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***Title:* Hemispheric specialization in spatial vs. ordinal**

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processing in the day-old domestic chick (*Gallus gallus*)

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***Running Head:* When number conflicts space.**

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9 **Authors:** Rosa Rugani^{1,2*}, Lucia Regolin¹

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12 **Affiliations:**

13 ¹ Department of General Psychology, University of Padova, Padova, Italy.

14 ² Department of Psychology, University of Pennsylvania, Philadelphia,

15 PA, United States.

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17

18 *Correspondence to: E-mail: rosa.rugani@unipd.it, rugani@sas.upenn.edu

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1 **ABSTRACT**

2 Different species show an intriguing similarity in representing numerosness in
3 space starting from left to right. This bias has been attributed to a right
4 hemisphere dominance in processing spatial information. Here, to disentangle
5 the role of each hemisphere in dealing with spatial vs. ordinal-numerical
6 information, we tested domestic chicks during monocular vs. binocular vision. In
7 the avian brain, the contralateral hemisphere mainly elaborates the visual input
8 to each eye. Four-day-old chicks learnt to peck at the 4th element in a sagittal
9 series of 10 identical elements. At test, chicks faced a left-to-right-oriented
10 series, where inter-element distance was manipulated, so that the 3rd element
11 was where the 4th had been at training; this compelled chicks to use either
12 spatial or ordinal cue. Chicks tested binocularly selected both the 4th left and (to
13 a lesser extent) right elements. Chicks tested monocularly chose equally the 3rd
14 and the 4th elements on the seeing side. Interhemispheric cooperation resulted
15 in the use of ordinal-numerical information; whilst, each single hemisphere
16 could rely on spatial or ordinal-numerical cue. Both hemispheres can process
17 spatial and ordinal-numerical information, but their interaction results in the
18 supremacy of processing ordinal-numerical cue.

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21 **KEYWORDS:** Number cognition; Asymmetry; Hemispheres; Lateralization;
22 Mental Number Line; Spatial-Numerical Association, Domestic chick.

1 INTRODUCTION

2 A peculiar characteristic of numbers is their spatial representation along a left-
3 right oriented continuum¹. Healthy participants are faster at processing small
4 numbers when responses are executed in the left side of space, and large
5 numbers when responses are executed in the right side of space (spatial-
6 numerical association of response codes, SNARC effect²). For a long time, the
7 orientation of the spatial numerical association (SNA) has been attributed to
8 cultural factors, such as reading and writing habits³. Israeli participants (who
9 read words from right to left and numbers from left to right) showed no SNA³,
10 while Palestinians, who read both words and numbers from right to left, show a
11 reversed SNA⁴. A growing number of studies in pre-verbal children⁵⁻⁹ and non-
12 human species¹⁰⁻¹⁷ support the hypothesis that SNA may be present soon after
13 birth and exist before experience or cultural influence, but see^{18,19}. Different
14 species (domestic chicks¹⁰⁻¹², adult Clark's nutcrackers¹¹, and adult Rhesus
15 macaques²⁰) showed an intriguing similarity in representing growing ordinal-
16 numerical information as oriented from left to right. Animals were trained to
17 select a target element (e.g. the 4th one), in a sagittally-oriented series of
18 identical and fixed elements. They were then tested with a series, identical to
19 the first one but frontal, i.e., rotated by 90°; hereafter we refer to this kind of task
20 as spatial-and-ordinal task. At test, even if the correct alternatives were two –
21 the left and the right target- animals chose the left one. This suggests that a
22 disposition to associate the numerical magnitudes from left to right may
23 originate from a neuro-biological precursor^{21,22}. Interestingly enough, if the use
24 of the spatial cues -the distance of the target element from the beginning or

1 from the end of the series- is prevented, chicks choose both the left and the
2 right target. When the distance between the elements was changed on each
3 trial, both during training and during fronto-parallel testing, chicks correctly
4 identified the target (the 4th element), but they did not show any left bias. The
5 left bias seems to be related to the possibility of using spatial and ordinal
6 information both during learning and recall. Bilateral responses occur also when
7 chicks learnt to identify the target on spatial and ordinal cues (spatial-and
8 ordinal training) but at test they could either use the spatial or the ordinal one
9 (spatial vs. ordinal test). Taken together this evidence indicates that the left-to-
10 right bias in this test occurs solely when both spatial and ordinal information
11 were available during learning as well as recall ¹². An intra-hemispheric coupling
12 of ordinal cues, which is bilaterally represented, and of spatial cues, unilaterally
13 (right) represented, has been proposed to explain differential attentional
14 allocation toward the left and right visual hemifields.

15 Nevertheless, the neural correlates of this left-to-right oriented numerical
16 association remain largely unknown. In humans, the posterior parietal cortex
17 has been indicated as the brain region dedicated to respond to numbers^{23–25}.
18 Moreover, in humans, populations of neurons tuned to small numbers have
19 been described in the parietal cortex²⁶. These neurons are organized
20 topographically, forming a map in the brain. Such a neural organization might
21 determine the organization of magnitudes along the MNL²². Neurons tuned on
22 numerosness are located in the intra-parietal cortices of the macaque brain²⁷,
23 and in a brain association area (*nidopallium caudolaterale*, NCL) of the avian
24 brain²⁸. Up to now the topographical organization of “number-neurons” in

1 animals has not been defined. Since non-verbal numerical cognition is shared
2 by many animal species²², it is plausible that a similar map of number-neurons
3 would be a common characteristic of the architecture of animal neural systems.
4 In performing the original version of the “spatial-and-ordinal” task, specifically
5 designed to investigate left vs. right processing of spatial and ordinal
6 information, both hemispheres of the avian brain store, retain and proficiently
7 retrieve both numerical as well as spatial information¹⁶. Spatial asymmetries in
8 the “spatial-and-ordinal” task may be accounted for by a model that assumes
9 differential encoding, processing and integration by the two hemispheres for
10 spatial and numerical information.

11 A first attempt to study the hemispheric correlates of left-right bias in a serial
12 ordinal task, has been done using the technique of temporary monocular
13 occlusion. Due to the conformation of the avian brain, visual input to each eye is
14 mainly elaborated by the contralateral hemisphere. Two main features of the
15 bird brain make this possible: i) the visual fibres decussate nearly completely at
16 the optic chiasm^{29,30}; ii) the absence of a structure homologous to the corpus
17 callosum (even though smaller tracts exist which allow inter hemispheric
18 communication^{31–34}). Thus, each eye mainly projects toward the visual system
19 of the contralateral hemisphere³⁵. By temporarily restricting the visual input to a
20 single eye (by simply patching of the other eye), it is possible to determine the
21 role of the contralateral hemisphere³⁵. Chicks learnt to find food reinforcement
22 into the 4th element, in a sagittally-oriented series of identical and fixed
23 elements. At a subsequent fronto-parallel test, the series was maintained
24 identical but it was rotated by 90°. The test was conducted in three different

conditions of vision: binocular, right monocular and left monocular. At fronto-parallel test, right monocular chicks pecked at the 4th right position, left monocular and binocular chicks pecked at the 4th left position¹⁶. This indicates that both hemispheres process, maintain and correctly use ordinal-numerical information. The similarity in the left-oriented responses showed by the left monocular and binocular chicks suggests that the left bias is linked to a right hemisphere dominance, which allocates attention toward the left hemispace³⁶. But did the hemispheres use different strategies to solve the task? To approach this question, in the present study we used a spatial-and-ordinal training, which allows learning of both spatial and ordinal information, and a “spatial vs. ordinal” test, in which we created a conflict between spatial and ordinal cues¹². We trained chicks to identify the 4th element in a series of 10 identical elements. During training the elements were in fixed positions, thus birds could identify the 4th both on the basis of (i) its ordinal information: the 4th position in the sequence; (ii) its spatial information: the element that is located at a given distance from the beginning of the series. At test inter-elements distances were increased so that the 3rd element was at the distance from the beginning of the series in which the 4th element had been experienced during training. By integrating this paradigm with the use of temporary monocular occlusion, we studied how the two hemispheres encode, process and integrate spatial vs. ordinal information. We expected that chicks would be able to solve the task, and select the 4th element, but that the leftward bias would be lost if based on spatial cues.

MATERIAL AND METHODS

Subjects

Subjects were 13 male domestic chicks (*Gallus gallus*). We weekly obtained hours-old chicks from a local commercial hatchery (Agricola Berica, Montegalda, Vicenza, Italy). Immediately after their arrival, they were caged, in groups of three, in standard metal cages (28.0 x 40.0 x 32.0 cm width, depth and height, respectively) at controlled temperature (28–31 °C) and humidity (68%). Food (chick starter) and water were available *ad libitum*. Three times a day we also fed them some mealworms (*Tenebrio molitor larvae*), to familiarize them with this food, that was used as reinforcement during training.

Chicks were reared in these conditions from Monday morning (11.00 am) to Wednesday afternoon (5.00 pm), when they were singly caged. On Thursday (10.00 am) the food jars were removed, while water was left available. A few hours later (1.00 pm) birds underwent pre-training. Pre-training began when they were 4 days old, due to their yolk sac reserves, chicks are little motivated to peck for food reward before day 4 post-hatching. Two hours after the pre-training was over, each chick underwent training. Once this was over, they were singly caged overnight with food and water available *ad libitum*.

On Friday, in the early morning (7.00 am) chicks were food deprived and then re-trained (8.00 am). Testing took place for each chick immediately after the re-training. At the end of the behavioural observations, all chicks were caged in social groups of five birds, with food and water available *ad libitum*, and a few hours later they were all (i.e. those used in this as well as in all other

experiments described) donated to local farmers. All procedures employed were evaluated and approved by the Committee for Animal Welfare of the University of Padova.

Apparatus

All the experimental phases (pre-training, training, re-training, sagittal test and fronto-parallel tests) took place in an experimental room located near to the rearing room. During experimental phases temperature and humidity were kept respectively at 25° C and 70%. The room was lit by four 58 W lamps, placed on the ceiling, 148 cm above the experimental apparatus. The apparatus consisted in a plastic square-shaped arena (80 x 80 x 40 cm), which floor was covered with wood-shaving. The apparatus was connected with a starting box (7 x 11 x 11.5 cm) located outside of the arena itself. A slit (7 x 11 cm) connected the arena with the starting box. The slit was normally closed by an opaque plastic partition (8 x 12 cm); the partition was lifted for a few seconds at the beginning of each trial to let the chicks enter the arena. Ten identical elements (plastic bottle tops, 3.2 cm in diameter and 0.8 cm height) were aligned along the midline of the arena's floor. The elements were spaced 2.5 cm from one another, for an overall length of 54.5 cm. The sagittal series was symmetrically placed in the centre of the apparatus, thus the beginning of the series was 13 cm apart from the slit and 39.6 cm from the side walls (**Fig.1a**). Each element was filled with wood-shaving. All the elements contained a mealworm, but only in the 4th element it was reachable by the chicks. At the beginning of each trial,

all elements looked identical. In each experimental phases, the apparatus was randomly rotated in the experimental room, from trial to trial, to avoid any possible use of external cues.

Pre-training

On Day 4, chicks singly underwent a pre-training. The bird was firstly placed, for about 3 seconds, in the starting box and then, by sliding the partition, it was allowed to enter the arena. It could then spend a couple of minutes in the arena, walking wherever it wanted to be acquainted with the novel environment. As soon as the bird stopped to emit distress calls, a pre-training procedure started. Initially a mealworm was visibly positioned (not covered by wood-shaving) into the niche of the 4th element. Thereafter, the mealworm was progressively buried in the wood shaving, up to be completely hidden into it. Only chicks' pecking responses to the target element were reinforced. Once the mealworm was completely hidden, the learning criterion of this experimental phases was three consecutive correct trials.

Training

Training begun two hours after the pre-training was over. During training, all elements were filled with wood-shaving, so that at the beginning of each trial, they looked identical. Only the 4th element hid a piece of mealworm. At the beginning of each trials, the chick was placed in the starting box for a few seconds. The removable partition was lift from above thus the chick could enter the arena and walk towards the series of elements and peck at one of them. A

trial was considered over when the chick pecked at one element. Only one peck on each trial was allowed. A trial was considered correct if the chick pecked at the 4th elements. A trial was considered null and thus terminated after 180 seconds in the absence of a peck at whichever element. The learning criterion for this experimental phase was -at least- eight correct responses across 20 valid trials^{15,24}. All the chicks reached it and advanced to the subsequent experimental phase.

Re-training

Before the beginning of each of the following tests, chicks underwent a re-training. The experimental procedure was identical to that used in the training phase. The re-training criterion was fixed at three consecutive correct responses. All chicks passed this phase in 5–10 minutes. A few minutes after the completion of the re-training, chicks underwent a test.

Sagittal test

Two hours after training each chick underwent a re-training. Immediately thereafter, chicks underwent the sagittal test. This consisted of 20 consecutive trials. The experimental apparatus and the series was exactly identical to those used during training. During each trial, the chick was allowed one peck. Only correct responses could be reinforced: the food reinforcement was available only in some pre-established trials (trial number 4, 5, 7, 10, 13, 14, 16 and 19), and chicks could gain the food only by correctly choosing in those trials^{16,37,38} this reward schedule was applied also for the fronto-parallel test. This rewarding

1 schedule is needed to prevent extinction of responses over multiple unrewarded
2 test trials^{12,16}. All other trials were unrewarded. If no response occurred within
3 60 seconds, the trial was terminated. At the end of each trial, the chick was
4 gently placed back at the starting box and after approximately 5 seconds it was
5 given a new trial.

6 7 **Fronto-parallel tests**

8 In the morning of the fifth day, each chick underwent a re-training and then at a
9 fronto-parallel test. During fronto-parallel tests, we used a series of 10 elements
10 (**Fig.1b**); the elements were fronto-parallel oriented with respect to the slit (that
11 allowed the chick to enter the arena) and approximately 35.0 cm away from it.
12 Thus, the new test series was rotated by 90° when compared with the training
13 series. The distance between elements was enlarged (i.e. 5.35 cm), with
14 respect to the inter-element distance which was used during training, but it was
15 kept constant throughout the 20 testing trials. We specifically selected this
16 distance, to create a conflict between the spatial and the ordinal information: the
17 3rd element of this new series was located at the same absolute distance, from
18 the end of the series, in which was the 4th element in the previous phases (20.3
19 cm from the beginning of the series). The overall length of the series was 73.15
20 cm and the ends of the series were at 3.4 cm from the arena's lateral walls.

21 The fronto-parallel test was conducted in three different conditions of vision:
22 binocular, left monocular and right monocular. The interval between two
23 consecutive tests was of two hours, and before each test chicks underwent re-
24 training. All chicks firstly underwent the binocular test, then they underwent the

two monocular tests, in balanced order. Even if the order of the monocular tests was shown not to have an effect chicks' performance in a "space and number" ordinal task¹⁶, a group of chicks (n= 6) underwent the left-monocular fronto-parallel test as second test, another group (n=7) underwent the right-monocular fronto-parallel test as second test.

Twenty minutes before the beginning of a monocular test, a temporary eye patch -made of special, removable, paper tape- was gently applied so as to occlude vision from one eye (without preventing normal blinking). This period allowed subjects to get acquainted to the new condition of vision before the testing time. Before the binocular test, chicks received an equal amount of handling and acquainting time, but in this case the eye-patch was not applied).

RESULTS

For each test, we scored the pecks emitted by each chick to any of the 10 elements and we computed the percentages of correct responses [(number of pecks to a given element/20) x 100] and averaged separately. We planned to use parametric paired t-tests to compare: i) the pecks emitted at the target elements (4th from left and 4th from right), ii) the pecks emitted in the correct ordinal position (to the 4th element), iii) the pecks emitted in the correct spatial location (to the 3rd element). Thus for this last analysis, the planned comparisons were: i) the 4th left element vs. the 3rd left element, ii) the 4th right element vs. the 3rd right element.

For each test, before each pairwise analysis we run a test to check of normality (Shapiro-Wilk). Since the Shapiro-Wilk test indicated that data were not

normally distributed, we used the non-parametric equivalent: the One-sample Wilcoxon Test to analyze departures from chance level (10%) and the Wilcoxon's signed-rank test to analyze differences among the 3rd and the 4th elements. We reported the effect size as the rank-biserial correlation (r_B). We conducted all analyses using Jasp 0.11.1. Sample size was calculated using the formula for quantitative variables: $n = (2\sigma^2) / (\mu_1 - \mu_2)^2 \times f(\alpha, \beta)$; with the following values: $\alpha = 0.05$; $\beta = 0.80$; average = 30%; standard deviation = 18%. In this formula: σ is the variance; μ_1 and μ_2 are the means of the two groups; $f(\alpha, \beta)$ is a function of type I error (α) and type of II error (β). The sample size was evaluated according to the principle of Reduction in animal research and approved by the University Committee for animal welfare.

Sagittal test:

Chicks pecked at the 4th element above chance ($N = 13$, Mean = 53.846, ES = 2.542; Median = 55.000; $V = 91.000$, $p = 0.002$, $r_B = 1.000$); also the 3rd element: was pecked above chance ($N = 13$, Mean = 19.231, ES = 3.041, Median = 20.000; $V = 59.500$, $p = 0.020$, $r_B = 0.526$) (see **Fig. 2a**). However, chicks chose more often the 4th rather than the 3rd element ($W = 91.000$, $p = 0.002$, $r_B = 1.000$).

Since the first rewarded trial in all tests was the 4th trial, we restricted the analysis to the first four trials, to exclude any effect of learning during testing. It appeared that from the very first trials chicks selected the 4th element above chance ($N = 13$, Mean = 63.462, ES = 3.598; Median = 75.000; $V = 91.000$, $p = 0.001$, $r_B = 1.000$); the 3rd element was not selected above chance ($N = 13$, Mean = 15.385, ES = 5.325, Median = 0.000; $V = 63.000$, $p = 0.020$, $r_B = 0.500$) (see

Fig. 3a). Chicks chose more often the 4th rather than the 3rd element (W=66.000, p=0.003, r_B =-1.000).

Fronto-parallel tests:

Binocular test:

Chicks selectively chose both elements in the 4th position above chance (4th left element: Mean=41.538, ES=3.978, Median=45.000; V=78.000, p=0.001, r_B =0.714; 4th right element: Mean=16.538, ES=3.222, Median=15.000; V=62.000, p=0.037, r_B =0.590) (see **Fig. 2b**). They did not peck on the elements in the 3rd position above chance (3rd left element: Mean=5.769, ES=2.029, Median=5.000; V=15.000, p=0.957, r_B =0.071; 3rd right element: Mean=3.462, ES=1.042, Median=5.000; V=0.000, p=0.999, r_B =-1.000).

The chicks pecked more often at the 4th left than the 4th right element (W=72.500, p=0.010, r_B =0.859). They selected more often the 4th elements than the 3rd element both on the left and on the right side (left: W=91.000, p=0.002, r_B =1.000; right: W=55.000, p=0.006, r_B =1.000).

For what concerns the first four trials, chicks selectively chose the 4th elements above chance (4th left element: Mean=44.231, ES=6.426, Median=50.000; V=90.000, p<0.001, r_B =1.308; 4th right element: Mean=21.154, ES=6.231, Median=25.000; V=70.000, p=0.044, r_B =4.000) (see **Fig. 3b**). No difference was there between choice of the 4th left and the 4th right element (W=54.000, p=0.064, r_B =0.636).

Left monocular test: Chicks selectively chose the 4th left element above chance (4th left element: Mean=31.923, ES=4.441, Median=30.000; V=76.500, p=0.002, $r_B=0.681$), but not the 4th right element (Mean=5.385, ES=1.647, Median=5.000; V=5.000, p=0.992, $r_B=-0.643$) (see **Fig. 2c**). Also the 3rd left element was pecked above chance (Mean=22.308, ES=2.164, Median=25.000; V=76.500, p=0.002, $r_B=0.681$), but not the 3rd right element (Mean=2.692, ES=1.342, Median=2.000; V=2.000, p=0.0999, $r_B=-0.600$).

Chicks pecked more often at the 4th left element than at the 4th right element (W=78.000, p=0.002, $r_B=1.000$). They pecked at the 4th just as much as the 3rd elements, both on the left and on the right sides (left: W=52.500, p=0.089, $r_B=0.591$; right: W=14.000, p=0.090, $r_B=0.867$).

For what concerns the first four trials, chicks selectively chose the 3rd and the 4th left elements above chance (3rd left element: Mean=23.077, ES=5.979, Median=25.000; V=76.000, p=0.017, $r_B=3.222$; 4th left element: Mean=32.692, ES=7.692, Median=25.000; V=81.000, p=0.007, $r_B=2.600$) (see **Fig. 3c**). No difference was there between choice of the 3rd and the 4th left element (W=22.000, p=0.193, $r_B=0.571$).

Right monocular: The chicks chose both the 4th right element and the 3rd right element above chance (4th right element: Mean=25.000, ES=3.397, Median=25.000; V=76.000, p=0.002, $r_B=0.670$; 3rd right element: Mean=29.625, ES=3.645, Median=30.000; V=85.500, p=0.001, $r_B=0.967$); but not the 4th left element (Mean=5.000, ES=1.179, Median=0.000; V=9.000, p=0.993, $r_B=-1.000$) and the 3rd left element (Mean=0.769, ES=0.521, Median=0.000; V=0.000, p=1.000, $r_B=-0.143$) (see **Fig. 2d**).

Chicks pecked more often at the 4th right elements than at the 4th left element ($W=76.000$, $p=0.004$, $r_B=0.949$). They identically selected the 4th right element and the 3rd right element ($W=24.000$, $p=0.445$, $r_B=-0.273$).

In the first four trials, chicks selectively chose the 3rd and the 4th right elements above chance (3rd right element: Mean=19.231, ES=5.027, Median=25.000; $V=76.000$, $p=0.016$, $r_B=3.222$; 4th right element: Mean=32.692, ES=7.153, Median=25.000; $V=85.000$, $p=0.003$, $r_B=2.091$) (see **Fig. 3d**). No difference was there between the 3rd and the 4th right element ($W=47.000$, $p=0.219$, $r_B=0.424$).

DISCUSSION

The aim of this study was to disentangle the engagement of either hemisphere in dealing with the ordinal and spatial information in a serial ordinal task. Results of the sagittal test, performed in binocular condition of vision, indicate that chicks correctly memorized and used the integration of spatial and numerical cues to locate the target element. These findings are in line with previous studies^{10–12,16} and sustain the idea that birds are precociously capable to use ordinal-numerical information to find a food source. Birds, in the sagittal test, chose the target element (the 4th one) over all the others. Whenever the analysis was restricted to the very first four trials, only the 4th element was selected above chance. If we consider the whole test, chicks pecked also at the 3rd element above chance, but the 4th element was selected more than the 3rd. A possible explanation for this could be that, in identifying the target element, chicks anchored their evaluation on the closest end of the sequence (i.e.,

somewhat similarly to the “working up strategy” described by Suzuki and Kobayashi (2000)³⁹. This possible interpretation is also supported by the outcome of a previous study. In such a study, different groups of day-old chicks learnt to identify the 3rd, the 4th or the 6th element in a sagittal series on ten identical elements. Whenever the 3rd (or the 4th) element were the target, chicks made more errors respectively on the 2nd (or on the 3rd) element; while chicks for whom the 6th elements was the target mistook more on the 7th element¹⁰. These results can indicate that birds would anchor to the 1st element, to identify the 3rd and the 4th element, hence making more mistakes on the element immediately preceding the target rather than on the element following the target. Similarly, the chicks may anchor to the last element (i.e., the 10th one) when identifying the 6th element (i.e., see the “working down strategy” described by Davis and Bradford (1986)⁴⁰. Results from the binocular fronto-parallel test showed that chicks identified the target element, basing on serial ordering, and neglecting the elements at the correct distance (the 3rd elements). Both 4th elements were pecked above chance; considering the whole test, chicks pecked more often at the left than at the right target, but restricting the analysis to the first four trials, the 4th elements were equally selected (for similar findings see¹²). They were able to generalize what learnt to a new series, characterized by i) a different orientation with respect to the one experienced during training, ii) a different metric, in fact by increasing the inter-element distance also the overall length of the series increased. Interestingly enough they selected the numerical-ordinal target elements in a presence of a conflict between the numerically correct target (the

4th elements) and the spatially correct one (the 3rd, which was the one located at the correct distance at training).

To disentangle the engagement of each hemisphere in dealing with the “spatial vs. ordinal” test, we used the monocular occlusion technique. The domestic chick is a unique model to study hemispheric specialization. In binocular condition of vision, the information from the eyes reaches, throughout the tectofugal and thalamofugal pathways, the respective telencephalic areas: the Ecostriatum and the visual Wulst³⁵. However, thanks to the decussation at the optic chiasm of both the tectofugal and thalamofugal pathways⁴¹ and to an almost total lack of interhemispheric connections (see introduction), a simple temporary patch over one eye can be used to discern hemispheric specializations⁴². A common feature of the two visual pathways is their lateralization⁴³, in chicks stimulus processing is usually carried out by the eye-system specialized for the given task⁴⁴. If one hemisphere is dominant in processing spatial or ordinal information, we would expect an asymmetry in behaviour when the information is elaborated with the “specialized” eye-system. In both monocular conditions of vision, the birds efficaciously performed at the fronto-parallel test, but in these cases they relied on both numerical and spatial cues. In fact, they identically chose the 3rd and the 4th element (either on the left or on the right) significantly above chance. This is true when we considered the whole 20 testing trials as well as when we limited the analyses to the very first four trials. In monocular conditions of vision birds mainly directed their responses on the elements located in their clear hemifield. Because of the eye patch, only one end of the series either left or right was clearly visible, and

1 chicks used this side as benchmark to start to “count”. Whenever their vision
2 was limited to the left eye, they equally chose the 3rd and the 4th left elements;
3 when they could see with their right eye, they equally chose the 3rd and the 4th
4 right elements. Thus in different trials the animals relied either on spatial or
5 ordinal cue. This shows that the spatial and ordinal information acquired during
6 binocular training can be correctly represented, and independently used by
7 each hemisphere, though with a different anchor end that identifies the starting
8 point for “counting”. This anchoring has been reported also when both spatial
9 and ordinal cues were available at fronto-parallel test¹⁶. The bilateral
10 hemispheric representation of numerical processing found here is coherent with
11 scientific literature^{45,46}. Overall these findings enlarge our knowledge on the left-
12 to-right oriented bias previously reported in birds¹¹ and non-human primates²⁰.
13 Here we show that both hemispheres encoded and used spatial and ordinal
14 cues to find a food source. The left bias reported in previous studies seems
15 therefore to be based on an integration of numerical and spatial cue by the right
16 hemisphere.

17 Up to now there is no study that has investigated how numerical-ordinal
18 information is elaborated and integrated by the two hemispheres. Future studies
19 are needed to understand (i) how and where this integration of information
20 occurs and (ii) if a same mechanism can be potentially extended to explain
21 other SNA effect, like associations between numerosities and sides in space,
22 which were described in infants⁷⁻⁹ and also in chicks¹⁵. An insightful reflection
23 could arise by comparing the behavioural responses in the fronto-parallel test
24 performed in binocular conditions of vision with those of the monocular

conditions of vision. Whenever the birds could use both cerebral hemispheres, they selectively relied on numerical-ordinal cues, whereas when the suitable hemisphere was only one (either the left or the right one), they relied on both ordinal-numerical and on spatial cues. This might allow to speculate that the basis of the spatial numerical association (SNA) might be accounted for by a model that assumes differential encoding, processing and integration by the two hemispheres for spatial and numerical information. Up to now there is no sufficient evidence to individuate the origin of SNA. Nevertheless, our results seem to indicate that a valuable answer will be obtained once we will have better understood how and where numerosnesses are represented within the brain.

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AUTHOR CONTRIBUTIONS

R.R. and L.R. designed the study; R.R. analyzed the data; both authors interpreted the results; R.R. wrote the paper and created the figures; L.R. critically reviewed the paper.

COMPETING INTERESTS

All authors must include full disclosure statements at time of submission as a section at the end of the manuscript text.

Figure legends

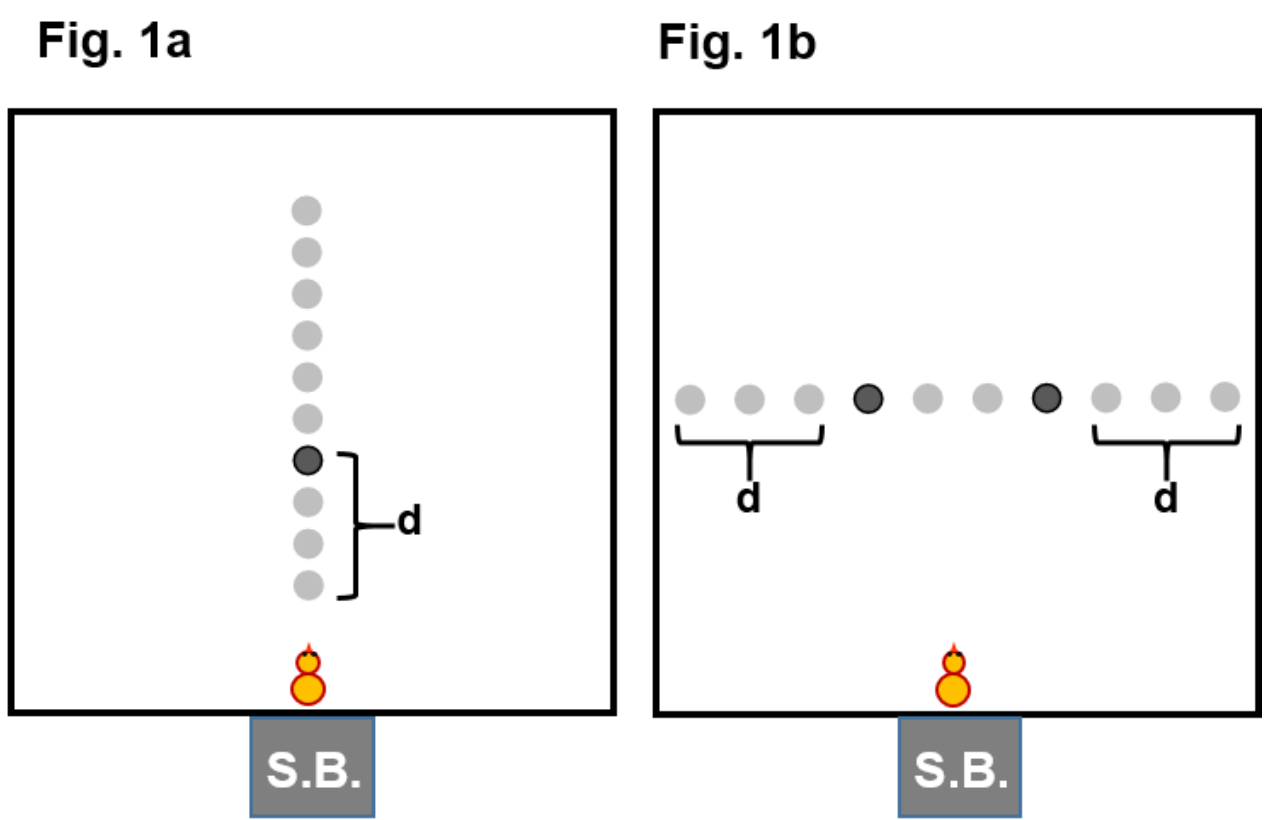
Fig. 1. Schematic illustration of the apparatus and the series of elements; “d” indicates the distance from the closest end of the series during training. All elements looked identical, but in the illustration, the target is grey. “S.B.” represent the starting box; the chick’s starting position. (a) The disposition of the series during training and during the sagittal test. (b) The disposition of the series during the fronto-parallel tests.

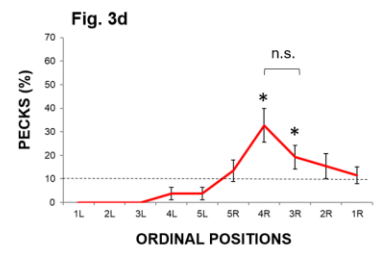
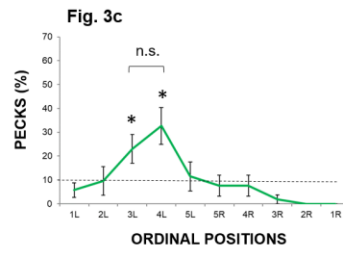
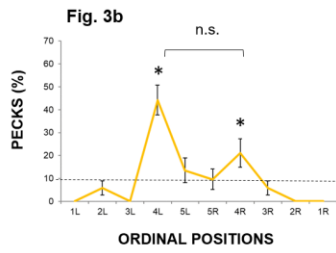
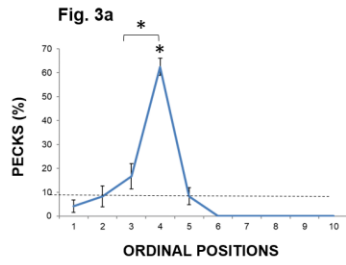
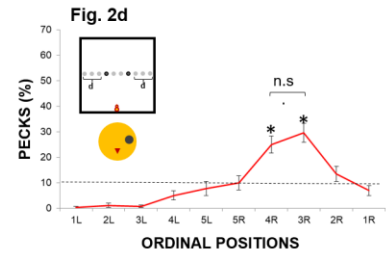
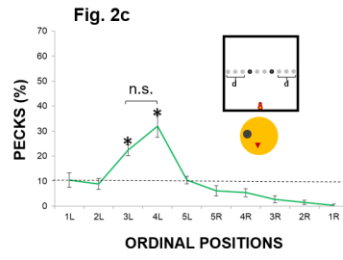
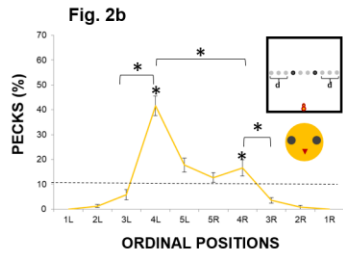
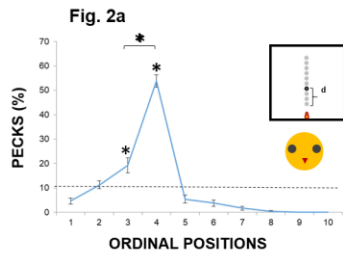
Fig. 2. The graphs represent the mean percentage \pm SE of choices for each element in the 20 testing trials; the dotted line represents chance level ($y=10$); asterisks indicate $p<0.05$; the schematic chick heads indicate the condition of vision; the schematic illustration of the apparatus and of the series indicates the test. (a) Results of the binocular sagittal test. (b) Results of the binocular fronto-

1 parallel test. (c) Results of the left monocular fronto-parallel test. (d) Results of
2 the right monocular fronto-parallel test.

3 Fig. 3. The graphs represent the mean percentage \pm SE of choices for each
4 position in the first four trials; the dotted line represents chance level ($y=10$);
5 asterisks indicate $p<0.05$ (a) Results of the binocular sagittal test. (b) Results of
6 the binocular fronto-parallel test. (c) Results of the left monocular fronto-parallel
7 test. (d) Results of the right monocular fronto-parallel test.

8
9 **Figures**





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