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Animal Welfare 2012, 21: 127-135
ISSN 0962-7286

The effect of keel fractures on egg-production parameters, mobility and behaviour in individual laying hens

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Abstract

A majority of laying hens fracture their keel bones during the laying cycle. It is not easy for a farmer to identify hens with fractures and hen survival rate seems high. Thus, the effect of both recent and healed fractures on bird welfare is unclear. We aimed to investigate the impact of these keel-bone fractures on hens' production and behaviour. The egg production, mobility and behaviour of Lohmann Brown hens without keel fractures were compared with that of hens with old healed fractures of varying severity. In addition, the keel-bone strength and body temperature around the fracture site was measured for each group. Hens with no fractures laid more eggs and had a higher egg-quality score (derived from measures of egg weight, egg surface area, shell weight, shell percentage and shell density). These hens had the highest keel area temperature, strongest keel bones, accessed perches more frequently and took a shorter time to negotiate a walkway obstacle test and to fly down from a raised perch. Hens without keel fractures were better in all investigated parameters than hens with keel fractures, indicating a detrimental effect of fractures on both welfare and economic return.

Keywords: *animal welfare, behaviour, egg production, keel fracture, laying hen, mobility*

Introduction

The prevalence of keel fractures in hens is high, and seems to have increased over the past 20 years. In the UK, non-cage systems, prevalences of 7.5% in percheries and 6.9% in free-range systems were first reported by Gregory *et al* (1990). More recent studies indicate far higher prevalences, 73% in a perchery system (Freire *et al* 2003), 60% in a commercial single-tier wire floor system (Nicol *et al* 2006), and 50 to 78% in free-range systems (Wilkins *et al* 2004). Elsewhere in the EU, Rodenburg *et al* (2008) recently reported a prevalence of keel-bone fractures in The Netherlands and Belgium of 97% in aviaries and of 82% in floor housing systems. The keel bone is highly susceptible to fractures due to its prominent anatomical position and damage can occur as a result of accidental impacts with equipment, perches or any hard objects within the house. These fractures and deformities are most likely to be associated with the presence of perches (Scholz *et al* 2008). Keel fractures may also occur as a result of wing flapping itself, as the flight muscles create forces on the skeletal bone (Alexander 1981). In non-cage systems, keel fractures can occur when hens access the perches incorrectly or due to landing accidents and flight (Gregory & Wilkins 1996; Freire *et al* 2003; Fleming *et al* 2004). Keel fractures also occur in conventional cage systems at a lower prevalence (0.2%: Gregory *et al* [1990]; 17.7%: Sherwin *et al* [2010]). Furnished cages seem to result in intermediate levels of damage (62%: Rodenburg *et al* [2008]; 31.7%: Sherwin *et al* [2010]). These fractures are mainly due to osteoporosis due to inability of birds to exercise fully (Fleming *et al* 1994; Keutgen *et al* 1999; Whitehead & Fleming 2000; Jendral *et al* 2008).

There is a positive correlation between bodyweight and bone strength (Knowles *et al* 1993; Vits *et al* 2005). However, the incidence of fractures also increases with bodyweight, because the increase in the skeletal bone strength is not enough to bear the extra weight load (Knowles *et al* 1993). There is a strain difference in the susceptibility of bone to breakage (Budgell & Silversides 2004; Riczu *et al* 2004) and this may be due to the differences between species or lines in bone structures, bodyweight and calcium metabolism (Clark *et al* 2008).

The impact of these fractures on the economics of egg production and egg quality is not clear and no previous studies have been carried out examining these relationships at an individual bird level. Thiruvenkadan *et al* (2010) suggested that egg production may decrease as a result of bone breakage during the production period, but there is no empirical evidence for this. If the production of birds with fractures is affected this could be an indicator of metabolic stress, difficulty accessing feed or stress associated with pain. It might also motivate farmers and breeding companies to take the problem more seriously.

Eggshell defects or abnormalities can be used as a non-invasive method to detect stress in laying hens (Hughes *et al* 1986). The eggshell constitutes the skeletal or external support of the egg (Ar *et al* 1979) and, as such, eggshell quality is very important to the poultry industry (Takahashi *et al* 2009). Any abnormalities or cracks of the eggshell mean that the eggs cannot be sold as first class, diminishing their economic value. These cracks may also have an effect on the internal and external quality of egg and food safety (because of bacterial invasion and contamination), thereby affecting marketability.

The impact of bone fractures on bird welfare is uncertain and it is not clearly known whether all fractures are painful or the extent to which they restrict or compromise bird movement and activity. Bone fractures in humans lead to an experience of pain (Yates & Smith 1995) and it is likely that keel fractures also cause pain in hens. However, most birds seem to survive after fractures occur and egg producers are unable to detect birds with fractures based on their overall behaviours. Despite the apparent lack of obvious change in bird behaviour, we should not interpret this as evidence that such fractures are inconsequential. What is required is a systematic examination (of individual birds) to determine whether healed fractures are associated with residual inflammation (Turner *et al* 1986; Purohit 2006) around the fracture site and whether they impact on bird movement.

To assess mobility we need to examine bird movement in the situations that require considerable physical effort. In the current study, we focused on perching activity, as it requires flight, balance, muscle strength, and motor and spatial skill. Domestic fowl are similar to their wild ancestors in terms of their motivation to perch in order to safeguard themselves from the floor predators (Newberry *et al* 2001), although their flight control is poorer, possibly due to the higher wing loading and heavier bodyweight of modern hens (Moinard *et al* 2004a). Laying hens prefer to access higher perches (Olsson & Keeling 2000; Wichman *et al* 2007; Struelens *et al* 2008; Schrader & Müller 2009). However, we also included a less demanding mobility test to increase the sensitivity of our results over a range of movement tasks.

The overall aims of this study were therefore to examine the impact of fractures of different severity level on egg production, eggshell quality, inflammation, mobility and behaviour. We predicted that birds with fractures would have lower bodyweights and poorer egg production due to greater difficulty in accessing feed or water resources or because of decreased feed consumption associated with pain or stress. Any of these factors could interfere with hormonal control of egg ovulation and formation. We predicted that hens with keel fractures would have a higher temperature at the fracture area compared to birds with no fractures as a result of residual inflammation. We also predicted that hens with fractures would show reduced agility and a lesser ability to access perches, particularly those that are high off the ground, in a range of mobility tests.

Materials and methods

Birds, classifying fracture severity, inflammation at fracture site and bone strength

Lohmann Brown laying hens (33 to 42 weeks of age) were collected from four different farms in four batches (Table 1). They had all been kept in free-range housing with access to perches. On-farm, before recruitment for the study, birds were examined by palpation of the keel to detect any abnormalities or fractures (Wilkins *et al* 2004). It is unlikely that palpation would cause pain to birds with healed fractures but we minimised any impact to birds that might have had more recent fractures by taking care not to manipulate or move the bone. Birds were selected according to the following criteria: (i) birds with a suspected severe healed keel fracture; (ii) birds with a suspected mild healed keel fracture; and (iii) birds with no suspected fracture. As the hens were unloaded from the transport crate they were identified individually with a coloured leg tag and housed together in one floor pen (3 × 3.5 m; length × width), bedded with wood-shavings, and provided with a metal, single-tier nest-box unit. During the initial holding period, perches were not provided to minimise the potential for new fractures. A perch was introduced two days before starting to record the perching behaviour. The hens were provided *ad libitum* with standard layers mash from two suspended poultry feeders and water from two suspended poultry drinkers. The lighting programme was 14 h light: 10 h dark and ambient temperature ranged between 19 and 21°C. The hens were kept for two weeks in one group at the experimental site for a separate experiment and then transferred to this experiment. Birds were acclimatised for three days before the start of the study. On the first day, hens were housed as a single group and on two subsequent days they were physically, but not visually, isolated overnight (from 1600 until 0900h the following day) in individual pens (50 × 50 × 100 cm; length × width × height) within the home pen to allow acclimatisation. The experiment was carried out under Home Office license.

Hens were weighed directly on arrival and then every alternate day during the experimental period (Mettler PE 12 (Mettler Instrument AG, Zurich, Switzerland with accuracy ± 0.001 kg) (Table 2). Thermal images of the keel area were taken on scheduled days for 76 hens (Table 2) by holding each hen on her back, with the ventral surface of the keel facing the thermal camera (FLIR ThermaCAM® E4, Professional Software, Kent, UK) and holding the feathers covering the keel area to one side. The images were taken at a distance of 1 m and analysed using specialised ThermaCAM® Reporter 2000 (Professional Software, Kent, UK). At the end of the experiment, hens were euthanised by cervical dislocation and dissected to validate the presence and severity of any keel fractures. Following dissection, the keel bone was removed, cleaned of all adhering soft tissue, and the severity of damage categorised using a five-point scale (Wilkins *et al* 2011). Keel-bone peak breaking strength was determined by three-point compression testing using a Stevens CR analyser (Mechtric Engineering Ltd, Benfleet, Essex) at two positions on the keel bone (area A, directly below the manubrial spine and area B, mid-lateral surface) (Nicol *et al* 2006).

Table 1 Details of the farms of origin of each batch of hens.

Batch number	Number of birds used in current experiment	Flock size	Age (weeks) when obtained for current experiment	Percentage egg production in flock of origin	Percentage mortality in flock of origin	Percentage keel fractures in flock of origin
1	12	6,793	40	85	2.8	20
2	32	16,000	39	92	0.3	25
3	32	12,000	42	90	1	30
4	26	12,000	33	91.3	0.4	15

Table 2(a) Testing protocol (days 1–10)

Procedure	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 9	Day 10
Group acclimatisation to the home pen	x									
Weighed	x		x		x		x		x	
Thermal image	Batch 1, 2 & 3			Batch 1, 2 & 3						Batch 1, 2 & 3
Home pen perch test (1230–1530h)			Batch 2 & 3*		Batch 2 & 3, 50 cm		Batch 2 & 3, 100cm		Batch 2 & 3, 150 cm	
Housed overnight in individual pens for egg collection		x**	x	x		x	x	x	x	
Eggs collected at 0900h				x		x	x	x	x	x
Training for mobility tests										
Mobility tests										
Behavioural observation (0830–1230h)										

Table 2(b) Testing protocol (days 11–20)

Procedure	Day 11	Day 12	Day 13	Day 14	Day 15	Day 16	Day 17	Day 18	Day 19	Day 20
Group acclimatisation to the home pen	x									
Weighed			x		x		x		x	
Thermal image									Batch 1, 2 & 3	
Home pen perch test (1230–1530h)										
Housed overnight in individual pens for egg collection										
Eggs collected at 0900h										
Training for mobility tests	Batch 1, 2 & 3	Batch 1, 2 & 3	Batch 1, 2 & 3							
Mobility tests				Batch 1, 2 & 3		Batch 1, 2 & 3		Batch 1, 2 & 3		
Behavioural observation (0830–1230h)									Batch 2 & 3	Batch 2 & 3

x Applies to birds from all four batches.
* Initial acclimatisation at perch height 50 cm.
** Initial acclimatisation to overnight pens without egg collection.
Video-recorded results of 150 cm perch height were excluded because only a very few number of birds accessed this perch height (2 or 3 birds).

https://doi.org/10.7120/096272812799129376 Published online by Cambridge University Press

Egg-quality assessment

Hens were physically but not visually isolated from 1600 until 0900h the next day in individual pens within the home pen to obtain individually identifiable eggs for egg-quality assessment. This procedure was followed for seven consecutive days (Table 2). During isolation, each hen was provided with *ad libitum* food and water and additional mealworms to familiarise the hens with this food, which was used to motivate the hens in subsequent mobility or agility tests. Eggs were labelled individually after collection and the following measures of egg quality were taken within 15 h of collection. External appearance: eggs were examined for deformed shape; shell calcification; or blood on surface of shell, classified as 'Yes' or 'No' for each individual parameter. Egg weight: measured using the Sartorius 1202 MP balance (GmbH, Gottingen, Germany) with accuracy ± 0.01 g. Egg length and width (mm): measured using an electronic digital calliper with accuracy ± 0.001 mm. Egg-shape index: calculated as egg width/egg length × 100 (Das *et al* 2010). Eggshell thickness (mm): measured using an electronic digital calliper once the eggshell was broken, taken as the mean of measures from both ends of the egg and from the middle (Das *et al* 2010). Eggshell weight (g): measured using the Sartorius 1202 MP balance after the eggshell had been washed and left to dry overnight at room temperature. Eggshell percentage: calculated as shell weight/egg weight × 100. Egg-surface area cm^2: calculated as $= 3.9782W^{0.7056}$ where $W =$ egg weight (g) and 3.9782 and 0.7056 are constants (Sezer 2007). Eggshell density or shell weight per unit surface mg cm^{-2}: calculated as shell weight (mg)/egg surface area) (Abdallah *et al* 1993; Vits *et al* 2005). Egg-production percentage was calculated as the average of seven days of egg production.

Mobility tests

On day 10, birds were housed again as one group for the whole 24-h period. They maintained their individual tags for the purposes of identification. Mobility or agility tests included home-pen, free-perch access, a walking velocity test, a flying and landing test and behavioural observations were carried out after appropriate training (Table 2). Four birds were excluded from mobility or agility tests because they could not be trained. The tests comprised the following.

Home-pen, free-perch access

A soft wood perch was introduced inside the home pen on days three and four for familiarisation. The dimension of the perch was 4 × 4 cm (length × width) in cross-section with a rounded edge and provided 25 cm perch length per bird. Video recordings of bird activity in the home pen were carried out for 3 h continuously from 1230 to 1530h for each single perch height: 50, 100 and 150 cm on days 5, 7 and 9, respectively. When a new perch was added to the pen the previous one was removed (Table 2).

Walking velocity test

Training for the walking velocity test was carried out by catching the hens and carrying them to the test room individually in a single box; hens could hear but not see other birds. The hens were trained to walk along a 3-m walkway to reach a mealworm food reward without any obstacles. Such tests have been used before to study motivation in broilers (McGeown *et al* 1999; Bokkers & Koene 2002) but we adapted our procedures from both of them to fit layers. Food was withdrawn 2 h before morning training and testing, but 1 h before afternoon training. The longer morning withdrawal period was to compensate for the fact that the hens' crops were fuller at this time, following their peak feeding after light onset. A hen was defined as 'trained' when she started moving towards the food after a latency of not more than 10 s. The test was carried out in trained hens by placing concrete blocks (40 × 20 × 10 cm; length × width × height) on the walkway and measuring the time taken for the hen to reach each obstacle and reach the end of the walkway. The hens were then given two trials per day on days 14, 16 and 18 with the first obstacle (height 20 cm) placed at 85 cm from the start, and the second (height 30 cm) placed at 175 cm from the start. The test was terminated after 5 min if the bird had not completed the course.

Flying and landing test

Training was carried out by catching the hens and carrying them to the test room individually in a single box; hens could hear but not see other birds. Food was withdrawn 2 h before morning training and testing time and 1 h before afternoon training. The aim of these tests was to assess the willingness of hens to fly up to a perch (flying test) or to descend from a raised perch to the ground (landing test); in both cases to access a mealworm food reward.

During training for the flying test, hens were picked up and lifted to one end of a perch, at a height of 20 cm from the ground. The hens were allowed to walk along the perch to reach a highly palatable food at the other end of the perch. Hens were defined as 'trained' if they started to move towards the food after a latency of not more than 10 s and ate the provided food. Trained hens were then placed individually on the floor of the pen and an observer assessed the time taken for hens to fly up to the perch.

During training for the landing test the hens were picked up and lifted onto the same perch, 20 cm from the ground and allowed to see a highly palatable food on the floor. Hens were defined as 'trained' if they began moving towards the food on the floor after a latency of not more than 10 s and ate the provided food. Trained hens were then placed on the perch and allowed to see mealworms on the floor. Again, an observer assessed the time taken for birds to move from the perch to the ground in the same order. Three perch heights (50, 100 and 150 cm) were evaluated in the same sequence for flying and landing tests. For both tests, hens were returned to the home pen without any food reward if they did not complete the test within 5 min.

Behavioural observation

Direct observations were conducted in the home pen with group-housed birds from batch two and three, by scan sampling every 10 min continuously from 0830 to 1230h on days 19 and 20 (Table 2) to record the behaviours of hens with and without keel fractures. The observer stood inside

https://doi.org/10.7120/09627281279912937 6 Published online by Cambridge University Press

the pen 10 min before starting the scan sampling to allow the hens to acclimatise. The recorded behaviours were foraging, eating, walking, preening, perch using (standing or sitting on the perch), sleeping, standing, drinking, spontaneous flight up to 100-cm perch height and nesting.

Statistical analysis

Statistical analysis was carried out using PASW Statistics (SPSS version 18.0 for Windows). An independent *t*-test was used to compare means from hens with and without keel fractures and analysis of variance to detect any effects of batch. For more detailed statistical examination, hens were grouped into four categories based on keel dissection at the end of the study (no fracture, fracture score one, fracture score two, and fracture score three). We did not find any birds with fracture score four. Linear regression and Pearson correlations were used to analyse the relationship between the fracture severity and different measured parameters.

Results

There was no significant effect of batch on the measured parameters except for the scan samples of eating and preening behaviour, where batch one ate less and preened more than batch two.

Bird assessment and thermal image

Palpation showed 62% of birds had fractures and 38% of birds were classified as no fracture but after dissection we found 25% of birds had no fractures. Based on dissection, the initial analysis grouped hens into two categories: hens with no fractures and those with fractures, regardless of severity. The temperature of the keel area was significantly higher in hens without keel fractures than hens with keel fractures (Table 3). Keel-fracture severity was negatively correlated with keel temperature (df = 1, 74 $F = 5.27$, $P = 0.025$, $\beta = -0.22$).

Egg quality assessment

There was no significant difference in bodyweight between hens with and without keel fractures. The external egg appearance did not show any signs of calcification, deformation or blood spots. Hens free from keel fractures had stronger keel bones in area B ($P = 0.000$). They also laid eggs with greater eggshell weight ($P = 0.04$). There was also a tendency for hens with no keel fractures to lay heavier eggs ($P = 0.07$), with greater surface area ($P = 0.07$) (Table 3). Keel-fracture severity had a negative significant relationship with egg weight (df = 1,100, $F = 5.47$, $P = 0.02$, $\beta = -0.99$), egg surface area (df = 1,100, $F = 5.40$, $P = 0.02$, $\beta = -0.81$) and a highly negative significant relationship with the keel-bone strength in area B (df = 1,100, $F = 13.12$, $P = 0.000$, $\beta = -0.89$) (Table 4).

The keel-bone strength in area A was positively correlated with keel-bone strength in area B ($r = 0.48$), and with shell density ($r = 0.20$). Egg weight was positively correlated with shell weight ($r = 0.66$) and egg surface area ($r = 0.97$) but was negatively correlated with shell percentage ($r = -0.21$). Shell weight was positively correlated with shell thickness ($r = 0.24$), shell density ($r = 0.79$), shell percentage

Table 3 Mean (± SEM) of keel-area temperature, egg quality, bodyweight and keel-bone strength in hens with and without keel fractures (classified following keel-bone dissection).

Factor	Hens with no keel fractures	Hens with keel fractures
Number of birds	16	60
Temperature of keel area	37.90 (± 0.17)**	37.29 (± 0.12)
Number of birds	26	76
Number of eggs	172	473
Egg weight (g)	63.25 (± 1.10)	61.26 (± 0.52)
Egg-shape index (%)	76.95 (± 0.45)	77.74 (± 0.22)
Egg-surface area (cm^2)	74.16 (± 0.91)	72.51 (± 0.43)
Shell thickness (mm)	0.41 (± 0.006)	0.41 (± 0.005)
Shell weight (g)	5.81 (± 0.09)*	5.57 (± 0.06)
Shell percentage (%)	9.21 (± 0.10)	9.10 (± 0.08)
Shell density (mg cm^{-2})	78.34 (± 0.77)	76.76 (± 0.70)
Bodyweight (kg)	1.83 (± 0.03)	1.81 (± 0.02)
Egg production (%)	94.51 (± 1.39)**	89.10 (± 1.58)
Keel strength area A (kg)	28.53 (± 1.06)	26.38 (± 0.61)
Keel strength area B (kg)	15.10 (± 0.55)**	12.64 (± 0.31)

Means in the same row differ significantly (*t*-test).
* $P < 0.05$, ** $P < 0.01$. Area A: directly below the manubrial spine; Area B: mid-lateral surface.

Table 4 Linear Regression of keel-area temperature, egg-quality parameters and keel-bone strength in hens with different keel-fracture severity (classified following keel-bone dissection).

Traits	df	*F*	*P*-value	β
Average keel-area temperature	1, 74	5.271	0.025	–0.22
Average egg weight (g)	1, 100	5.469	0.021	–0.985
Average egg-shape index (%)	1, 100	1.902	0.171	0.248
Average egg-surface area (cm^2)	1, 100	6.646	0.011	–0.812
Average eggshell thickness (mm)	1, 100	0.028	0.867	0.001
Average eggshell weight (g)	1, 100	0.681	0.411	–0.040
Average eggshell percentage (%)	1, 100	1.731	0.191	0.079
Average eggshell density (mg cm^{-2})	1, 100	0.558	0.457	0.320
Average egg production percentage during 7 days	1, 100	0.218	0.642	–0.524
Keel strength area A (kg)	1, 100	3.006	0.086	–0.821
Keel strength area B (kg)	1, 100	13.12	0.000	–0.886

Area A: directly below the manubrial spine; Area B: mid-lateral surface. β: Regression coefficient.

https://doi.org/10.7120/096272812799129376 Published online by Cambridge University Press

Table 5 Mean (± SEM) of walking velocity, flying, landing, keel-bone strength and spontaneous perch access in hens with and without keel fractures (classified following keel-bone dissection).

Factor	Hens with no keel fractures	Hens with keel fractures
Number of birds	15	49
Spontaneous access perch height 50 cm	11.33 (± 2.10)	9.14 (± 1.03)
Spontaneous access perch height 100 cm	3.67 (± 0.69)	3.04 (± 0.37)
Number of birds	16	56
Reach first obstacle (s)	1.92 (± 0.71)	2.86 (± 0.34)
Reach second obstacle (s)	5.61 (± 1.67)	10.44 (± 1.37)
Reach food (s)	9.41 (± 2.15)*	16.74 (± 2.11)
Flying from ground to perch height 50 cm (s)	134.38 (± 31.97)	151.47 (± 16.45)
Flying from ground to perch height 100 cm (s)	203.98 (± 30.39)	231.04 (± 11.40)
Landing from 50 cm perch height to floor (s)	9.33 (± 2.02)*	33.63 (± 9.18)
Landing from 100 cm perch height to floor (s)	25.90 (± 6.94)**	80.10 (± 11.99)
Landing from 150 cm perch height to floor (s)	78.70 (± 24.50)	127.78 (± 12.57)
Keel strength area A (kg)	33.24 (± 1.16)**	26.08 (± 0.65)
Keel strength area B (kg)	15.49 (± 0.63)**	12.50 (± 0.34)

Means in the same row differ significantly (*t*-test). * $P < 0.05$; ** $P < 0.01$.
Area A: directly below the manubrial spine; Area B: mid -ateral surface.

Table 6 Linear Regression of home-pen, free-perch access, walking velocity, flying, landing and keel-bone strength in hens with different keel-fracture severity (classified following keel-bone dissection).

Traits	df	*F*	*P*-value	β
Spontaneous access perch height 50 cm	1, 62	4.87	0.031	–1.808
Spontaneous access perch height 100 cm	1, 62	2.97	0.090	–0.502
Average of time taken to reach 1st obstacle	1, 70	3.11	0.082	0.509
Average of time taken to reach 2nd obstacle	1, 70	4.00	0.049	2.11
Average of time taken to reach food	1, 70	2.61	0.110	2.62
Average of time taken to fly from ground to perch 50 cm height	1, 70	1.20	0.276	14.97
Average of time taken to fly from ground to perch 100 cm height	1, 70	4.54	0.037	21.69
Average of time taken to land from perch 50 cm height to ground	1, 70	1.51	0.223	8.33
Average of time taken to land from perch 100 cm height to ground	1, 70	7.25	0.009	23.75
Average of time taken to land from perch 150 cm height to ground	1,70	4.37	0.040	21.80
Keel-bone strength area A (kg)	1, 74	22.97	0.000	–2.620
Keel-bone strength area B (kg)	1, 74	26.29	0.000	–1.389

Area A: directly below the manubrial spine; Area B: mid lateral surface. β: Regression coefficient.

($r = 0.60$), egg surface area ($r = 0.64$) and keel-bone strength in area A ($r = 0.23$). Shell thickness was positively correlated with shell density ($r = 0.30$) and shell percentage ($r = 0.28$). All these correlations were significant.

Mobility tests

Home-pen, free-perch access

There was no significant difference between hens with and without keel-bone fractures in frequency of using perches (Table 5). There was a negative relationship between fracture severity and the frequency of accessing the 50 cm perch (Table 6).

Walking velocity

Hens without keel-bone fractures were quicker to successfully complete the walkway test with the obstacles than hens with keel fractures (Table 5). There was a positive relationship between keel-fracture severity and the time taken to reach the second obstacle (df = 1, 70, $F = 4.00$, $P = 0.049$, $\beta = 2.11$) (Table 6).

Flying and landing

The latency to fly from the ground to all perch heights was shorter in hens free from keel fractures, and their latency to fly down and land on the floor from all perch heights was shorter than for hens with keel fractures (Table 5). The difference for landing was significant from the 50- ($P = 0.01$) and 100-cm ($P = 0.000$) perch heights, but failed to reach significance from the 150-cm perch height ($P = 0.07$) (Table 5).There was an overall positive relationship between keel-fracture severity and the time to flying or landing. This relationship was statistically significant for flying from the ground to perch height 100 cm (df = 1, 70, $F = 4.54$, $P = 0.037$, $\beta = 21.69$), landing from perch height 150 cm (df = 1, 70, $F = 4.37$, $P = 0.040$, $\beta = 21.80$) and highly significant for landing from perch height 100 cm (df = 1, 70, $F = 7.25$, $P = 0.009$, $\beta = 23.75$) (Table 6).

Keel-bone strength

Hens without a keel fracture had the strongest keel bones in areas A and B and there was a highly significant difference between these hens and hens with keel fractures regardless of severity (Table 5). There was a negative relationship between keel-fracture severity and keel-bone strength in both area A (df = 1, 74, $F = 22.97$, $P = 0.000$, $\beta = –2.62$) and B (df = 1, 74, $F = 26.29$, $P = 0.000$, $\beta = –1.39$) (Table 6).

Behavioural observation

The scan-sampling behaviour data showed that there was no significant difference in the frequency of measured behaviours between hens with and without keel-bone fractures except for sleeping. Hens with no keel fractures spent less time sleeping on the floor than hens with keel-bone fractures (0.00 [± 0.00] vs 0.18 [± 0.09]). There was a negative relationship between keel-fracture severity and drinking (df = 1, 62, $F = 5.17$, $P = 0.026$, $\beta = –0.29$) and between accessing the 100-cm perch (df = 1, 62, $F = 6.12$, $P = 0.016$, $\beta = –0.74$). There was a positive relationship between keel-fracture severity and preening (df = 1, 62, $F = 4.57$, $P = 0.036$, $\beta = 0.44$) (Table 7).

https://doi.org/10.7120/096272812799129376 Published online by Cambridge University Press

Discussion

The primary objective of this study was to investigate the effect of keel-bone fractures on the temperature of keel area, egg quality, activity and behaviour. Thermal imaging is a non-invasive tool to detect inflammatory processes which are accompanied by changes in the skin temperature in different animal species (Purohit 2006). The higher temperature measured over the keel area in hens without keel fractures in comparison to those with keel fractures was unexpected. It is possible that fractures of the keel may, over a period of time, result in disuse of the *pectoralis major* and *supracoracoideus* muscles particularly during the early stages of remodelling and healing, leading to subsequent muscle degeneration or disuse atrophy and lowering of the temperature in the affected area. The hens in this experiment had old, healed, keel-bone fractures and a different result might be expected with more recently fractured bones.

Hens without keel fractures had a greater egg production, as predicted by Thiruvenkadan *et al* (2010). This difference in production between hens with and without keel fractures may occur if pain or stress disrupts the hormones which are responsible for ovulation or decrease the responsiveness of granulosa cells to luteinising hormone, as has been found for heat-stressed birds (Donoghue *et al* 1989; Novero *et al* 1991).

Shell thickness was similar in hens with and without keel fractures, although egg weight was heavier in hens with no fractures. Heavier eggs generally have a larger surface area (Buss 1988), requiring more calcium for eggshell formation. Where keel fractures occur calcium may be used in bone healing and callus formation instead of eggshell formation (Scholz *et al* 2008). It is possible that hens without keel fractures laid heavier eggs because they were more acclimatised to the new environment, and hence were less stressed than hens with keel fractures. Further investigation would be necessary to confirm whether fractures do affect the capacity to adapt or cope with a new environment. Hens with no keel fractures had the strongest keel bones. In addition, keel-bone strength, in area A and B, showed a non-significant, weak correlation with egg production. There have been conflicting results on the relationship between bone strength and egg production. Whilst some authors have found no relationship (Rennie *et al* 1997; Bishop *et al* 2000; Jendral *et al* 2008), others have reported that birds with stronger bones might provide less calcium for eggshells (Cox & Balloun 1971; Harms & Arafa 1986; Graveland & Berends 1997; Whitehead 2004). However, our results are in accordance with the majority of studies that have found that good bone strength and good eggshell quality can go together (Hurwitz 1964; Riczu *et al* 2004; Schreiweis *et al* 2004).

This is the first time that a walking velocity test has been used in hens to assess the impact of injury, and it was adapted from a similar test in broilers. The time taken for hens with no fractures to finish the runway test was shorter than for hens with keel fractures and suggests either a physical impairment to moving or a reduced motivation to move, perhaps because of associated pain. Similar results were obtained when comparing broilers suffering from lameness with birds free from lameness (McGeown *et al* 1999).

Table 7 Linear Regression of scanned behaviour observation every 10 min summed over the 4 h and bodyweight in hens with and without keel fractures (classified following keel-bone dissection).

Traits	df	*F*	*P*-value	β
Forage	1, 62	0.0047	0.829	–0.062
Eat	1, 62	0.542	0.464	–0.288
Walk	1, 62	0.050	0.0823	–0.040
Preen	1, 62	4.573	0.036	0.435
Perch	1, 62	0.254	0.616	–0.173
Sleep	1, 62	1.391	0.243	0.075
Nest	1, 62	0.188	0.666	0.089
Drink	1, 62	5.168	0.026	–0.293
Stand	1, 62	1.157	0.286	0.147
Access 100-cm perch height	1, 62	6.124	0.016	–0.737

β: Regression coefficient.

The flying and landing tests showed that birds without fractures took a shorter time to fly from the ground to perches of different heights and to land from these perches to the ground, but that, generally, all birds were quicker to land rather than to fly upwards and this may be because flying upwards is against gravity so that hens need more wing flapping and more power to fly upwards than to land. Hens with no fractures used perches more frequently than hens with fractures. This may be because the keel fractures or deformities interfere with the ability of hens to use and rest on perches either due to a reduced motivation, as suggested earlier, or a decrease in the amount of force that an injured bird can generate during wing flapping for flight (Sandilands *et al* 2009). Again, hens may try to avoid muscle forces that press on the keel bone during flying or landing due to associated pain. However, when negotiating between perches, (Scott *et al* 1997; Moinard *et al* 2004a,b) found that birds could cross perches more successfully by flying upwards rather than downwards.

Hens with no fractures were more active during the behavioural observation period. But, hens suffering from fractures spent more time sleeping on the floor than hens free from fractures. More time was spent in the nest by hens with keel fractures, possibly because they used the nest-boxes for resting. Foraging behaviour did not differ between hens with and without keel fractures. This may be because foraging is a behaviour priority for hens (Weeks & Nicol 2006).

We conclude from our data that keel-bone fractures may prevent hens from performing normal behaviours or accessing resources, so keel fractures may be considered detrimental to hens' welfare. This may be similar to lameness in broilers. Butterworth *et al* (2002) found that severely lame broilers become moribund and dehydrated because they are unable to reach resources.

Animal welfare implications and conclusion

The occurrence of keel-bone fractures in laying hens is high. This study found keel-bone fractures to reduce egg production and quality and potentially bird welfare. Hens with keel fractures laid a lower number of eggs with lower egg-quality scores and reduced keel-bone strength. They took longer to successfully complete the walkway, flying and landing tests and accessed perches less frequently than hens without fractures. Further investigation is necessary to determine if keel fractures are accompanied by pain perception, which would further impact on hen welfare.

Acknowledgements

The authors would like to thank the BBSRC for funding the study and The Egyptian Government (The Egyptian Educational and Cultural Bureau) for funding Mohammed AF Nasr.

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