched on. Birds of the six treatments were exposed to the same photoperiods: constant light during the first week, weeks 2–18, 7L:17D, increasing by 1 h light per week to reach 16L:8D after 23 weeks of age. At 18 weeks of age three nests (30 cm × 30 cm × 40 cm) were connected to each pen and egg production was recorded every day by pen. The present experiment concerned only the growing stage, methods and results of the laying stage can be found in Bruggeman et al. (2005).

## 2.2. Behavioural observation

In each pen three target birds randomly chosen by tag number were colour marked on the back (black, red, green) for individual identification. From 6 to 13 weeks of age, two types of video sessions were simultaneously recorded. Four cameras surveyed four pens starting 4 h after feed distribution (2 pm). Each pen was filmed for 30 consecutive minutes. Four days a week, cameras were changed to 4 new pens to have a complete cycle of all 48 pens over 3 weeks (4 cameras  $\times$  4 days  $\times$  3 weeks = 48 pens). A fifth camera equipped with a remote operated electronic zoom and swivelling turret was used during the morning meal (1 h after feed distribution) and at 4 pm at the end of the first session. The camera on a moving stand was placed above the pen wall (2 m high) and the operator left the room for 15 min before recording the black bird in close up for 1 min. The beginning signal of a session was 'one peck given at something'. Only SA, SR, EA and ER pens were recorded for the 1 min files in two pens/day. The order of filming the two sessions followed the same (randomised block) pen order and lasted 3 weeks. The first series lasted from 6 to 9 weeks of age and the second one from 11 to 13 weeks of age. All recorded sessions were kept on the hard disks of a digital recorder (Alcatraz, Macrosystems, Belgium) for further behaviour analyses by focal sampling of the target birds.

Behaviour of the target birds of each pen was coded by the same person according to the ethograms defined in [Table 1](#) using The Observer Video Pro 4.0 ([www.noldus.com](http://www.noldus.com); Noldus et al., 1999). Two types of files were coded: 10 min files of the three marked hens at normal speed and 1 min files of the black marked hen at slow motion (in general five times slower speed and eventually image by image). Prior to coding, files were digitalized on the computer using a Miro-video 10 card. For the 10 min files the 30 min recorded session of one pen was surveyed and the start of a file was determined for each marked chicken as 'one peck given at something' and lasted 10 consecutive minutes whatever the activities of the bird. Digitalized files were carefully identified by the number of the pen and colour of the mark and then opened in the Observer program for direct coding using a configuration corresponding to the ethogram ([Table 1](#)). A single class of exclusive states was used, preening or dust-bathing being preferentially coded than standing or resting. Short events, mainly pecks with the beak or specific leg movements (steps excluded) were concomitant to standing or resting states.

Coding of 1 min digitalized files used a distinct ethogram that takes into account the duration of short movements such as pecks ([Table 1](#)). Two classes of states were defined as general position of the bird's body for the first class and specific head and leg movements for the second class of states. States were exclusive within a class but behaviours belonging to distinct classes could be concomitant. In the first class, when the bird moved in the pen, the movement was coded as state 'step' and not as separated leg movements in the second class.

## 2.3. Calculations and statistical analysis

For 10 min files, frequencies and duration (for states) of each file were computed using the 'elementary statistic' module of Observer and then exported to a spreadsheet. Data were summed within pen and the mean length of bouts of states were computed by dividing

Table 1  
Ethogram definitions

10 min files	Definition
<b>States</b>	
Stand	Stands upright without identifiable activity
Step	Moves in the pen
Rest	Lying on the floor without identifiable activity
Preen	Directs attention with the beak towards body and feathers
Dust-bathe	While lying on floor, kicks litter onto body and wiggles body in litter dust
<b>Events</b>	
Peck at feed	Pecks inside the feeder where feed is present
Peck at nipples	Pecks at the water nipples
Peck at litter	Pecks at the litter or anything on floor bedding
Peck at empty feeder	Pecks at the outside of the feeder or inside an empty feeder
Peck at bird	Non-aggressive pecks with companion birds (no avoidance)
Peck at environment	Pecks at the wall of the pen or at any spot not describe before
Stretch leg	Unilateral extension of one leg and/or wing towards bird sides
Scratch litter	Backward strokes with legs as if to dig litter
Fight	Aggressive peck inducing avoidance (received or given)
1 min files	Definition
<b>First class states<sup>a</sup></b>	
Rest	Lying on the floor
Stand head-up	Stands upright with head above line of back
Stand head –down	Stands upright with head below line of back
Step	Moves in the pen
<b>Second class states<sup>a</sup></b>	
Inactive	No identifiable activity
Move head	Movement of the head (in general rotation or lateral) without identifiable pecking target (see below)
Peck at litter	Pecks at the litter or anything on floor bedding
Peck at feed	Pecks inside the feeder where feed is present
Peck at bird	Non-aggressive pecks at companion birds (no avoidance)
Peck at nipples	Pecks at the water nipples
Peck at empty feeder	Pecks at the outside of the feeder or inside an empty feeder
Preen	Directs attention with the beak towards body and feathers
Peck at environment	Pecks at the wall of the pen or at any spot not describe before
Scratch litter	Backward strokes with legs as if to dig litter
Stretch leg	Unilateral extension of one leg and/or wing towards bird sides

<sup>a</sup> States belonging to different classes can be concomitant although states within the same class are exclusive.

the duration by the corresponding frequency. Two-way ANOVA (genotype  $\times$  diet) was applied and when significant, mean differences were tested by Newman and Keuls multiple comparison test ( $P < 0.05$ ). Frequencies of events when sufficiently expressed (see Section 3) were compared by non-parametric tests (Mann and Whitney for genotype and Kruskal–Wallis for diets). Transitions between behaviours were calculated using the ‘lag-sequential analysis’ module of Observer (with no lag) grouping all the files of the same treatment together. Transition rate (higher than 1%) was represented by arrows where thickness was proportional to the percentage of the total number of transitions between distinct

behaviours (repeated events excluded). The duration of states were represented on the same diagram as a rectangle where the surface area was proportional to the overall percentage of time spent in a state for one treatment. Frequencies of events were represented as circles where the surface area was proportional to the percentage of the overall number of events expressed by the chickens on each treatment.

Original data files coded with The Observer were transferred to Theme 4.0 ([www.noldus.com](http://www.noldus.com)) software for T-pattern detection (Magnusson, 2000; Martaresche et al., 2000; Borrie et al., 2002). A T-pattern is a combination of two events or more separated by non-random critical intervals of time indicating that these events occur repeatedly ( $n \geq 3$  per file) within a time sequence that differs significantly from random ( $P < 0.0001$ ). The number of occurrences of detected T-patterns of behaviours and their characteristics (average duration of time lag between behaviours) were computed for each file and exported to a spreadsheet. Patterns were detected in most of the files (see Section 3) and data were subjected to two-way ANOVA (genotype  $\times$  diet) of pen means.

The 1 min files were analysed using Theme to detect T-patterns of behaviours and their characteristics following a procedure similar to the calculation applied to 10 min files. However, the behaviour of hens was completely different in the morning (eating) and afternoon files, and the two periods were analysed separately. Given the high number of coded variables compared to the actual number of replications ( $\leq 8$  per treatment), comparisons were limited to the most frequently detected T-patterns and the analysis of 1 min files must be considered as exploratory.

### 3. Results

#### 3.1. Production results

Production results were published by Bruggeman et al. (2005). Briefly, the results obtained in the present experiment were confirmed by a similar experiment run in another research centre (University of Leuven, see Bruggeman et al., 2005). When culling and mortality were combined, average percentages of liveability to 40 weeks of age were: EA = 91%, EI = 92.5%, ER = 99%, SA = 81.5%, SI = 88.5%, and SR = 97.5%. Most mortality was due to hot weather during the laying stage and no target hen died before 14 weeks of age. BW of ad lib fed hens rapidly diverged from their restricted counterparts. At 15 weeks of age the average body weight were: EA =  $2578 \pm 29$  g; EI =  $1878 \pm 7$  g; ER =  $1461 \pm 11$  g; SA =  $3904 \pm 16$  g; SI =  $2612 \pm 21$  g; SR =  $1565 \pm 16$  g (average and S.E. for pens). However, immediately after feed restriction was released (end of week 15), a compensatory feed intake for 9 weeks led to similar adult BW for EA and EI hens and for SA and SI hens.

#### 3.2. Behaviour in 10 min files

The six files coded per pen (twice for the three target hens) were summed to represent 60 min of behaviour. Dust-bathing was observed only three times in two pens during the experiment and could not be analysed. There was no significant genotype  $\times$  diet

Table 2

The frequencies, duration and mean length of behavioural states recorded during 1 h of pen observations of two different genotypes of broiler breeders (E and S) fed ad libitum (A), restricted to 55% of ad libitum (I) or restricted to match a standard growth curve (R)

Behaviour	Treatment						ANOVA <sup>a</sup>			
	EA	EI	ER	SA	SI	SR	Genotype <sup>b</sup>	Diet	G × D	SEM
<b>Stand</b>										
Nb of bouts	63	106	136	60	93	112	–	***	–	10
Duration (s)	1509	2583	2843	2137	2632	3078	*	***	–	145
Mean length (s)	24.1	25.1	22.3	38.4	28.9	30.1	***	–	–	2.8
<b>Step</b>										
Nb of bouts	40	85	111	43	75	92	–	***	–	8
Duration (s)	96	212	276	142	223	263	–	***	–	21
Mean length (s)	2.37	2.59	2.51	3.27	2.97	2.95	**	–	–	0.21
<b>Rest</b>										
Nb of bouts	28	16	13	19	9	2	*	**	–	5
Duration (s)	1702	556	184	1176	503	89	*	***	–	134
Mean length (s)	66.0	36.2	21.8	74.2	47.6	26.8	–	**	–	11.7
<b>Preen</b>										
Nb of bouts	24	22	23	12	18	16	*	–	–	4
Duration (s)	220	195	215	88	113	116	**	–	–	37
Mean length (s)	9.4	8.7	10.0	7.3	5.6	7.6	**	–	–	1.1

<sup>a</sup>  $N = 8$  pens per treatment with three target birds recorded for 10 min twice in each pen (1 h in total for each pen).

<sup>b</sup> Genotype: E vs. S.

interaction for the remaining states (Table 2). The mean length of one standing bout was significantly shorter in E-hens (24 s) than in S-hens (32 s). The same difference existed for stepping bouts (E = 2.5 s; S = 3.1 s) suggesting overall faster changes of activity in the dw-genotype. A larger number of resting bouts per hour (19 versus 10) and a larger number (23 versus 15) and mean length (9.4 s versus 6.8 s) of preening bouts confirmed the trend for the E-bird to rest and preen more with more frequent changes of activities than the standard large S-hens. The total number of changes of state recorded per hour was 222 for E versus 184 for S and, on average, E-hens rested for 22.6% of the time versus 16.4% for S-hens ( $P < 0.05$ ).

The number of bouts of standing and stepping increased proportionally to the severity of feed restriction and there was a highly significant corresponding decrease in the number and mean length of resting bouts (Table 2). On average, ad libitum fed hens rested 40%, the intermediate restricted birds (I) 15% and the restricted (R) 4% of their time. The total number of changes of state recorded per hour was proportional to the feed restriction intensity (144, 213 and 253  $\text{h}^{-1}$  on average for diets A, I and R, respectively). However, the mean length of stepping or standing bouts and the preening activities were not significantly affected by feed restriction (Table 2).

The total number of events occurring in 1 h was less in E-hens ( $2569 \pm 160$ ) compared to S ( $3198 \pm 199$ ). This difference was mainly due to a larger number of pecks given to the empty feeder in SI- and SR-hens compared to EI- and ER-hens (Mann–Whitney,  $z = 2.6$ ,

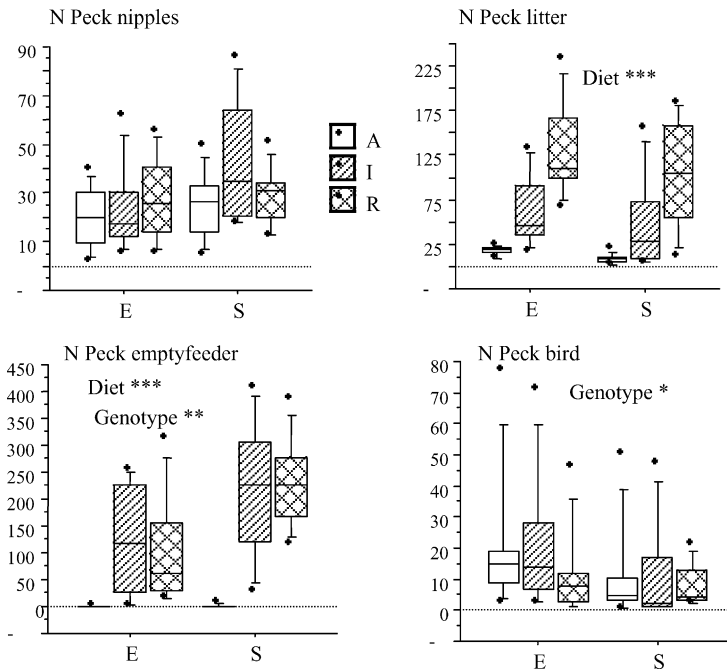


Fig. 1. Box plot of the median and quartile of frequencies of major events recorded during 1 h (6 files of 10 min) of pen observations of two different genotypes of broiler breeders (E and S) fed ad libitum (A), restricted to 55% of ad libitum (I) or restricted to match a standard curve of growth (R). Genotypes were compared by Mann and Whitney and Diets by Kruskal–Wallis tests.  $N = 8$  pens per treatment.

$P < 0.01$ ). Only five events were consistently observed: pecks at feed, litter, nipples, empty feeder and other birds. The other behaviours (stretch leg, peck at environment and fight) were too rare or variable from one pen to another to be analysed. Peck at feed was conditioned by the irregular (for diet I) constant (for diet A) and nil (for diet R) presence of feed in the afternoon. Scratch litter was closely associated with peck at litter. There was no significant difference of frequency of pecking at water nipples (Fig. 1) but restricted hens (I and R) did peck much more at the litter and at the empty feeders than A. The frequency of foraging in the litter (pecking and scratching) was proportional to the intensity of feed restriction in both genotypes although spot-pecking at an empty feeder was more variable from one bird to another and did not show a proportional response (Fig. 1). Social non-aggressive pecks to other birds were more frequently observed in E than S hens but feed restriction did not significantly change the frequency of this event.

The transition diagrams (Fig. 2) summarize the overall organization of behaviours in restricted and ad libitum fed S-hens. In SR, resting was replaced by more frequent stepping and standing bouts, and eating events by pecking at the empty feeder and at the litter. The diagram of the I diet were similar to R and no clear genotype differences appear on these transition diagrams (expressed in percentage of total activities or transitions, cf. Section 2).

When Theme was applied to the 288 10 min files recorded in this experiment, T-patterns were detected in 269 files. Eight SA-, 5 EA-, 4 SI- and 2 ER-files had no significant T-patterns.



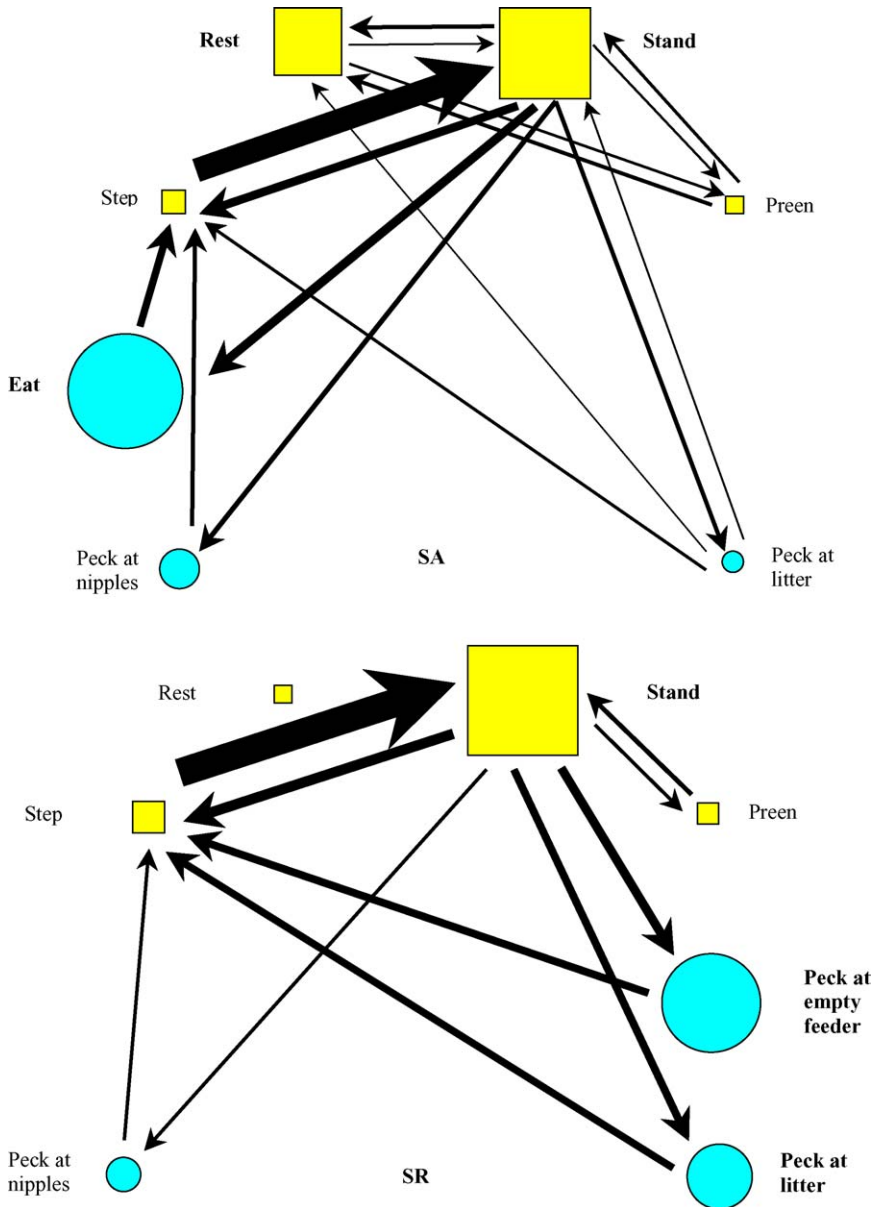


Fig. 2. Transition analysis comparing the behaviour of standard broiler breeders fed ad libitum (SA) or fed restricted (SR) 4–5 h after feed distribution. The size of squares is proportional to the time spent in corresponding states; that of circles is proportional to the number of pecks (events); and the size of the arrows is proportional to the frequency of transitions (all expressed in % of total of 48 10 min files per treatment, only values >1% are represented).

Table 3

The frequencies and mean length of major behavioural transitions detected by Theme during 1 h of pen observations of two different genotypes of broiler breeders (E and S) fed ad libitum (A), restricted to 55% of ad libitum (I) or restricted to match a standard growth curve (R)

T-patterns	Treatment						ANOVA <sup>a</sup>			
	EA	EI	ER	SA	SI	SR	Genotype <sup>b</sup>	Diet	G × D	SEM
Average number of T-pattern occurrences	93	156	177	73	131	141	*	***	–	13
Step → Stand										
Nb of bouts	36	85	110	39	75	90	–	***	–	9
Mean length (s)	2.47	2.59	2.51	3.15	2.97	2.99	**	–	–	0.21
Stand → Step										
Nb of bouts	13	24	25	10	21	16	–	*	–	4
Mean length (s)	4.8	4.8	4.2	4.1	6.0	4.2	–	–	–	0.7
Preen → Stand										
Nb of bouts	15	11	12	7	12	12	–	–	–	4
Mean length (s)	7.5	6.2	5.0	6.4	6.3	6.3	–	–	–	0.9

<sup>a</sup>  $N = 8$  pens per treatment with three target birds recorded for 10 min twice in each pen (1 h in total for each pen).

<sup>b</sup> Genotype: E vs. S.

Most of the files had one to four different T-patterns that occurred on average nine times but this frequency and the complexity of the T-patterns identified were very variable from one file to another. Most T-patterns included the transition between two behaviours and the most frequent were analysed (Table 3). The transition Step → Stand was found significant by Theme for almost all the stepping bouts observed. For example, there were on average 40 bouts of stepping per hour in treatment EA (Table 2) and Theme identified on average 36 of them as being time structured (Table 3). This means that most stepping bouts were time stable and this observation was true for all the treatments. On the contrary, the transition Stand → Step was detected as a significant T-pattern in a much lesser frequency than the total number of standing or stepping bouts. Those T-patterns were shorter (4–6 s) than the mean length of a standing bout (22–38 s). This means that there were two different types of standing bouts, the shorter ones being time-structured and their frequency was increased in the feed restricted hens. Similarly the preening bouts detected by Theme are only a part (60%) of the total, mostly those that were done while standing. The preening bouts included in T-patterns did not differ between genotypes suggesting that the preening differences between E and S were mainly those done while resting or were preening bouts followed by a behaviour other than stand.

The overall number of time structured T-patterns detected by Theme were more frequent per hour of behaviour in genotype E (on average 142) compared to S (115) and for the feed restricted hens (151) compared to the ad libitum fed hens (83, Table 3). A classical objection raised to Theme analysis is that the number of T-pattern occurrences is proportional to the number of data per file. This is not the case in the present experiment as illustrated by the complete lack of correlation ( $R^2 = 0.03$ ) existing between the number of data per file and the number of T-patterns detected by Theme (Fig. 3). However, if the short T-pattern between two behaviours could be statistically compared between treatments

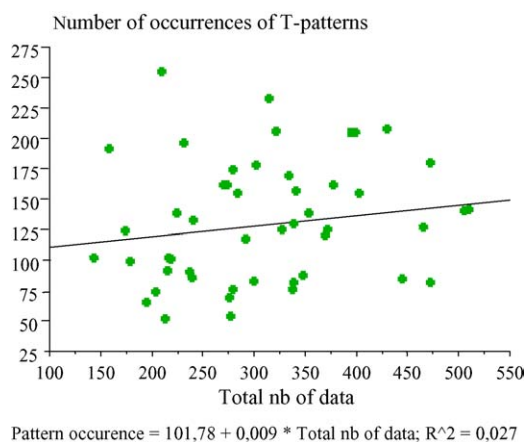


Fig. 3. Lack of correlation between the total number of data recorded per pen and the number of occurrences of T-patterns detected by Theme in the corresponding pens.

because they were consistently observed, larger T-patterns associating 3–5 behaviours were observed in a too small number of files to be statistically compared between treatments.

### 3.3. Behaviour in 1 min files

A close-up analysis of behaviour during the morning meal after feed distribution and during the afternoon was done on the black marked hen from SA, SR, EA and ER pens. One hundred and seven 1 min files were obtained. Unfortunately, in the afternoon several files could not be analysed because the observed hen was just resting. This reduced the number of repetitions to a critical four pens for SA birds in the afternoon. As a consequence, we have concentrated the following presentation on results of Theme analysis.

When Theme was applied to the 107 detailed coded 1 min files, all of them showed significant T-patterns. Many behavioural acts such as a peck at the feed or a head movement had relatively stable duration. For example, over 95% of the 1384 head movements, 4227 pecks at the feed, 1198 pecks at the litter or 968 pecks at the empty feeder that had been coded were included into T-patterns. For each case, the average duration of T-pattern could be compared (Table 4).

During the morning meal the overall number of T-pattern occurrences and number of distinct T-patterns per file were higher in feed restricted hens compared to ad libitum fed counterparts. The number of head movements (peck excluded) per minute was significantly less in feed restricted than in ad libitum fed birds. The duration of a head movement or of a peck at the feed was not affected significantly by the diet. The mean length of a peck at the feed was shorter in E-hens (145 ms on average) compared to S-hens (161 ms;  $P < 0.05$ ).

In the afternoon, no significant differences between treatments were observed in the number of occurrences and number of different T-patterns observed per minute. Head movements were more frequent ( $17.3 \pm 1.4$  versus  $10.7 \pm 1.0$ ;  $P < 0.01$ ) and longer ( $138 \pm 5$  ms versus  $112 \pm 6$  ms;  $P < 0.01$ ) in the afternoon than in the morning but not

Table 4

The frequencies of “T-patterns” of behaviours detected by Theme during 1 min detailed observations and characteristics of the most frequently observed activities of two different genotypes of broiler breeders (E and S) fed ad libitum (A), or restricted to match a standard curve of growth (R)

T-patterns	Treatment				ANOVA <sup>a</sup>			
	EA	ER	SA	SR	Genotype <sup>b</sup>	Diet	G × D	SEM
During the morning meal								
T-pattern occurrences	116	193	150	207	–	**	–	22
Nb of T-patterns/file	4.2	7.0	5.0	8.8	–	**	–	1.0
Move head								
Nb of bouts	13.9	8.0	13.1	7.4	–	**	–	1.8
Mean length (ms)	136	99	109	105	–	–	–	10
Peck at feed								
Nb of bouts	65	87	72	83	–	–	–	12
Mean length (ms)	146	143	160	162	*	–	–	7
During the afternoon								
T-pattern occurrences	77	74	70	99	–	–	–	14
Nb of T-patterns /file	3.7	4.1	3.5	5.3	–	–	–	0.6
Move head								
Nb of bouts	19.1	15.8	22.5	14.3	–	–	–	2.7
Mean length (ms)	145	133	148	131	–	–	–	10
Peck at litter								
Nb of bouts	7.5	36.4	28.3	28.0	–	**	**	4.3
Mean length (ms)	182	164	177	174	–	–	–	12
Peck at empty feeder								
Nb of bouts		37.0		50.9	–	<sup>c</sup>	<sup>c</sup>	9
Mean length (ms)		218		176	–	<sup>c</sup>	<sup>c</sup>	18

<sup>a</sup>  $N = 8$  pens per treatment with one target bird (60 s on average per pen) in the morning;  $N = 7$ (EA); 8(ER); 4(SA) and 7(SR) in the afternoon.

<sup>b</sup> Genotype: E vs. S.

<sup>c</sup> Not determined (not enough values for A diet).

significantly modified by the treatments. The number of pecks given to the litter was much higher in ER compared to EA but no difference was observed between SA (with only 4 birds) and SR (interaction,  $P < 0.01$ ). The average duration of a peck given to the litter was 174 ms and that of a peck given to the empty feeder 193 ms without a measurable effect of the treatments (Table 4).

Some other transitions were not systematically included into T-patterns. For example, the transition: “Stand head-down → Stand head-up” appeared 1317 times in the files but it was detected as a stable T-pattern only 178 times in 36 files. In the morning these transitions included into a T-pattern had a duration varying between 80 and 150 ms whereas the overall duration of all transitions “Stand head-down → Stand head-up” (those included or not in T-patterns) ranged from 20 to 800 ms. Out of respectively 25 and 27 files analysed in the morning eating sessions, 5 ad libitum birds only exhibited this T-pattern although 13 restricted hens showed it. It was a too small sample to be conclusive but it

illustrates the fact that the same behaviour might have distinct interpretation whether it was included in a T-pattern or not.

#### 4. Discussion

The production results suggested that the efficiency of ad libitum feeding a diluted diet in broiler breeders depends on the genotype and the *dw* gene is probably a favourable factor. Intermediate feeding strategies such as moderate feed allowances from 6 to 15 weeks of age did not change the adult body weight of hens and had limited effect on egg abnormalities but may improve laying rate. A severe feed restriction was the only way to maintain reproductive fitness in the tested S broiler breeder hen (Bruggeman et al., 2005).

One major variation observed between genotypes and diets was the activity of the birds. E hens consistently rested longer than S with the measurements done previously by scan sampling (Puterflam et al., 2005). However, this longer resting activity was due to an increased frequency of resting bouts of constant length at the expenses of the mean length of movements (stepping) and standing states. There is a major difference between resting more because of longer bouts of resting and resting more because of more frequent bouts of resting. The overall number of changes of states were more frequent in E-hens compared to S-hens suggesting a more 'lively' E-bird with shorter standing and moving bouts because their movements were probably faster than in S-hens. Bouts of preening of E-hens were more frequent and of longer mean length than in S-hens, which may also be the expression of more time or attention given to comfort behaviour. Similarly, social pecking was more frequently observed in the E- than in the S-hens. This difference referred to "allo-preening" rather than to "allo-pecking" for which genetic influences have been reviewed in layers (Kjaer and Hocking, 2004). The social pecking differences in the present experiment could therefore be considered as an extension of the preening differences between the two genotypes.

The stimulating effects of feed restriction on activity, in both genotypes, were clearly due to shorter and rarer resting bouts and an increase of the number of standing and stepping bouts of a relatively constant average duration. This increased activity or arousal in feed restricted birds has been reported by several researchers (Savory and Maros, 1993; Hocking et al., 1996; Nielsen et al., 2003). It may be linked to enhanced feeding motivation expressed by pecking behaviours oriented towards the litter and the empty feeder in restricted birds. Genotype and diet modulation of activity followed different paths and interactions between genotype and diet were not significant. In the comparison between genotypes, the organisation of the time was different because the birds had different size and possibly genetic differences in temperament. In the comparison between diets the behaviour was displaced from one activity (eating) towards substituting alternative oral behaviours (Hocking et al., 1996) as illustrated in Fig. 2.

In both E and S genotypes that were feed restricted, pecking at the litter in the afternoon was, in general, proportional to feed restriction severity and expressed in most hens, although pecking at an empty feeder was observed in several restricted birds at a high rate whatever the level of restriction. Pecking at an empty feeder or 'spot-pecking' is a stereotyped activity that might alleviate stress by its de-arousing effects (Kostal et al.,

1992; Savory et al., 1992) although the relationship with corticosterone or fear remains unclear (Hocking et al., 1997; Savory and Mann, 1997). There is a large difference with pecking at and scratching the litter, a natural foraging behaviour in chickens that is too often thwarted by the farming practise of “ad libitum” feeding a single highly nutritious balanced feed. Feed intake behaviour is not just ‘eating’ (Nielsen, 1999, 2004). The identification of parameters that can quantify the effects of feed restriction on hunger is a major issue (de Jong et al., 2002). Pecking at the litter was expressed proportionally to feed restriction in the present experiment. However, only three feed allowance levels were tested and when comparing E and S genotypes, feed restriction in proportion to ad libitum was similar in both genotypes for I (55%) but much more severe for SR (29%) than for ER (45%). The response evaluated on pecking at litter or spot-pecking at an empty feeder was not associated with these differences. Pecking and scratching at litter were probably one of the most reliable traits that express feeding motivation and spot-pecking at an empty feeder a good expression of frustration although Hocking (1993a) and Hocking et al. (1996) suggested that spot-pecking might also be a redirected foraging activity. Both are too variable to be used as routine quantitative indicators.

Detailed pecking characteristics measured in the 1 min files did not reveal major differences in the time duration of those acts associated with the experimental levels of feed allowance. Interestingly, pecks at feed were shorter in E compared to the S genotype consistent with the more active behaviour of E already mentioned or anatomical differences that might explain a quicker peck. However, if anatomical considerations were concerned this should have been noted for the other measures such as peck length that did not differ significantly between genotypes. The faster eating pecks in E hens were probably due to the more active behaviour of the bird.

The use of a relatively diluted feed (10 MJ/kg) in the present experiment might have interfered with the expression of feed motivation (Hocking et al., 2004). However, Savory and Lariviere (2000) demonstrated that the relationship between feeding motivation and reduction of growth rate was not altered by using feed dilution. Displaced pecking was observed even after a meal when the gut is full. Hunger and feed intake behaviour are not only monitored by gut feelings (Nielsen, 2004; Jones et al., 2004). Feed dilution and small particle size of the meal feed may have enlarged the duration of the eating time in restricted birds and this is the main reason for its utilisation in the present experiment. Unfortunately such a fine meal cannot help a broiler breeder to self-restrict, at least to the level required for adequate reproduction.

In the 10 min files coded at normal speed, Theme detected as T-patterns activities included major states (step, stand, preen) but much less frequently resting. Resting was less frequently observed and the duration of 10 min might have been too short to allow the observation of three resting bouts within a same file. No large T-patterns of 3–5 behaviours could be consistently identified in one treatment, although some individual birds exhibited a more structured behaviour than others. Stepping and short bouts of preening were consistently found as T-patterns in all treatments. Approximately the same proportion of short standing bouts before stepping were time stable whatever the diet. Chicks exposed to a minor change of form of feed particles increased the proportion of non-synchronized acts not included in T-patterns (Martaresche et al., 2000). Conversely here, no major decrease of synchronization of the behaviour of feed restricted chickens could be detected. On the

contrary, the number of T-pattern occurrences was higher in restricted hens than in ad libitum fed counterparts in 10 min files. However, in those 10 min files, pecking was coded as an event and the analysis could not evaluate the variation in the duration of those behaviours because Theme does not take into account repeated behaviours (i.e., peck at a feed – peck at feed) in a sequence.

In the 1 min detailed files, most of the pecks given to feed, litter or feeder were included in T-patterns of consistent mean length whatever the feed restriction levels. This shows that the regime did not disrupt the fine time structure of pecking under our conditions. Head movements were shorter and less frequent in the morning because during feed intake behaviour head rotations are mostly done within a narrow zone to select the target of the subsequent peck, although in the afternoon several head movements are given head-up to observe the environment. The reduction of the number of head movements during eating in restricted hens might be interpreted as a reduction of exploration while eating (Yo et al., 1997; Picard et al., 1999). The interesting transitions “Stand head-down → Stand head-up” detected by Theme, probably more frequent in restricted birds than in ad libitum fed counterparts, could be due to the facilitation of feed swallowing when the bird is eating fast or by the scanning for predators or competitors during a critical moment of the day. The possibility to discriminate T-patterns of transitions from non-synchronised transitions of the same behaviours that would have normally been analysed as a single behavioural process is definitely a useful contribution of Theme. The software requires precise coding facilitated by a slow motion video system in birds that typically act very quickly. The duration of the file is critical (the longer, the better) and the minimum number of T-patterns per file arbitrarily fixed to three here, requires further consideration when a large number of files are compared.

## 5. Conclusion

Feed restriction did not “disorganize” the behaviour of broiler breeder hens and “hyperactivity” of feed restricted hens was not due to changes in the duration of individual acts but was affected by the transitions between various activities. A transfer of feeding activities toward foraging and spot-pecking to the empty feeder and a reduction in the number and duration of resting bouts were the major observed change. No specific sequences of behaviour attributable to feed restriction could be consistently identified under our conditions using Theme. Although exhibiting quite different behaviour and reproductive fitness, the two genotypes of broiler breeders adapted similarly to the different feed allowances. The E genotype that rested more frequently also changed its behavioural activity more often. A more careful and detailed analysis of resting in heavy genotypes might bring new insights on the adaptability of broiler breeders to feeding system.

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