1 Conserved abilities of individual recognition and genetically

2 modulated social responses in young chicks (*Gallus gallus*)

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14 Abstract

The ability to recognise familiar individuals and the motivation to stay in contact with conspecifics are important to establish social relationships from the beginning of life. To understand the genetic basis of early social behaviour, we studied the different responses to familiar/unfamiliar individuals and social reinstatement in 4-day-old domestic chicks (*Gallus gallus*) in three genetically isolated breeds: Padovana, Polverara and Robusta. All breeds showed a similar ability to discriminate between familiar and unfamiliar

individuals, staying closer to familiar individuals. Social reinstatement motivation 21 22 measured as the average distance between subjects, latency to the first step and exploration of the arena (a proxy for the lack of fear), differed between breeds. More 23 socially motivated chicks that stayed in closer proximity, were also less fearful and 24 explored the environment more extensively. These results suggest that modulation of 25 social behaviour shows larger genetic variability than the ability to recognise social 26 partners, which appears to be an adaptive ability widespread at the species level even for 27 very young animals. 28

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30 Keywords

Individual recognition, imprinting, social reinstatement, social motivation, chicks, *Gallus gallus*, early cognition

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35 1. Introduction

In many species, the ability to recognise familiar individuals is important to establish 36 37 social relationships from the first moments of life. In domestic chicks (Gallus gallus) and other precocial animals that move around soon after birth, the mechanism of imprinting 38 enables to quickly learn the features of familiar individuals such as the mother and the 39 40 siblings (Bateson, 1966; Bolhuis, 1991; McCabe, 2013; Vallortigara & Versace, 2018). After imprinting, hatchlings exhibit strong affiliative responses towards the imprinting 41 stimulus while avoiding unfamiliar stimuli, a behaviour that requires the ability to 42 43 recognise familiar individuals. Moreover, separation from the imprinting objects elicits 44 attempts to signal and join the social partners (Jones & Merry, 1988; B. R. Jones & 45 Williams, 1992; Suarez & Gallup, 1983; Zajonc, Wilson, & Rajecki, 1975), as well as adrenocortical activation in domestic chicks (Jones & Williams, 1992). Imprinting is not 46 limited to general species recognition, as initially hypothesised by Lorenz, but enables to 47 discriminate between particular individuals. This is shown by the different affiliative and 48 aggressive responses to familiar and stranger social partners of the same chicken breed 49 (Rajecki, Ivins, & Rein, 1976; Väisänen & Jensen, 2004; Vallortigara, 1992; Vallortigara, 50 Cozzutti, Tommasi, & Rogers, 2001; Vallortigara & Andrew, 1994; Zajonc et al., 1975 see 51 also Schweitzer, Poindron, & Arnould, 2009 for quails). Moreover, Johnson and Horn 52 53 (1987) have shown that the ability of chicks to imprint on a specific hen depends on the 54 intermediate and medial mesopallium (IMM) and Town (2011) has shown that this area 55 responds differently to familiar and unfamiliar conspecifics.

For wild hatchlings, the ability to imprint and recognise familiar individuals is 56 important to identify the care giver and maintain flock/group cohesion. However, 57 recognition of familiar individuals has been documented also in hatchlings of solitary 58 59 species, such as land tortoises of different species (Versace, Damini, Caffini, & Stancher, 2018). Filial imprinting is particularly suitable to investigate in the laboratory since 60 imprinting can be established also for artificial objects that are more easily manipulated 61 62 in appearance than living animals. Thanks to these features, imprinting has become a model system for memory, learning and social behaviour (Bateson, 1966; Bolhuis, 1991; 63 Horn, 1985, 2004; McCabe, 2013, 2019). In the laboratory, imprinting is studied using 64 65 well controlled artificial stimuli such as balls, cubes, cylinders or two-dimensional stimuli 66 presented on cardboards or computer screens (Rosa-Salva et al., 2018; Versace, Schill, Nencini, & Vallortigara, 2016; Versace, Spierings, Caffini, ten Cate, & Vallortigara, 2017; 67 Wood & Wood, 2015). These experiments have shown that chicks discriminate subtle 68 differences of the imprinting objects, such as rotation of the features located inside the 69 imprinting object (Vallortigara & Andrew, 1991), the configuration of items that 70 compose the imprinting stimulus (Rosa-Salva et al., 2018) and even the underlying 71 structure of the stimuli independent of their physical appearance (Martinho & Kacelnik, 72 2016; Versace, Regolin, & Vallortigara, 2006; Versace, Spierings, et al., 2017). Less is 73 known on the genetic differences in the early ability of chicks to imprint on living 74 75 objects and to discriminate between familiar and unfamiliar individuals. Väisänen and Jensen (2004) have explored the differences in responses to familiar and unfamiliar social 76 stimuli in red jungle fowls (a breed considered to be close to the ancestral 77 undomesticated chicken (Miao et al., 2013) and White Leghorns (a modern breed 78

relected for laying eggs), using 3-4 week old animals. This study showed greater
affiliative responses in White Leghorn compared to red jungle fowls but did not clarify
the onset of these differences.

82 Here we explore the genetic differences of chicks in responding to familiar/unfamiliar individuals in 4-day old chicks of three different breeds of domestic 83 84 fowl: the Padovana, Polverara and Robusta breed. Chicks originated in the same farm within the conservation programme Co.Va, that kept these local breeds in genetic 85 isolation for more than 20 years (De Marchi, Cassandro, Targhetta, Baruchello, & 86 87 Notter, 2005). This particular arrangement reduced the environmental differences while 88 ensuring low admixture between breeds (Zanetti, De Marchi, Dalvit, & Cassandro, 89 2010). We previously investigated the predisposed visual preferences of these breeds to approach a stuffed hen vs. a scrambled version of it in visually naïve chicks (Versace, 90 91 Fracasso, Baldan, Dalle Zotte, & Vallortigara, 2017). Predisposed responses precede imprinting for they are exhibited before any prior visual experience has occurred and do 92 93 not depend on experience (Di Giorgio et al., 2017). When given the choice between a stuffed hen and a scrambled version of it, we observed that all three breeds initially 94 95 preferred to orient towards the stuffed hen, which is the predisposed stimulus that 96 several breeds preferentially approach (see Egorova & Anokhin, 2003; Johnson & Horn, 97 1988; Mayer, Rosa-Salva, Lorenzi, & Vallortigara, 2016; Versace, Fracasso, Baldan, Dalle 98 Zotte, & Vallortigara, 2017). Interestingly, we observed a difference between breeds as 99 early as the first 10 minutes of visual experience: while the Polverara breed showed a 100 steady attachment for the stuffed hen throughout the test, the Robusta and Padovana

101 breeds were attracted by the alternative stimulus. This suggests that behavioural 102 strategies that drive early attachment and orientation have a genetic basis, and that 103 genetic differences are apparent in the first minutes of visual experience. It is not clear, 104 though, whether differences in the ability to recognise familiar individuals and social 105 motivation are present between these breeds. The peculiar skull and brain anatomy of the Padovana breed is a further reason of interest for these breeds. Both the Padovana 106 107 and Polverara breed have a striking crest on the head but only in the Padovana breed the crest covers a perforated skull with an associated enlargement of the brain (Verdiglione 108 109 & Rizzi, 2018). The behavioural and cognitive implications of this striking anatomy 110 remain elusive, although Darwin (1868) hypothesised potential deficits in a closely 111 related breed, the white crested Polish. Historical documents suggest that the Padovana 112 breed, whose traces go back to Roman times (Brothwelp, 1979), was introduced in Italy 113 from Poland more than seven centuries ago (De Marchi et al., 2005), originating from 114 the white crested Polish. In the White crested Polish/Padovana breed the endocranium is enlarged and the brain has dramatically expanded to fill this gap (Frahm & 115 Rehkamper, 1998). It is not clear which behavioural consequences this peculiar brain 116 117 organisation has produced, and whether the abilities of individual recognition and affiliative responses for this breed differ from those of other chickens. 118

119 To investigate the genetic variability in early individual recognition and in social 120 motivation we tested the three breeds in an open field test. In this setting, chicks are 121 located in a novel empty arena larger than their home cage and their spontaneous 122 behaviour is observed in the presence of familiar or stranger conspecifics. The ability of

123 chicks to recognise familiar/stranger individuals can be inferred looking at whether the 124 distance kept between familiar animals is different. Due to the process of filial imprinting, familiar chicks are expected to stay closer than stranger chicks, as previously 125 126 documented (see for instance Vallortigara, 1992; Zajonc et al., 1975). Based on their 127 antipredatory and affiliative behaviour, it has been suggested that animals located in an open field with other conspecifics experience both the fear of being in an open 128 129 environment – which has the effect of reducing activity and exploration –, and social reinstatement, namely the motivation to reach the group and remain in contact with 130 conspecifics (Suarez & Gallup, 1983; Vallortigara, 1992; Vallortigara, Cailotto, & 131 132 Zanforlin, 1990). In a range of species, greater latency of movement/tonic immobility indicates antipredatory responses, while shorter distance between individuals indicates 133 stronger social/reinstatement motivation (Jones, Mills, & Faure, 1996; Versace, Caffini, 134 Werkhoven, & Bivort, 2019). 135

136

137 2. Methods

138 2.1 Breeds and conservation scheme

We studied three genetically isolated breeds of domestic fowl (*Gallus gallus*) raised in the same farm (Istituto Istruzione Superiore Agraria "Duca degli Abruzzi", Padova, Italy): Padovana, Polverara and Robusta maculata. These breeds entered the Co.Va conservation project more than 20 years before this project (De Marchi et al., 2005). The breeding and conservation scheme included no gene flow between breeds and were aimed at increasing the number of pure breed animals while maintaining genetic 145 variability within the breed. We included individuals from gold, silver and buff Padovana 146 and white and black Polverara, because previous studies showed high homogeneity 147 within these breeds (Zanetti, De Marchi, Abbadi, & Cassandro, 2011; Zanetti et al., 148 2010). The Robusta maculata breed was developed in 1965 from crosses between Tawny 149 Orpingtons and White Americans. Zanetti et al. (2010) documented genetic isolation (low level of admixture) between these breeds and a closer phylogenetic relationship 150 151 between Padovana and Polverara, which are also more similar at phenotypic level compared to Robusta. More details are provided in Versace et al. (2017). 152

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154 2.2 Subjects and rearing conditions

We tested 221 pairs of chicks: 94 pairs of the Padovana breed (PD), 88 pairs of the 155 156 Polverara breed (PL), 39 pairs of the Robusta breed (RB). During the test, 156 pairs moved: 58 Padovana, 60 Polverara and 38 Robusta. Eggs were obtained from the 157 Agricultural High School "Duca degli Abruzzi" (Padova, Italy), which is pursuing the 158 159 Co.Va conservation programme for the maintenance of local biodiversity described above (De Marchi et al., 2005). We incubated and hatched eggs in darkness at 37.7 °C. 160 161 During incubation, humidity was kept at 40% and then increased to 60% during the last three days of incubation. Chicks hatched in groups of the same breed and were then 162 163 housed in pairs of the same breed within 24 hours from hatching without any visual exposure to conspecifics prior to housing. Chicks were maintained in standard rearing 164 conditions (temperature 28°C, 14:10 day:night cycle) for three days in 28x38x32 cm 165

166 cages, and tested at an age of 4 days after hatching. After the experiments, animals were167 donated to local farmers.

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169 2.3 Apparatus

The test apparatus used for the test was a black square area (40x40x36 cm), illuminated with an incandescent lamp (100 W) located 1 m above the centre of the arena. A camera recorded the experimental scene from above during the test.

173

174 2.4 Test procedure

175 The experiment consisted of two phases: a familiarization phase and a test phase (see 176 Figure 1). The familiarization phase started soon after hatching (day post hatching 0) and 177 lasted 3 days. During this phase a pair of chicks of the same breed was housed in the 178 same cage. At test, each subject was visually isolated for 10-15 minutes in an opaque box 179 (14.5 cm height, 8.5 cm large, 11 cm width) located inside its cage and then transferred 180 to the testing room together with another experimental subject. During the test we 181 observed two chicks previously housed together (Familiar condition) or previously housed with a different animal (Stranger condition). At the beginning of the test, 182 experimental chicks were placed simultaneously in two opposite corners of the arena 183 and left free to move for 5 minutes. 184

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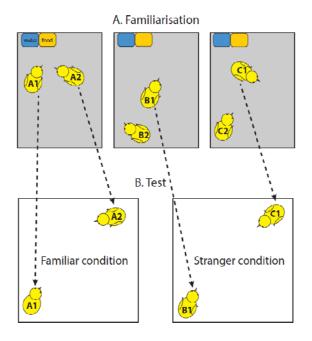


Figure 1. (A) Chicks were initially housed with another chick of the same breed for three days (familiarisation). (B) At test, chicks were located in a new arena either with the familiar companion (Familiar condition) or with a novel chick (Stranger condition). For each pair we recorded the latency of the first step, the distance between the centroid of the chicks and overall distance run.

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191 2.5 Data analysis

We initially established whether chicks in the pair moved or remained still throughout the test and counted how many pairs moved/did not move during the test. We used a Chi-square test to check whether more pairs moved in the Stranger or in the Familiar condition.

For all pairs that moved, we used an Anova to analyse the average distance (cm) between the centroid of subjects throughout the test, the overall distance moved by the pair (cm) and the average latency of the first step (s) using Condition (familiar, stranger), 199 Breed (Padovana, Polverara, Robusta) and their interaction as independent variables. 200 Exploratory analyses showed that a log-transformation of the distance moved and 201 latency normalised the residuals of these variables, hence we used log-transformed values for these analyses. We ran post-hoc t-tests to explore significant differences. We 202 203 also analysed the relation between latency, distance run and distance between subjects using the Spearman rank order correlation test and fitting linear and polynomial models 204 205 until finding the minimal adequate model. Significance level was set to p < 0.05. Statistical analyses were performed with the R software (version 3.5.2). 206

207

208 3 Results

209 3.1 Pairs that moved in the Familiar and Stranger condition

There was no significant difference in the frequency of pairs that moved/did not move
between the Familiar and Stranger condition (X-squared=0.834, df=1, p=0.361) or
Breed (X-squared=0.989, df=2, p=0.610).

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214 **3.2 Distance between individuals**

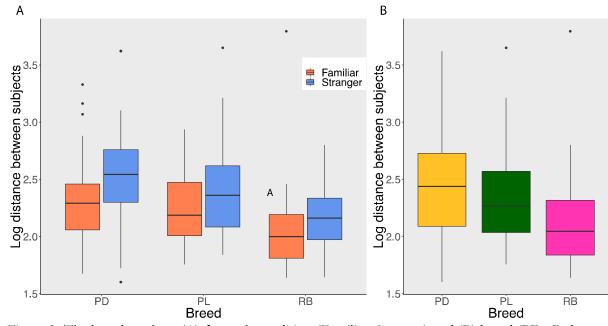
Looking at the log-transformed distance between subjects we found a significant effect of Condition ($F_{1,150}$ =8.311, p=0.005) and Breed ($F_{2,150}$ =5.664, p=0.004) and no significant interaction ($F_{2,150}$ =0.155, p=0.857). These results are shown in Figure 2. Familiar chicks stayed closer than stranger chicks (t=-2.817, df=153.34, p=0.005). The

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average distance between Familiar chicks was 17.57 cm (median 10.26 cm), the average

distance between Stranger chicks 20.75 cm (median 15.35 cm).

Chicks in the RB breed stayed significantly closer than PL (t=2.536, df=76.942, p=0.013) and PD (t=3.455, df=80.728, p<0.001) chicks, whereas there was no significant difference between PL and PD chicks (t=1.139, df=115.01, p=0.257).



224BreedBreed225Figure 2. The boxplots show (A) for each condition (Familiar, Stranger) and (B) breed (PD=Padovana,226PL=Polverara, RB=Robusta) the median, quartiles, maximum, minimum and outliers of the distance227between subjects expressed in log centimetres.

228

229 3.3 Distance run

Analysing the overall distance run in each pair, we found a significant effect of Breed ($F_{2,150}=6.102$, p=0.003) and no significant effect of Condition ($F_{1,150}=0.409$, p=0.523) and no significant interaction ($F_{2,150}=1.027$, p=0.360). These results are shown in Figure 3.

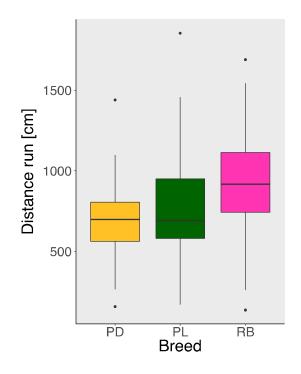


Figure 3. The boxplots show for each breed (PD=Padovana, PL=Polverara, RB=Robusta) the median,
quartiles, maximum, minimum and outliers of the distance run expressed in centimetres.

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Polverara chicks did not run significantly more than Padovana chicks (t=1.890, df=110.13, p=0.061) although there was a trend in this direction, Robusta chicks ran significantly more than Polverara chicks (t=2.530, df=71.406, p=0.014), and significantly more than Padovana chicks (t=4.186, df=58.757, p<0.001).

242

243 3.4 Latency first step

Analysing the latency of the first step in each pair, we observed a significant effect of Condition ($F_{2,150}$ =4.026, p=0.047, with chicks in the Familiar condition moving sooner than chicks in the Stranger condition) and Breed ($F_{2,150}$ =11.02, p<0.001) and no

significant interaction ($F_{2,150}=0.336$, p=0.715). The mean latency for the Familiar condition was 43.29 s (median 20.25 s) and for the Stranger condition 48.19 s (median 36 s). The results are shown in Figure 4.

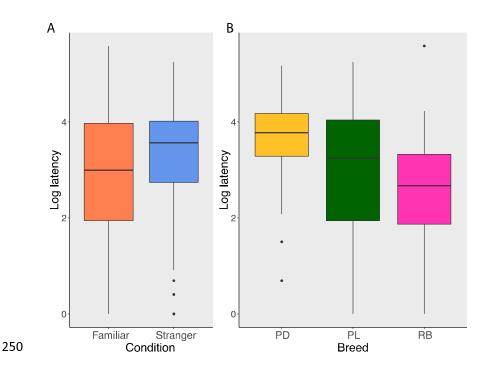


Figure 4. The boxplots show for each (A) condition (Familiar, Stranger) and (B) breed (PD=Padovana,
PL=Polverara, RB=Robusta) the median, quartiles, maximum, minimum and outliers of the pair
latency of the first step expressed in log seconds.

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255 Polverara chicks moved significantly earlier than Padovana chicks (t=-27.937, df=78.274,

256 p < 0.001), Robusta chicks moved significantly earlier than Polverara chicks (t=-27.229,

df=71.209, p<0.001), and also Robusta chicks moved significantly earlier than Padovana

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261 3.5 Relation between latency, distance between subjects and distance run

We observed a significant positive correlation between the latency of the first step and the average distance between subjects (S=162850, p<0.001, rho=0.743, see Figure 5A) and a significant negative correlation between the latency of the first step and the distance run (S=899280, p<0.001, rho=-0.421, see Figure 5B).

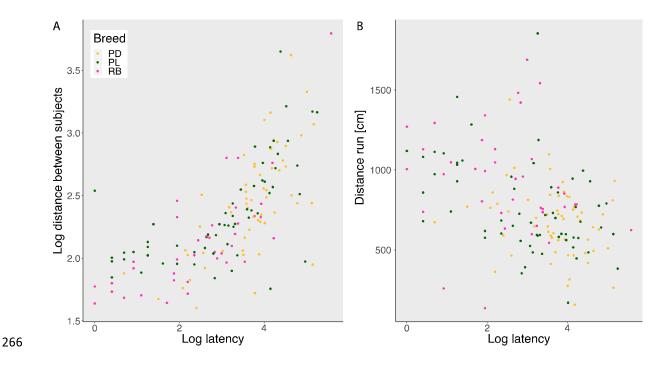


Figure 5. The scatterplots show the relation between (A) latency of the first step and average distancebetween subjects and (B) latency of the first step and distance run.

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The minimal adequate model for the dependent variable Distance between subjects using as predictors Latency, Condition and Breed included a quadratic polynomial with Latency ($F_{2,150}$ =98.035, p<0.01), Condition ($F_{1,150}$ =7.13, p=0.008) and Latency x Condition ($F_{2,150}$ =4.77, p=0.001) as significant effects. The full table of coefficients is reported in Table 1.

	Estimate	Std. Error	T value	$\Pr(\geq t)$
Intercept	2.266	0.032	70.122	<2e-16
Latency (linear)	3.138	0.370	8.493	1.83e-14
Latency (quadratic)	0.886	0.366	2.421	0.017
Condition (Stranger)	0.120	0.045	2.642	0.009
Latency (linear) x Condition	1.135	0.576	1.970	0.051
Latency (quadratic) x Condition	1.373	0.572	2.400	0.018
Residual standard error: 0.2797 of	n 150 degre	ees of freedor	n	
Multiple R-squared: 0.587	Adjusted R-sq	uared: 0.573		
F-statistic: 42.55 on 5 and 150 df,	p<2.2e-16			

²⁷⁵ Table 1. This table shows the coefficients of the minimal adequate model for the dependent variable

277

278 The minimal adequate model for the dependent variable Distance run using as

predictors Latency, Condition and Breed included Latency ($F_{1,151}$ =29.100, p<0.01) and

280 Breed ($F_{2,151}$ =4.322, p=0.015) as significant effects. The full table of coefficients in

281 reported in Table 2.

	Estimate	Std.	T value	$\Pr(> t)$			
		Error					
Intercept	715.233	42.460	16.845	<2e-16			
Latency (linear)	-1156.56	286.562	-4.036	< 0.001			
Condition (Stranger)	-3.876	42.880	0.090	0.928			
Breed (PL)	40.333	50.206	0.803	0.423			
Breed (RB)	168.909	59.122	2.857	0.005			
Residual standard error: 26	4.6 on 151 o	degrees of fr	eedom				
Multiple R-squared: 0.2		Adjusted R-squared: 0.179					
F-statistic: 9.436 on 4 and 1	151 df, p<0.	001	-				

²⁸² Table 2. This table shows the coefficients of the minimal adequate model for the dependent variable

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²⁷⁶ Distance between subjects using as predictors Latency, Condition and Breed.

²⁸³ Distance run using as predictors Latency, Condition and Breed.

286 4. Discussion

From the beginning of life, the ability to recognise familiar individuals and the 287 288 motivation to stay in contact with conspecifics are important to establish social 289 relationships. Little is known, though, on whether early social behaviours have a genetic basis that determines behavioural differences. To address this issue, we focused on 290 291 domestic chicks (Gallus gallus). Chicks are an ideal model to investigate the first social responses not only because they are a precocial species (Versace & Vallortigara, 2015; 292 293 Versace, 2017) and move around autonomously soon after birth but also because they 294 live in flocks and have to recognise the mother and siblings to maintain social cohesion 295 (Nicol, 2015). We previously showed that the initial predisposition to orient towards a 296 (stuffed) hen does not vary between breeds, while exploratory responses to social stimuli 297 differ between breeds after just 10 minutes of experience (Versace, Fracasso, Baldan, 298 Dalle Zotte, & Vallortigara, 2017). Here, we investigated early social responses mediated by previous social experience and learning. We tested whether 4-day-old chicks have 299 standing genetic variation for the ability to recognise conspecifics and social 300 reinstatement by using three genetically isolated breeds: Padovana, Polverara and 301 Robusta. 302

In all breeds, we observed a similar ability to discriminate between particular individuals and recognise a familiar one. As a proxy for recognition, we used the different distance kept between familiar and unfamiliar individuals. When located in a new environment with a familiar or an unfamiliar chick, all tested breeds equally discriminated between familiar and unfamiliar individuals staying significantly closer to

308 familiar chicks. and showing shorter latency of movement with pairs of familiar 309 individuals. Even the Padovana breed, that shows the neuroanatomical peculiarity of a 310 perforated skull with an associated enlargement of the brain (Verdiglione & Rizzi, 2018), 311 did not show cognitive differences in this ability. This performance of Padovana chicks suggests that the behavioural limitations noticed by Darwin (1868) in the white crested 312 Polish – a breed ancestral to Padovana (De Marchi, Dalvit, Targhetta, & Cassandro, 313 2005; Verdiglione & Rizzi, 2018) - do not derive from deficits in social motivation or 314 315 individual recognition (they might instead be due to difficulties in visual perception 316 linked to the long plumage of the crest as suggested by Vallortigara, personal 317 communication). Hence, the ability to promptly recognise familiar individuals appeared widespread at the species level in chickens. Previous studies conducted on quails 318 (Coturnix coturnix japonica) (Kovach, 1990) showed a slow response to selection for 319 low/high imprintability. In this study, imprintability was selected as low/high ability to 320 exhibit imprinting responses for colours that were not initially preferred by young birds. 321 The observed sluggish response to selection for imprintability observed in quails is in 322 line with the absence of interbreed differences in the recognition of familiar individuals 323 that we have documented in chicks. 324

We have previously shown that even hatchlings of non-social species such as tortoises are able to recognise familiar and unfamiliar individuals in the first days of life (Versace, Damini, Caffini, & Stancher, 2018). The early age and little experience required (see also Suarez & Gallup, 1983; Vallortigara, 1992; Zajonc et al., 1975), together with the low genetic variability of this trait, shows how crucial individual recognition is at the

330 onset of life for a range of different species. This contrasts with other social traits that 331 showed significant genetic variability. In fact, we observed very clear differences between breeds and conditions when looking at measures of social reinstatement and fear in an 332 333 open field. We measured social reinstatement, the motivation to join the social group 334 and keep in contact with it, looking at the average distance between subjects. Shorter distance indicates greater social reinstatement motivation (Jones et al., 1996; Schweitzer 335 336 et al., 2009; Suarez & Gallup, 1983; Vallortigara et al., 1990; Vallortigara, 1992). Both the Polverara and Padovana breed, that are closely genetically related (Zanetti et al., 2010), 337 338 maintained a larger distance than the Robusta breed, providing another indication of the 339 fact that the observed differences have a genetic basis. We also assessed the fear elicited by being in a novel open field by looking at the latency to the first step and overall 340 distance run. Since an adaptive antipredator response for chicks in an open field is 341 342 freezing/tonic immobility, fear is expected to induce greater latency, and less exploration 343 in the arena as antipredator behaviours. We observed that chicks with lower latency 344 stayed closer and explored the arena more extensively. While fear/antipredatory responses and social reinstatement can dissociate (for instance with short distance 345 between subjects that indicates high social reinstatement and little distance moved that 346 indicates high fear), we observed that they were inversely correlated. This shows that 347 348 pairs of chicks that approached each other explored the arena while remaining in close 349 proximity.

350 Differences in social reinstatement and fear responses were apparent from the351 first days of life, in animals with very limited social experience. The differences in social

352 reinstatement and fear had a clear genetic basis, with an effect of breed on latency, 353 distance run and distance between individuals. In particular, the Robusta breed showed 354 the shortest latency and distance between subjects as well as greater distance run during 355 the test. These results are in line with the fact that quails respond to selection for high/low social reinstatement and fear (Formanek et al., 2008; Mills & Faure, 1991; 356 Mills, Jones, & Michel, 1995), that older chicks of jungle fowl and White Leghorn exhibit 357 358 different social reinstatement behaviour (Väisänen & Jensen, 2004), and that neonate chicks differ in exploratory responses to social stimuli (Versace, Fracasso, Baldan, Dalle 359 360 Zotte, & Vallortigara, 2017).

361 Our findings suggest that the modulation of social behaviour shows larger genetic 362 variability than the ability of recognizing familiar individuals. This points at the crucial 363 adaptive role of individual recognition, an ability widespread across taxa and available 364 from the earliest developmental stages also in tortoises (Versace, Damini, Caffini, & 365 Stancher, 2018). Interestingly, while we have shown that chicks consistently tend to 366 aggregate with other individuals, tortoise hatchlings – that are solitary until reaching sexual maturity - ignore familiar individuals and avoid strangers (Versace, Damini, 367 Caffini, & Stancher, 2018). The ability to promptly recognize familiar individuals can 368 hence sustain both affiliative and avoidance responses. The pivotal role of individual 369 370 recognition is apparent in invertebrates as well. For instance, in studying aggression, 371 Yurkovic et al. (2006) have documented the ability of fruit flies to respond differently to 372 familiar and unfamiliar opponents. Not only familiar opponents had significantly fewer encounters but the fighting strategies depended on whether the opponent was familiar 373

374 or unfamiliar: losers tested with unfamiliar winners were more aggressive than losers 375 paired with familiar winners. Individual recognition has been documented in other insects such as paper wasps (Tibbetts, 2002; Tibbetts & Dale, 2007) and ants (D'Ettorre 376 377 & Heinze, 2005). It hence appears that individual recognition has a crucial role in a 378 broad comparative context. Using newly hatched chicks of different breeds, we have shown the ability to recognize familiar individuals and differentially respond to them is 379 widespread in this species from the earliest stages of life. We suggest that identifying the 380 381 core abilities exhibited by young animals at the beginning of life might also guide experts 382 in artificial intelligence in understanding which are the fundamental components of 383 general intelligence (Versace, Martinho-Truswell, Kacelnik, & Vallortigara, 2018). Further studies should clarify the role of different perceptive cues chicks in 384 385 discriminating between familiar individuals.

386

387 Animal welfare note

All experiments comply with the ASAB/ABS Guidelines for the Use of Animals in Research and with the current Italian and European Community laws for the ethical treatment of animals and the experimental procedures were approved by the Ethical Committee of University of Trento and licensed by the Italian Health Ministry (permit number 1138/2015 PR). At the end of the experiments, all chicks were donated to local farmers.

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