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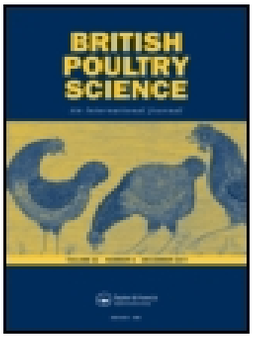
**6 authors**, including:

Carlos Sanchez
Game & Wildlife Conservation Trust


Marta Alonso
Universidad de León


Emilio Jorge Tizado
Universidad de León


Jose A Armenteros
University of Castilla-La Mancha

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# Anti-predator behaviour of adult red-legged partridge (*Alectoris rufa*) tutors improves the defensive responses of farm-reared broods

C. SÁNCHEZ-GARCÍA[1], M.E. ALONSO, E.J. TIZADO[2], J. A. PÉREZ[3], J. A. ARMENTEROS, AND V. R. GAUDIOSO

*Department of Animal Production, Facultad de Veterinaria, University of León, Campus de Vegazana, Research Group on Game Species Breeding and Management, León, Spain, [1]The Game & Wildlife Conservation Trust, Burgate Manor, Fordingbridge, Hampshire, United Kingdom, [2]Department of Biodiversity and Environmental Management, University of León, Ponferrada, Spain, and [3]Global Sigma S.L., León, Spain*



**Abstract** 1. The aim of this work was to improve natural anti-predator behaviour of farm-reared gamebirds.
2. We evaluated the anti-predator behaviour of reared red-legged partridge *Alectoris rufa* chicks kept in brooder houses in large groups (>350 chicks), trained and not trained by parent red-legged partridges acting as experienced tutors.
3. The experiment consisted of two conditioned tests (a raptor model and a human) and two control tests, which were conducted during three consecutive phases of life (1–4, 15–17 and 30–32 d after hatching). The motor anti-predator behaviour, its duration, the intensity of response in chicks and alarm calls elicited by adults were recorded.
4. Tutors elicited aerial alarm calls (76% of tests) and showed prolonged crouching (59% of tests) in response to the raptor model whereas uttering the ground alarm call (73% of tests) and showing vigilance behaviour (78% of tests) was the main pattern during the human test.
5. Trained and not trained chicks showed similar motor behaviour in response to the raptor model (crouching) and the human test (escaping), but frequency of strong responses (all chicks responding) from chicks trained with tutors was double that of chicks trained without them, and chicks trained with tutors showed a higher frequency of long responses (41–60 s).
6. This study indicates that anti-predator training programmes before release may improve behaviour of farm-reared partridges which may confer benefits to survival of birds.

## INTRODUCTION

Every year, millions of farm-reared gamebirds are released across Europe for shooting purposes of which only few survive and even fewer reproduce (Sokos *et al.*, 2008). Several studies have shown that low survival and breeding success after release are in part due to a consequence of the rearing systems of commercial game farms, which typically ignore the importance of the development of natural behaviour including anti-predator responses (Csermely *et al.*, 1983; Dowell, 1990; Putaala and Hissa, 1998; Pérez *et al.*, 2010). If reared gamebirds learn correct anti-predator behaviour during the pre-release stage, this may increase their survival rates (WPSA and IUCN, 2009) and also guarantee appropriate behaviour for shooting.

Compared to commercially reared birds, partridges bred and reared by their natural or foster parents (i.e. natural and semi-natural rearing) have higher survival rates, as chicks learn from parents crucial anti-predator behaviour, though their breeding success is still lower than that of their wild counterparts (Buner *et al.*, 2011; Pérez *et al.*, 2015). Parent-rearing is the best option for re-establishing purposes when translocation of

Correspondence to: Carlos Sánchez-García, The Game & Wildlife Conservation Trust, Burgate Manor, Fordingbridge, Hampshire, SP6 1EF, United Kingdom. E-mail: csang@unileon.es
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wild birds is not possible (WPSA & IUCN, 2009), though it is much more expensive than commercial game rearing, and parent-reared gamebirds are currently not commercially available across Europe, except for the grey partridge *Perdix perdix* in the United Kingdom (www.perdixwildlifesupplies.co.uk). Hence, a possible alternative to improve the behaviour of farm-reared gamebirds may be to teach them at least some basic anti-predator behaviour during the rearing process.

Training anti-predator behaviour in captive animals for conservation purposes has been used in some species during recent decades, and there is evidence that behavioural conditioning to avoid predators is effective (Griffin *et al.*, 2000). Slaugh *et al.* (1992) imprinted chukar partridge *Alectoris chukar* chicks on adults and used a hawk model and dogs as aversive stimuli, resulting in a higher survival after release into the wild in comparison to the commercially reared birds. Zaccaroni *et al.* (2007) used repeated human intrusions as aversive stimuli for rock partridge *Alectoris graeca* poults and the escape response was elicited at greater distances in comparison to not-trained partridges.

Among Galliformes, one of the key factors for correct development of anti-predator behaviour are alarm calls attached with "wild-like" behaviour that a parent gives to its brood in the presence of a predator (Putaala, 1997). This effect is not restricted to genetically related parent birds, as foster parents are also able to modify the behaviour of unrelated young (Dowell, 1990; Bertin and Richard-Yris, 2004). Thus, in order to induce anti-predator responses in not-related chicks during the early stages of life, a possible improvement to current rearing methods may be the introduction of experienced foster parents (i.e. tutors) and the exposure to simulated predator events to these systems.

In this paper, we assess the behavioural responses towards artificial predator events of captive-held red-legged partridge adults and the effects of their behaviour on the responses of reared chicks under otherwise intensive commercial conditions.

## MATERIALS AND METHODS

### Game farm description

The research was conducted on a private intensive game farm in the province of León (north-western Spain), between 2008 and 2010. The farm was chosen based on its policy of avoiding the otherwise widespread hybridisation of red-legged partridges (hereafter simply red-legs) with the closely related but not native chukar partridge (Blanco-Aguiar *et al.*, 2008). Also, the selection criteria of breeding birds of the farm were not based on easy handling (tameness) or maximum egg production (Mourão *et al.*, 2010). Large outdoor enclosures (75 m length × 25 m width × 5 m height) with permanent natural ground cover and comparatively low stocking densities in the flight-pens (2–3 $m^2$/bird) were provided. Additionally, the farm's rural location and natural surroundings guaranteed the presence of avian predators such as the goshawk (*Accipiter gentilis*) and Eurasian eagle-owl (*Bubo bubo*) to which the adult partridges used in our experiments were exposed while kept in the outdoor pens. Apart from these improvements, the farm followed classical intensive rearing methods for red-legs (Sánchez-García *et al.*, 2009; González-Redondo *et al.*, 2010).

### Broods

A total of 6 standardised batches of 400–500 eggs were taken from 120 red-leg pairs, hatched in incubators at the same premises because commercial game farms typically rear partridges in large groups. Each of the 6 groups of chicks (further called simply broods) was kept on wood shavings close to a heat source within a protective circular ring (3.14 $m^2$, height 30 cm) for 7–10 d in separate 32 $m^2$ brooder houses, though each brood occupied one half of the brooder house (16 $m^2$, Figure 1). After this period, the chicks were given the whole brooder house allowing an approximate area of 335 $cm^2$/chick at a constant room temperature between 28°C and 30°C. A nutritionally balanced commercial food specific for gamebirds (NANTA, Nutreco España SA, Madrid), was provided *ad libitum* in 4 feeders together with fresh water in three water troughs. All individuals were subjected to daylight (14–15 h) and no cover or shelter for hiding was provided. Owing to fatalities which were usual within the commercial rearing environment (chicks dying from smothering after piling-up and starving during the first day), and a Colibacillosis outbreak, it was impossible to keep the group sizes constant and after 4 weeks, each brood had from 385 to 478 poults (Table 1).

### Tutors

Adult pairs of red-legs were used as experienced tutors. To avoid the maladaptation problems of wild red-legs to captivity (Sánchez-García *et al.*, 2011) and considering the experience collected in the well-studied grey partridge (Buner *et al.*, 2011), each pair consisted of a captive-reared female and a wild male caught under license in December–early January. These forced pairs of red-legs were housed in semi-natural breeding pens at the same game farm in January–early February (Figure 1). Only pairs which hatched their own chicks were used as tutors. Once the parent-hatched chicks were 8–9 d old, they were separated from their parents and incorporated


**Figure 1.** *Anti-predator training system: (a) semi-natural rearing pen, (b) a tutor with its brood, (c) brood house, (d) tutors in the cage with brood during phase 1, (e) phase 2 and (f) life-sized model of a flying raptor.*

**Table 1.** *Number of chicks and poults per brood and year and number of repetitions per test*

| Year | Brood | Chicks at start, *n* | Poults at 4 weeks, *n* | Raptor test | Raptor control | Human test | Human control |
|---|---|---|---|---|---|---|---|
| 2008 | Trained | 500 | 463 | 14 | 10 | 18 | 12 |
| | Not trained | 500 | 478 | 10 | 12 | 19 | 11 |
| 2009 | Trained | 400 | 385 | 14 | 11 | 17 | 11 |
| | Not trained | 500 | 474 | 10 | 11 | 18 | 11 |
| 2010 | Trained | 450 | 432 | 14 | 10 | 20 | 11 |
| | Not trained | 450 | 427 | 9 | 13 | 17 | 12 |
| Total | Trained | 1350 | 1280 | 42 | 31 | 55 | 34 |
| | Not trained | 1450 | 1379 | 29 | 36 | 54 | 34 |
| Grand Total | | 2800 | 2659 | 71 | 67 | 109 | 68 |

into the breeding stock which was not part of the experiment.

Due to the high number of chicks and the potential risks of disease and accidents, each pair of tutors was introduced into a separate experimental cage in the brooder house 4 h prior to the chicks (Figure 1). The cage was positioned at 22.5 cm above the ground in one corner of the brooder house to ensure visual contact between the tutors and the groups of incubator-hatched chicks. Food and water was provided *ad libitum*. Physical contact between chicks and tutors was prevented until the protective ring was removed at an age of 7–10 d. At the end of the experiment, tutors were transferred into the outdoor flight pens together with the trained group of juveniles. Once broods and tutors were placed in the brooder house, handling was avoided as much as possible. One week before the beginning of the experiments each year, tutors were subject to preventive treatment for parasite diseases (Flubendazole) and no medication was provided to the chicks unless disease symptoms were observed.

### Experimental design

According to the presence of tutors, two types of broods were studied: trained, where tutors were introduced in the brooder house and kept together with the brood and not trained, where the brood was reared without tutors.

Treatments were based on predation stimuli previously tested on grey partridges and ring-necked pheasants (*Phasianus colchicus*) (Dowell, 1990; Beani and Dessì-Fulgheri, 1998). Tutors and broods were exposed to 4 types of tests: Raptor test (*Rt*), a life-sized model of a flying raptor mounted on a stick was used. The model was shown across the two brooder house windows at a steady pace of 5 m/s, allowing the tutors and the chicks to see the "raptor" for one second; Raptor control (*Rc*), the stick without the raptor model was shown across the window at the same pace; human test (*Ht*), a person entered the brooder house avoiding noise for 10 s. During this test, levels of feed and water were checked quickly; human control (*Hc*), which consisted in opening the door partially for 2–3 s, was considered as control because of the slight noise and movement caused.

To understand the effects of consecutive artificial predator stimuli on tutors and considering that the anti-predator behaviour of broods may be influenced by age and escape ability, tests were conducted at three different life phases:

Phase 1: As the sensitive imprinting period in red-legs lasts 48 h after hatching (Csemerly *et al.* 1983), tests started in the first 1–2 d of life to ensure that they covered this crucial stage and continued until d 3–4. Apart from minimum contact with game farmers immediately after hatching, chicks were not subject to predatory stimuli before this phase.

Phase 2: As chicks start to flutter-fly from an age of 11–12 d, a second series of tests was carried out between 15 and 17 d after hatching.

Phase 3: Juvenile partridges are able to complete a medium distance flight from an age of one month. Therefore, a last series of tests were carried out between 30 and 32 d after hatching.

All tests were carried out in July and August, between 09:00–14:00 and 15:00–19:00. Tests were applied at least once but no more than twice per day. To avoid possible day-time effects, we swapped the time of the tests from morning to evening between days. We did not evaluate the behavioural responses of chicks after training without tutors as we opted for a subsequent radio-tracking study to compare the survival of trained and not trained broods after release, as this was taken as a measure of the ultimate effect of training (see Gaudioso *et al.*, 2011).

Cameras and a peep-hole in each brooder house allowed the investigators to assess whether the majority of chicks (>80%) were showing no anti-predator behaviour before the start of each test. No tests were conducted if the optimal temperature and humidity conditions of the brooder house were not present, as this could lead to either aggregation of the chicks under the heat source (cold) or the chicks being too dispersed (hot). Two video cameras (S130277, Superinventos S.L.) were fixed on the ceiling of the brooder house to record the behaviour of tutors and chicks. The cameras were connected to a central video recording unit, which consisted of a multiplexor (DVR Triplex 4/8, Superinventos S.L.), a video recorder and a monitor. A stereo microphone (YOGA, EM-278) connected to a digital audio recorder (PHILIPS Digital Voice TracerLFH 660) was used to record vocalisations.

### Behavioural study

The anti-predator motor behaviour of tutors and chicks observed after the presentation of the predator stimulus (named Behaviour) was divided into 4 exclusive and comprehensive classes (Prieto, 2008; Binazzi *et al.*, 2011): (1) vigilance, outstretched neck, upright posture and intermittent scanning of the head from side to side; (2) crouching (including freezing behaviour), a rapid squatting of the bird, followed by freezing and remaining motionless; (3) escape, walking and running, also including flying attempts; (4) note, any anti-predator response or other behaviour response different from 1 to 3 after 5 s of the presentation of the stimulus.

When a behavioural response was observed, its duration (Time, s) after the reaction of at least one adult or chick during the first 60 s after the stimulus. In adults, Time was counted until one adult turned to a not-anti-predator behaviour (mainly pecking around feeders, water troughs and the experimental cage) and in broods when at least 50% of the chicks responding turned to not-anti-predator behaviours. This variable was categorised in an ordinal scale as short (1–20 s), medium (21–40 s) and long (41–60 s). In a preliminary study, it was observed that the response of each chick was not entirely independent and when a stimulus was applied, broods responded as a whole or just a proportion of the chicks. Thus, we categorised the overall brood response (Intensity), as strong (all chicks responding) and weak (when not all the chicks responded, often continuing feeding and foraging after the stimulus). Finally, we recorded the type of first alarm call uttered by one of the adults within the first 5 s

(Sound), and classified as three types (Prieto, 2008; Binazzi *et al.*, 2011): aerial, short guttural "keer", usually related to the sight of a raptor; ground, high energy short calls, associated to ground-living predators; note, calls related to feeding, reproductive and social behaviour.

A total of 6 broods (three trained and three not trained) were studied and 315 observations were used for analysis (162 on trained and 153 on not trained). The number of observations carried out for each brood ranged from 50 to 56, and the number of repetitions for each test ranged from 9 to 20 per brood. Aiming to detect significant differences, in trained broods the number of raptor-trained (*Rt*) observations was deliberately higher in comparison to raptor-control (*Rc*) treatments as a higher variability in alarm calls was expected to be elicited by adults. For both types of broods, more human-test (*H*) observations were carried out as the levels of feed and water were quickly checked during these tests, reducing the total number of visits required to refill food and water troughs (Table 1). In the pair of adults used in 2010, the hen was removed during the first phase as she was suspected to suffer an infectious disease. In one trained brood, a Colibacilosis outbreak was detected in 2008 during the first phase of the experiment, resulting in 37 dead chicks.

### Data analysis

Log-linear models for contingency tables (i.e. generalised linear models, GLM) for count data by using Poisson distribution and the log link were used (Agresti, 2007). For adults, response variables tested were the count data of Behaviour, Time and Sound and the potential explanatory variables were Phase and Test. For broods, response variables were the count data of Behaviour, Time and Intensity and the potential explanatory variables were the Type of brood, Test and Phase. In each of 3 years, the study was conducted using two broods, one trained with tutors and the other not trained. The tests were not applied more than twice per day and phases were separated by at least for one week. The data were pooled together across years because the chicks came from the same game farm and husbandry and handling did not change during the 3-year period. Thus year was considered as a replication, not as an explicative factor of the study. The saturated-models for tutors (1) and for broods (2) were:

$$\log(m_{ijk}) = \lambda + \lambda_i^{\mathrm{X}} + \lambda_j^{\mathrm{Phase}} + \lambda_k^{\mathrm{Test}} + \lambda_{ij}^{\mathrm{X\cdot Phase}} + \lambda_{ik}^{\mathrm{X\cdot Test}} + \lambda_{jk}^{\mathrm{Phase\cdot Test}} + \lambda_{ijk}^{\mathrm{X\cdot Phase\cdot Test}} \quad (1)$$

$$\begin{aligned}\log(m_{ijkl}) = {} & \lambda + \lambda_i^{\mathrm{X}} + \lambda_j^{\mathrm{Phase}} + \lambda_k^{\mathrm{Test}} + \lambda_l^{\mathrm{Trained}} \\ & + \lambda_{ij}^{\mathrm{X\cdot Phase}} + \lambda_{ik}^{\mathrm{X\cdot Test}} + \lambda_{il}^{\mathrm{X\cdot Trained}} \\ & + \lambda_{jk}^{\mathrm{Phase\cdot Test}} + \lambda_{jl}^{\mathrm{Phase\cdot Trained}} \\ & + \lambda_{kl}^{\mathrm{Test\cdot Trained}} + \lambda_{ijk}^{\mathrm{X\cdot Phase\cdot Test}} \\ & + \lambda_{ijl}^{\mathrm{X\cdot Phase\cdot Trained}} + \lambda_{ikl}^{\mathrm{X\cdot Test\cdot Trained}} \\ & + \lambda_{jkl}^{\mathrm{Phase\cdot Test\cdot Trained}} \\ & + \lambda_{ijkl}^{\mathrm{X\cdot Phase\cdot Test\cdot Trained}}\end{aligned} \quad (2)$$

where $m$ corresponds to the cell frequencies, $\lambda$ represents the overall mean, $\lambda_i^{\mathrm{X}}$ the $i$-th marginal effect of the variable $X$ (being Behaviour, Sound or Time plus Intensity for brood), $\lambda_j^{\mathrm{Phase}}$, $\lambda_k^{\mathrm{Test}}$ and $\lambda_l^{\mathrm{Trained}}$ are the $j$-th, $k$-th, and $l$-th marginal effect of the other variables to test whether there were interactions with $X$, and the other terms were the corresponding interaction terms.

The GLM residuals and the interaction between categorical variables were checked using Chi-square tests. Differences with $P < 0.05$ were considered significant. Absolute percentages of count of Behaviour, Time, Intensity and Sound were presented in Tables and Figures. All statistical analyses were carried out using R software v.3.1.2 (R Development Core Team, 2009).

### Ethical statement

This study complies with the ethical guidelines of the International Society for Applied Ethology (Sherwin *et al.*, 2003). The experimental cage for the tutors was considered harmless and all tests were designed to avoid prolonged distress. Once the tests had finished on d 32, tutors were moved to outdoor flight pens together with trained broods.

## RESULTS

Significant effects of explanatory on response variables included in the models fitted for tutors and broods are shown in Table 2. The same shows the significance of the analysis of deviance between the null and fitted models.

### Tutors

Test was a key variable to explain tutors' Behaviour. In the *Rt*, the overall response was characterised by crouching (59.2%), whereas vigilance was the most frequent behaviour observed in the *Rc*, *Ht* and *Hc* tests (>64.7%, Figure 2). Fitted models explaining variation for Time in tutors included Test and Phase, with a higher number of long responses recorded in the *Rt* and *Ht* when compared to *Rc* and *Hc*, and the

**Table 2.** *Analysis of deviance of the several models fitted by selection of the terms in the full model (null). The fitted-model line shows the response (Behaviour, Time, Sound, Intensity) and explanatory variables (Test, Phase, Trained) for tutors and broods analysed and the significant interactions (P < 0.05) included. The P-value in the null line shows the significance in the analysis of deviance table between the fitted-model and the saturated-model*

| | df | Deviance | Residual df | Residual deviance | *P* (>Chi) |
|---|---|---|---|---|---|
| **TUTORS** | | | | | |
| *Fitted-model*: Behaviour + Phase + Test + Test × Behaviour | | | | | |
| Null | | | 47 | 307.43 | 0.156 |
| Behaviour × Test | 9 | 116.09 | 30 | 37.77 | <0.001 |
| *Fitted-model*: Time + Test + Phase + Test × Time + Phase × Time | | | | | |
| Null | | | 35 | 160.0 | 0.475 |
| Time × Test | 6 | 46.38 | 22 | 46.28 | <0.001 |
| Time × Phase | 4 | 28.57 | 18 | 17.71 | <0.001 |
| *Fitted-model*: Sound + Test + Phase + Test × Sound | | | | | |
| Null | | | 35 | 232.57 | 0.009 |
| Sound × Test | 6 | 153.93 | 22 | 40.80 | <0.001 |
| **BROODS** | | | | | |
| *Fitted-model*: Behaviour + Test + Phase + Test × Behaviour + Phase × Behaviour | | | | | |
| Null | | | 95 | 677.28 | 0.229 |
| Behaviour × Test | 9 | 412.07 | 78 | 136.05 | <0.001 |
| Behaviour × Phase | 6 | 55.50 | 72 | 80.54 | <0.001 |
| *Fitted-model*: Test + Phase + Time + Phase × Time + Test × Time + Trained × Phase × Time | | | | | |
| Null | | | 71 | 331.94 | 0.153 |
| Time × Test | 6 | 84.89 | 54 | 93.63 | <0.001 |
| Time × Phase | 4 | 42.27 | 60 | 178.52 | <0.001 |
| Time × Trained × Phase | 9 | 38.94 | 45 | 54.69 | <0.001 |
| *Fitted-model*: Test + Phase + Sound + Test × Sound + Phase × Sound + Trained × Phase × Sound + Test × Phase × Sound | | | | | |
| Null | | | 71 | 445.30 | 0.967 |
| Sound × Test | 6 | 101.62 | 54 | 115.21 | <0.001 |
| Sound × Phase | 4 | 133.27 | 60 | 216.84 | <0.001 |
| Sound × Trained × Phase | 9 | 45.54 | 45 | 69.67 | <0.001 |
| Sound × Test × Phase | 18 | 54.54 | 27 | 15.14 | <0.001 |
| *Fitted-model*: Test + Trained × Intensity + Test × Intensity + Test × Phase × Intensity + Test × Trained × Intensity | | | | | |
| Null | | | 47 | 257.82 | 0.120 |
| Intensity × Trained | 3 | 44.63 | 41 | 179.05 | <0.001 |
| Intensity × Test | 3 | 86.89 | 38 | 92.16 | <0.001 |
| Intensity × Test × Phase | 16 | 39.01 | 22 | 53.15 | <0.001 |
| Intensity × Test × Trained | 6 | 30.37 | 16 | 22.78 | <0.001 |

number of longer responses increased across Phases. As reported for Behaviour, the fitted model explaining Sound included Test; after the *Rt*, the aerial call was uttered in 76.2% of the tests and no calls were heard in 96.7% of *Rc*, whereas in the *Ht*, the *ground* call was the most uttered vocalisation (72.7%) and *no-alarm* call was heard in 76.4% of *Hc*.

## Broods

Fitted models for Behaviour of broods included Test and Phase. Crouching was the most frequent response in the *Rt* (trained = 73.7%, not trained = 65.5%), vigilance in the *Rc* (trained = 51.5%, not trained = 72.3%) and escape in the *Ht* (100% in both trained and not trained) and *Hc* (trained = 56%, not trained = 62%). Crouching was the main behaviour observed among phases in the *Rt*, though during phase 1 the other behaviours were observed at minor proportions. In the *Rc*, no anti-predator behaviour was observed in 61% of the tests, being vigilance the main behaviour in phases 2 (82%) and 3 (100%). In the *Hc*, vigilance and no anti-predator behaviour were the main responses in phase 1 (summing 78%), and escape was the main behaviour in phases 2 and 3 (>90%).

Of all models tested for Time, the fitted effects included Test, Phase and a Phase × Type of brood interaction. Pooling trained and not trained broods, a higher frequency of long responses were recorded in the *Ht* (50%) and *Rt* (34%) compared to *Rc* (0%) and *Hc* (6%) tests, and longer responses were more frequent in phase 3 (44%) compared to phases 2 (21%) and 1 (16%). Overall, trained broods showed a higher frequency of longer responses across phases for the *Rt* (69%) and *Ht* (44%) tests when compared with not trained broods (24% for both tests) (Figure 3(a)).

The fitted model explaining Intensity included Type of brood, Test, and interactions between Test × Phase and Test × Type of brood.

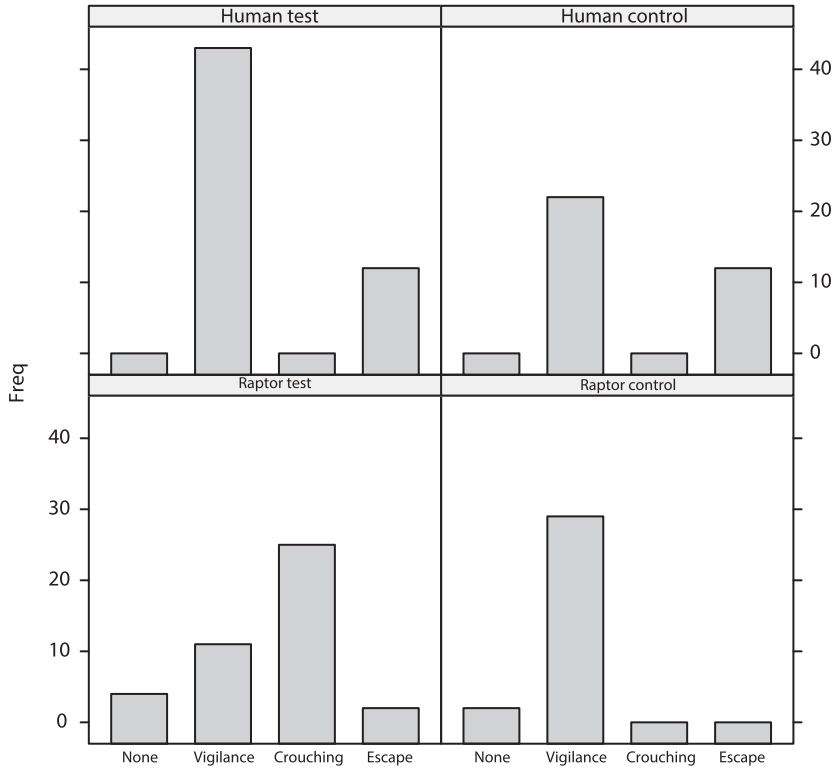


**Figure 2.** *Frequency of behaviour in tutors considering number of tests (Rt = raptor test; Rc = raptor control; Ht = human test; Hc = human control).*

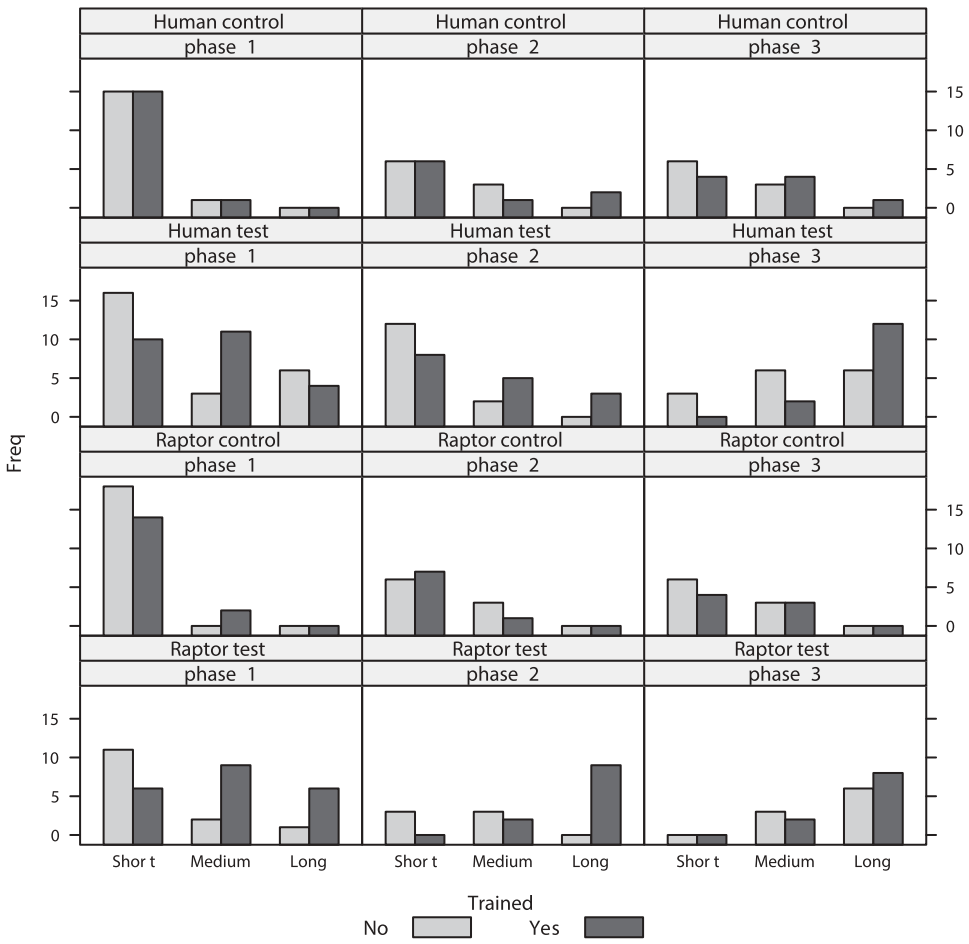


**Figure 3.** *(a) Time (left) and (b) Intensity (right) in broods considering number of tests. Rt = raptor test; Rc = raptor control; Ht = human test; Hc = human control; across Phases (Ph) and Type of brood (Yes = trained, No = not trained).*

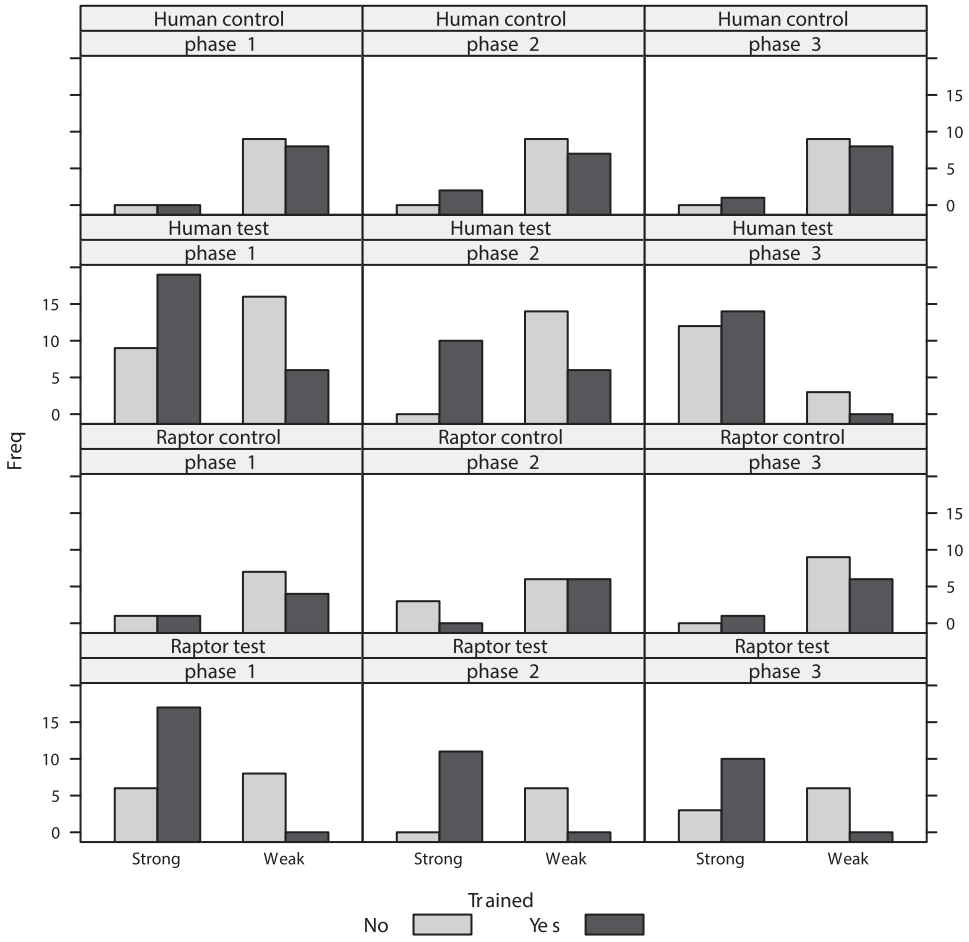


**Figure 3.** *(Continued)*

For both trained and not trained broods, a higher frequency of strong responses were recorded in the *Rt* and *Ht* when compared to their controls, although the overall frequency of strong responses from trained broods (62%) was approximately double that of not trained broods (33%). In trained broods, frequencies of strong responses decreased across phases in the *Rt*, whereas in the *Ht* a decrease of strong responses was observed in phase 2 when compared to phases 1 and 3. This was also observed in not trained broods for both *Rt* and *Ht* tests (Figure 3(b)).

## DISCUSSION

To our knowledge, this is the first study providing results of an anti-predator training technique for gamebirds using experienced tutors under commercial rearing. Although the type of motor behaviour was similar among trained and not trained broods, significant differences were found in the duration and intensity of responses, as trained chicks showed a higher frequency of long and strong responses when compared to not trained broods. Hence, these results confirm that the anti-predator behaviour of broods was affected by the responses of tutors (Dowell, 1990; Bertin and Richard-Yris, 2004).

Despite the space limitation in the experimental cage, tutors showed natural anti-predator behaviour in response to the stimuli given, which is in agreement to similar studies for the same species in captivity (Venturato *et al.*, 1997; Binazzi *et al.*, 2011). Undoubtedly, tutors were affected by the experimental cage, as they were not able to flee into cover (Beani and Dessì-Fulgheri, 1998). However, as observed in the wild by Pintos *et al.* (1985), their responses were affected by the nature of the stimulus; the *Rt* resulted in prolonged crouching and aerial alarm call, whereas the *Ht* elicited prolonged vigilance and ground alarm call. It seemed that in tutors no habituation to tests occurred, as the fitted model explaining type of motor behaviour did not include phase, though the duration of behaviour (Time) was best explained by the type of test and phase. It may be that the experimental cage disrupted the stress response during the first day in the cage, consistent with the findings of Dickens *et al.* (2009) for Chukar partridges.

The anti-predator behaviour of broods did not correspond entirely to that observed in the wild (Sánchez-García, personal observation), but

both trained and not trained broods showed inherited galliforme anti-predator behaviour by crouching in the presence of a raptor and escaping once encountering a human (Pintos *et al.*, 1985). While fitted models explaining variation in motor behaviour did not include type of brood, the explanatory variables type of test, phase and their interactions were included in the best models for duration of behaviour and intensity. Hence, experienced adults may have transmitted predator avoidance (fearful) behaviour to trained chicks, which is in agreement with previous studies in Galliformes (Dowell, 1990; Slaugh *et al.*, 1992; Bertin and Richard-Yris, 2004). We did not test whether alarm calls uttered by tutors were more important than motor behaviour for the response of trained broods. However, the inability of adults to show complete motor behaviour and differences in the alarm calls elicited between tests and their controls suggest that alarm calls may be the key factor for the anti-predator responses of chicks. Further research is needed to address how audience affects alarm calls of birds used as tutors, as tutors and chicks were unfamiliar (Zaccaroni *et al.*, 2013). Among not trained broods, 25% of tests resulted in strong behavioural responses, which suggests that social learning from conspecifics may play an additional part in the development of anti-predator behaviour (Nicol, 2006), though skills acquired by learning from "chick to chick" may not be enough for a correct development of anti-predator behaviour, especially during the all-important early day of life. In conclusion, the combination of alarm calls and exemplary behaviour from tutors in combination with direct sightings of the predator stimuli may explain the differences in the responses between trained and not trained broods (Thaler, 1986).

For both types of broods, we found variation in the type of motor behaviour and its duration across phases. Together with the effects of tutors on trained broods, differences among phases may be explained by the change on risk perception and anti-predator ability as phases progressed (chicks may flutter-fly from the second phase). However, we cannot discard the effects of the nature of the stimuli and the circumstances in which anti-predator behaviour is performed. Buner (2007) observed that free-living grey partridges mainly crouched in response to raptors, whereas when facing humans reactions were adapted to the circumstance (e.g. walkers, traffic and farming). This suggests that some stimuli induce more predictable responses (i.e. raptors), while responses elicited by other stimuli are also affected by the circumstance (i.e. humans). Additionally, anti-predator responses observed in the control tests may have reduced the effects of the training, which suggests that the type of stimuli given should be careful chosen and, at least for this study, training involving humans should be reconsidered. As demonstrated in other species, anti-predator training should be an important part of captive management for both re-establishment and releasing purposes (Griffin *et al.*, 2000; Shier and Owings, 2006; Teixeira and Young, 2014), though other forms of training (such as food unpredictability, see Homberger *et al.*, 2014) should be further investigated in gamebirds. Previous studies have demonstrated some benefits of parental anti-predator teaching for gamebird chicks (Dowell, 1990; Anttila *et al.*, 1995) and the ability of red-legs to discriminate between different predator stimuli in captivity (Binazzi *et al.*, 2011), but to our knowledge training has not been incorporated in commercial rearing.

As the improvement of behavioural responses during training may indicate a better preparation for releasing but is not a predictor of post-release survival (Van Heezik *et al.*, 1999), a subsequent study was carried out comparing the survival of the two different types of broods (Gaudioso *et al.*, 2011). Trained broods had a 4.8-times higher post-release survival when compared to not trained ones, with the majority of the latter failing to survive more than 2 weeks after release. Out of 40 radio-tagged not trained chicks, 87% were predated within the first month after release.

We should be cautious with our results, as the sample size of our behavioural and radio-tracking study was rather low, and further research with larger samples should be conducted. However, the present research has shown that the behaviour of red-legs reared under commercial systems can be improved through cost-effective training. Hence, we encourage gamebird farmers, conservationists and wildlife managers to consider carefully planned training techniques, although measures should be taken to avoid detrimental effects such as habituation (WPA–IUCN, 2009).

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## DISCLOSURE STATEMENT

No potential conflict of interest was reported by the authors.

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