

**UNIVERSITY OF PADOVA**

**Department of General Psychology**

**Bachelor's Degree Course 'Psychological Science'**

**Final Dissertation**

*Domestic Chicks' Preferential Choices for Sets of Odd Numerosity*

*Supervisor*

*Professor Lucia Regolin*

*Co-Supervisor*

*Maria Loconsole*

*Academic Year: 2023-2024*

*Candidate: Areia Romero Alcober*

*Matricola Number: 2037295*

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## **0. Abstract**

In this current study newly hatched domestic chicks were tested in their first stages of development, to assess their performance in two testing paradigms that in previous studies had led to different results when administered to separate groups of animals: spontaneous preference following 1 hour habituation, or prolonged exposure. In the present study each chick underwent both paradigms, therefore it was tested twice and in both cases the chick was presented with the numerical comparison of 5vs9 objects. Chicks were first tested following 1 hour habituation to heterogeneous sets of 2D elements. Based on previous studies a preference for the set of 5 elements is expected. Results confirmed this hypothesis. Thereafter each chick underwent prolonged exposure to a set of 7 elements (red balls) and through filial imprinting the chick was expected to learn the features of this set, before being tested again in the 5vs9 discrimination. However, the test following prolonged exposure (imprinting) did not result in a significant choice, differently from previous studies in which chicks in this test preferred the numerosity 9. The difference could depend on the two conditions and tests interfering with one another, preventing the understanding of the underlying differences among the two paradigms.

## **1. Introduction**

### **1.1 Numerical Discrimination**

Numerical discrimination is an important skill within cognitive development, and essential for non-human animals to make decisions that could increase chances of survival (Nieder, 2020) in contexts such as foraging (Hunt et al. 2008), defense from predation (Hager, & Helfman, 1991), or territorial fighting (McComb et al. 1994). It includes the ability to equate, compare and make decisions based on the quantity given of a set of a group. For instance, with the essential skill of foraging, it's important to aim for optimal food selection, through set group distinction to select food patches and areas that provide the highest quantities of feed. New Zealand robins are an example of a species that shows the evolution of numerical competency, by retrieving and stealing stored food using a sophisticated numerical sense (Hunt et al. 2008). As for predation, we've seen lions estimate whether a group of opponents is larger or smaller in number than theirs, and thus make a decision of whether to engage or not (McComb et al, 1994). Additionally, for territorial fighting, we can see fathead minnow, which prefers larger shoals for safety but lacks precise counting abilities. They reliably choose larger groups in extreme cases, like avoiding solitude with a predator nearby and in high-risk situations, to maximise survival chances (Hager & Helfman, 1991)

### **1.2 Domestic chicks as a model species**

Newborn chicks, as highly gregarious creatures, have a remarkable innate ability to spontaneously estimate numerosities, as well as a preference for larger groups of familiar stimuli. This behaviour is probably motivated by evolutionary advantages in terms of safety and survival,

as it is beneficial for them to be able to identify which group has the most members when the groups are separated. This has been corroborated by a number of studies that showed that within the first week after hatching, newborn chicks were able to distinguish between different numbers and shown a preference for the larger set of artificial objects they had been reared with (Rugani et al., 2013, 2017, 2020, 2022). Seeing as chicks saw these objects as social companions, it shows us that it is advantageous to be able to find and join the larger group if the social group is split into two or more subgroups. This early capacity to recognise small numbers from birth suggests that, like human infants, chicks recognise individual objects as discrete units, suggesting that numerical identification is an innate rather than learnt capability. This makes chicks an excellent subject for numeric discrimination studies.

### **1.3 Prime Numbers vs Composite (Novelty vs Majority)**

In addition to numeric discrimination in groups, a particular phenomenon in numerical cognition is seen with prime numbers. Prime numbers are of interest to us because of unique perceptual qualities recognizable in nature, their main characteristic being that they can not be symmetrically divided. Symmetrical grouping refers to the ability to take a group of elements and create equal subsets within that number. For example the number 9 is easily divisible into equal subgroups, three groups of three, ( $3 \times 3 \times 3$ ), whereas attempting to conduct the same process with 5, a novel number, that cannot be separated, is much more tasking. This form of grouping creates a balanced and harmonious configuration, aligning with Gestalt principles like symmetry and perceptual simplicity (Loconsole & Regolin, 2022). Composite numbers, as research has shown, being divisible into symmetrical subgroups, allow for a mental multiplication that facilitates a faster, more accurate judgment of numerosity. This mental shortcut simplifies

perceptual grouping and assessment of the sets (Ciccione & Dehaene, 2020). For this reason, this study uses a comparison choice between a prime, or novel, number (5) and a larger, composite number (9).

We have also seen this phenomenon in species such as cicadas, which emerge every 13 or 17 years, as an example of prime-number timing that minimizes predator encounters. In studies with baby chicks, researchers found that chicks preferred groups of objects that couldn't be symmetrically divided, during exploration of new stimuli. This indicates that chicks might instinctively process numerical patterns based on perceptual cues, like novelty, rather than the object's magnitude, and that the ability to distinguish prime numbers could be an innate perceptual skill shared across species (Loconsole et Regolin, 2022). In the work leading to this paper, the experimental group of chicks was habituated with heterogeneous sets of 2D stimuli, and when tested, would most likely prefer to go towards, or be near, groups with 7 or 11 elements rather than those with 9. This would suggest their choice was not based on the numerosity's quantity alone. Chicks seemed to use perceptual cues, such as the asymmetrical grouping of prime numbers, which they found novel or visually appealing, in spontaneous preference tasks, following habituation procedures (Loconsole et al., (2021). Since the 2D stimuli is not as exciting, baby chicks have been found to be attracted to mild novelty during exploration, when presented with two complex but different stimuli (one familiar and one unfamiliar), chicks spent more time near the novel image, meaning a group of 5 elements, over one of 9 elements (Jones et al., 1996). Additionally, results showed that the chicks spent longer exploring the set of a prime numerosity, irrespective of the prime set being the smaller or the larger of the pair (Loconsole et al., 2021, 2022).

On the other hand, when the one-day-old chicks were made accustomed to homogenous 3D stimuli, and made to choose between two different sets in a similar free-choice task, afterwards favored the larger set (Rugani et al., 2010, 2011, 2013). This points to the possibility of how imprinting could help accustom the chicks to foreign stimuli and whether that had an impact on their numerical choice. Filial imprinting refers to a phenomenon by which birds that frequent the ground follow form a rapid and special attachment to any object they are exposed to within a specific period of their life, the critical period, occurring shortly after hatching, usually within 1-3 days (McCabe, 2019). Imprinting helps increase a newborn organism's chances of survival by creating a bond with their closest guardian, this usually being their mother. This recognition is critical for maintaining close proximity to their mothers, who provide food and protection, as an evolutionary advantage. We can see that shortly after hatching, domestic chicks will respond socially to a wide range of living or inanimate objects, and view them as companions (Rugani et al., 2010). As a result of imprinting, their social behaviour is increasingly limited to objects with which they have had visual experience and, generally, chicks develop a stronger preference for an object the longer they have been exposed to it, favoring it over alternatives when presented with a choice. This is mainly important during the early stages, which is why we will stick to this window of time, where imprinting seems to have the strongest connection to forming a relation with a conspicuous object (Jackson et al. 1974).

## 1.4 Present Study

By combining these two conditions, which were tested isolated from one another, and instead using a repeated measures method with the same subject group to—first, habituation with heterogeneous 2D stimuli in a 9 vs. 5 free-choice experiment, followed by an imprinting task with 3D stimuli in a similar 9 vs. 5 experiment— it can be determined whether there is an interaction between these mechanism, or whether they occur independently. As mentioned, the object being imprinted on is not limited to their own kin, this process could be done interspecies, or even on inanimate objects, which will be used in this study. There also needs to be a consideration of task complexity, as seen in previous studies that they are able to distinguish numbers with around a 0.5 ratio, similar to previous literature, and lower numerosity (Rugani et al., 2014; Loconsole et al., 2021). Thus the reason for the number of elements to not be too large and demanding to process, choosing the simpler 5 vs 9.

The predicted hypothesis is that in the Habituation phase, the baby chicks will show a preference for the smaller, novel set (5) consistent with prior research, and, conversely, in the Imprinting phase, the same group of subjects, will have an inclination towards the larger numerosity (9). These results will showcase that their abilities to discriminate numbers are flexible, they adapt depending on what the circumstances require. The findings will tell us whether this mechanism is or isn't a rigid one, and explore novelty vs magnitude in preferential choices. Already having these conditions prior to this thesis, this work aims to bridge the gap between them, examining whether there is a relationship or interaction between them, or if they operate independently.

## 2. Materials and Methods

### 2.1 Subjects and Equipment

The subjects of this study were domestic chickens (*Gallus gallus*) in their first days after hatching. Newborn chicks (N=36 chicks, 18f and 18m), were tested, 6 subjects per week along 6 weeks of testing 12 fertilized eggs will be used each week. Incubation conditions were controlled, temperature (37.5°C) and humidity (55-66%). From these 12 eggs, which will hatch into newborn chicks, 6 take part in the study, the other 6 were kept in groups determined by their gender, and, in case of one of the subjects failing the habituation on the first day, will be swapped in. The eggs were kept in an incubator, and when selected (if hatched) for the experiment, they will be given a name (M1, M2, M3,...) to keep a track of their progress, both for the experiment, and to take their weight, to control health. Controlling health refers to ensuring that the chicks were not losing weight, or staying the same weight.

The testing area is a triangular arena of dimensions 93 cm x 62 cm x 30cm. The triangular shape encourages chicks, to move forward, and make a choice, as opposed to get distracted by other corners and reducing possible backtracking. There is also a monitor showing the stimuli for habituation, which works as the 3rd wall of the shortest side of the triangle. For the imprinting task, we have a device consisting of two columns, for it to stand on, and 3D plastic red balls, (4x3x3 cm), 14 in total, hanging from plastic strings, which can slide over the stick they were being suspended by, in order to optly change the number of objects (5 vs 9, or 9 vs 5) in one side or the other of the vertical divisor, a partition (5x30 cm), which is there to clarify which choice of right vs left the chick has made. A choice is only counted if the chick crosses this threshold, as they were not able to see the other side at all. Lastly, to record the data, a video camera (Canon-Legria HF-R606) was placed about 30cm above the arena, to record each

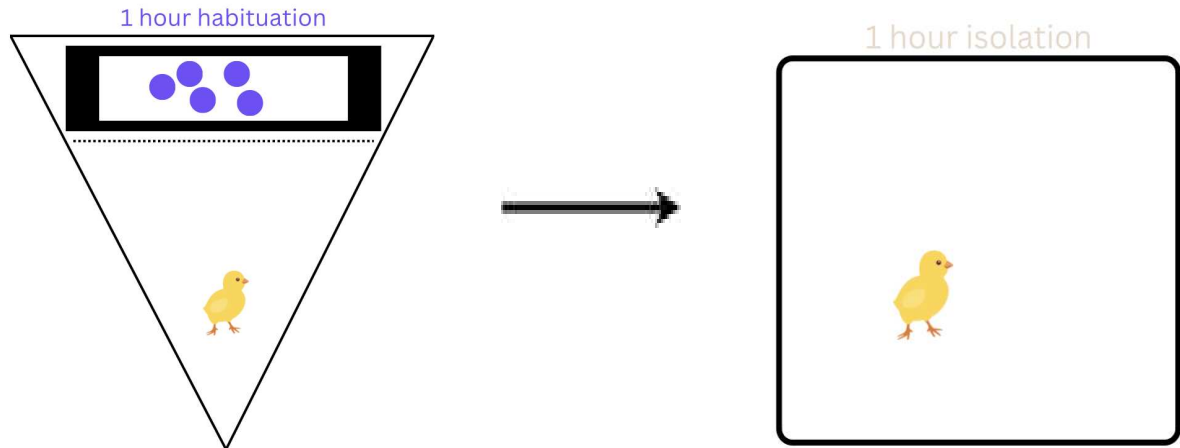
chick's experience is overlooking the testing arena, and a white board to display their name, sex, the location of the prime number (left or right) and which type of test they were taking part in (HAB for habituation or IMP for imprinting). The enclosure room consists of standard metal cages (28x32x40cm). Experimental conditions were controlled within the procedure room, such as lighting, since the same standard ceiling lights was used, consisting of a 36W lamp, 45cm above the cage floor, with temporized light-dark cycle (light from 7am to 7pm, followed by dark-light alternated phases of 2/3h each). These conditions remain constant for all subjects, in the experimental room, as well as the enclosure room, to ensure comfortable and uniform conditions for the baby chicks.

In order to further ensure and track the chick's safety and health, a general rearing procedure that must be conducted once the chicks were placed in the enclosure room following habituation or imprinting. The chicks were sexed by the wing, "named" according to their number in the experiment (M1, M2, M3,...), and their weight is noted. Weighing of the chicks happens every day, at the end of both habituation testing and imprinting testing, to monitor their health and ensure they were gaining weight. It is essentially that on the first day of life chicks eat enough and gain weight, thus they were weighted daily to ensure that they were eating and correctly absorbing food, otherwise it could be dangerous to their future development. For chicks that have issues gaining or maintaining weight, a closer look at their eating habits is necessary, as well as administering extra worms to increase their feed intake. The subjects not taken part in the tests and data collection will be placed in enclosures with 2-3 chicks, subjects were now grouped off depending on their gender. There is an observation in the first hour to ensure none of them fight or peck at each other, in the event of an aggressive incident, the perpetrator will be placed

behind a see-through screen for a few minutes so it would understand the behavior is inappropriate then reintegrated into the group. Testing subjects will be grouped off this way once imprinting testing is complete, as they need to be in isolation before that. Overall one glass of water will be provided per chick, and one glass of food per cage, the glass containers were wide and short in order for them to be able to reach them fine. They were also taught how to catch a worm by dropping them in the home cage in front of them.

## **2.2 Habituation of 2D Objects**

Starting with the Habituation (Fig. 1), on the first day of life, the newly hatched chicks were transported from the incubator to the testing area. The chicks were transported in a large plastic rectangular box, with tissue at the bottom, and when picking them up, handled with care and black gloves to not contaminate them. For an hour, in a dark room, the chicks were exposed to a display of an alternating number of objects on a computer, (Fig. 1) showed in a random order, in different in shapes, including squares, triangles and circles, and colors, including blue, green, yellow, red, as well as amount, always in even numbers, (4, 6, 10, 12). Even numbers were used to get them acquainted with the stimuli, but not with the characteristics of prime numbers, and not with the specific number of stimuli used in this experiment, which were numbers 5 and 9. Therefore all HAB stimuli were composite, to not alter their decision making by overexposing them to one or the other. An hour was necessary for them to get used to the environment and familiar with the shapes and stimuli. Following this hour, chicks were put into the feeding enclosure areas in isolation for another hour (Fig. 1), so they can process the information they received and to not interact with another chick yet as they'd be distracted being highly social animals.

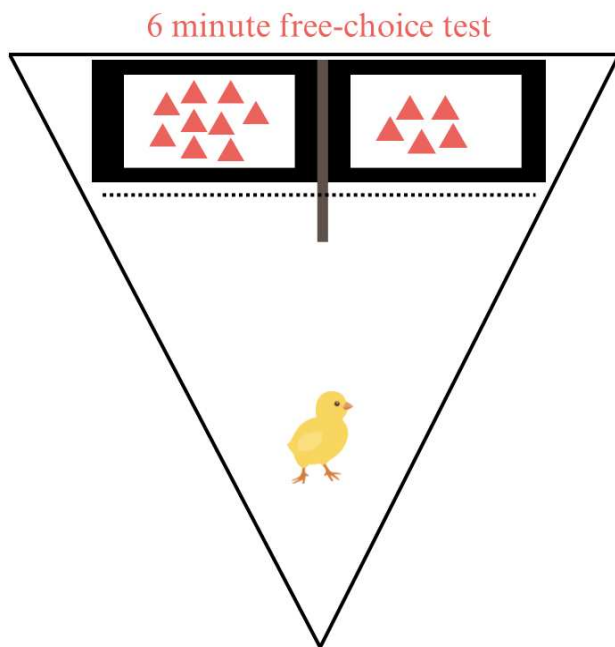


**Figure 1. Habituation and Isolation procedure.** Immediately after hatching, chicks were exposed for one hour to an experimental arena featuring two-dimensional (computer-generated) stimuli that varied in shape, color, and even numbers. Following these exposures, the chicks were kept in isolation in their home cages for an hour to process information.

### 2.3 Free-choice test following Habituation

Following one hour of isolated time, the chick were tested for 6 minutes, in a free-choice task (Fig. 2) with either 5vs9 or 9 vs 5 comparison, randomly assigned, but an equal number of chicks will be tested in both. Testing took place in the dark as well, so the monitor was the main stimuli they focused on. The difference between the conditions is, in 5 vs 9, 5 objects, similar to the ones shown during the one hour of habituation, (Fig. 1) were shown on the left of the monitor screen, and 9 on the right, whereas 9 vs 5 sees 9 objects projected on the left, and 5 on the right. Both sections were separated by a cardboard screen, opaque to properly note which side the chick has chosen. It is important for the tester to not be visible to the subjects whilst they make the choice as it could confuse or distract them, thus they were monitored through the camera to

check if they made a choice during the 6 minutes or not. Time scoring of choice was counted by the tester, while using the event-logging program Boris, (Friard, 2016) only if the chick crosses the invisible line where the cardboard begins, meaning once they've crossed far enough that they were only able to see the side they picked. In the case that one of the chicks fails to make a choice within the 6 minute examining period, the chick will be taken out of the testing area for a few minutes and tried a second time, if the lack of choice persists, the chick will be swapped for another subject and will no longer participate in the study. Once the 6 minutes were up, the chick is placed in the transportation container again, now to be put into the enclosure with food, water, grain, and mealworms as additional diet, and the 3D objects, which were composed of 7 red balls, identical in size, shape, and weight, of 4x3x4 cm, hanging from a transparent thread. The reasoning behind the number 7 for imprinting is because it is a prime number, in between 5 and 7, not easily chunked into smaller bits, to not interfere with their recognition of one or the other. Additionally, 7 is equidistant from 5 and 9, the testing stimuli, meaning not leaning towards one or the other, avoiding a priming effect, or preference bias, due to it being “more similar” and familiar to the rearing numerosity.

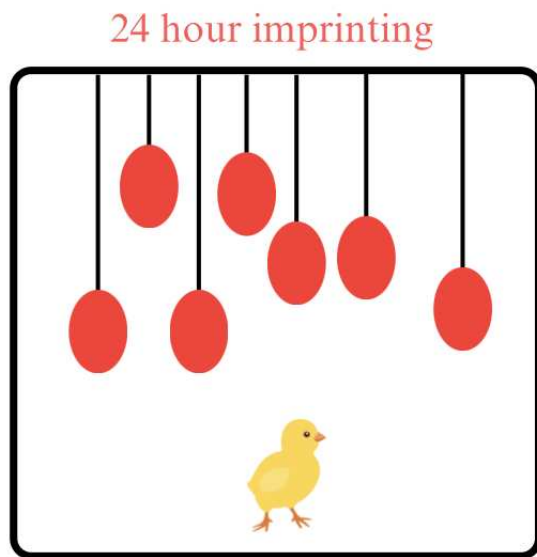


**Figure 2. Free-choice Habituation Test.** Once the hour of Isolation was up, the chicks underwent a 6-minute free-choice test involving a 5 vs. 9 numerosity comparison, in the same triangular testing area as the Habituation (Fig. 1). The numbers were displayed on a computer screen, and separated by a partition (5x30cm) to make each choice of set clear.

#### **2.4 Prolonged Exposure to 3D Elements**

Once the habituation is concluded for all the subjects, they were kept in their standard metal home cages with the 3D imprinting objects (Fig. 3), 7 identical red balls (4x3x3 cm) suspended at 2-3 cm from the cage floor via a transparent thread for about a day, in which they were expected to form a social bond with, as previously mentioned they are highly social animals, and will develop a familiarity with the objects, becoming highly motivated to join the larger of two groups of these objects (Rugani et al. 2011, 2013, 2020). These stimuli were

identical to the ones in imprinting tasks the following day (Fig. 3).

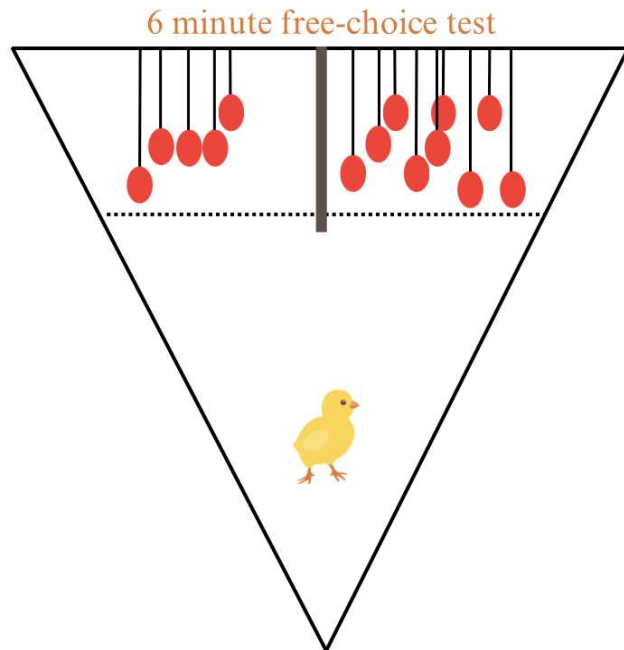


**Figure 3. 24 hour Imprinting.** Chicks were reared for 24 hours with seven identical, 3D red balls, suspended from above, in their home cage.

## 2.5 Imprinting Task

For the imprinting test (Fig. 4) following prolonged exposure, the contraption with 3D stimuli of hanging red balls was used. This was separated by a cardboard wall, in a 5 vs 9 or 9 vs 5 comparison, matching according to their condition in the habituation the prior day. This means in a 5 vs 9 condition, the 5 (prime) hanging balls will be on the left side, and 9 (non-prime) on the right, and vice versa for the 9 vs 5 condition. Placing the chick gently in the testing area, specifically in the corner in front of the contraption, it's important to drop them a couple centimeters off the ground so they don't start running in a random direction as they usually do if you place them directly on their feet. Subsequently, we record their choices for 6 minutes, in a similar free-choice task. In accordance with the previous habituation choice recording, a choice

is only counted if the subject crosses the threshold where they can no longer see the other side, so inside the cardboard border. In this case, in the event of them not making a choice, it will be noted as null. Once the experiment is up, they were placed back into the home cages until all subjects were finished with the test, conducting the rearing procedure detailed 2.1 before.



**Figure 4. Free-choice Imprinting Test.** Following prolonged exposure to the 3D objects, chicks were subjected to a 6-minute free-choice test involving a 5 vs. 9 numerosity comparison, where the prime was on the left, or 9 vs. 5, were the prime was on the right, depending on the chick's previous Habituation test (Fig. 2), to keep consistency. The testing objects were identical to those encountered during 24h Imprinting (Fig. 3), but the test was conducted in the testing arena. The stimuli were suspended by the apparatus described in *2.1 Subjects and Experimental Area*, and the two sets were once again separated by the same partition as in Habituation Test (Fig. 2), to clearly note which choice the chick made.

## **2.6 Data Collecting and Analysis**

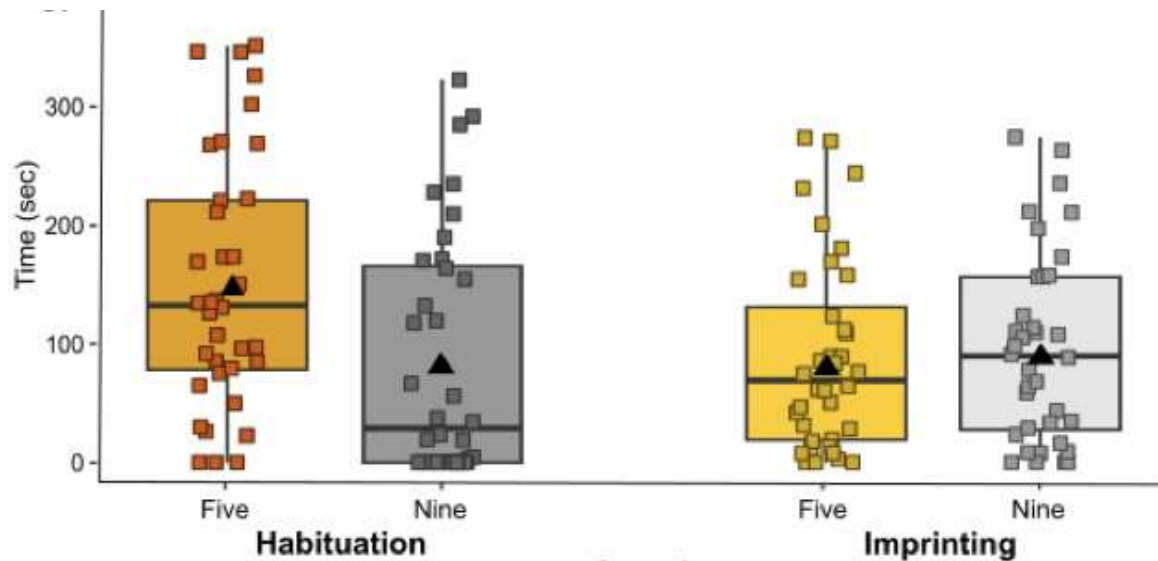
Once both tests were conducted and recorded, using a video camera, data was collected and analysed using a video free program called Boris, (Friard, O., 2016), useful for logging events in audio and video formats, to make observations, particularly useful for live subjects with many versatile movements. This software can note the timestamp when the chick would start and end the experiment, cutting it at the 6 minute mark. Additionally the program allows you to collect how much time the chick spent in each group of stimuli, 5 or 9, only after passing the vertical partition separating both of them, noting that as a choice being made. The number between the prime and non prime the chick chose to explore first was also noted. Data was organised in an excel spreadsheet, with the number of the subjects in accordance with their spot in the testing, as well as their sex, their choices and any additional notes and observations. Data was then analyzed using R 4.2.0, (R Core Team, 2019). A linear mixed model was employed, here the dependent variable was the time spent in each choice area, and the independent variable was the number of elements of the comparison (i.e., 5 or 9), and then processed data was displayed in a box plot graph with said conditions.

## **2.7 Ethical Considerations**

The experiments complied with all applicable national and European laws concerning the use of animals in research and were approved by the Italian National Ministry of Health (N.I.H.). Experimenters underwent a safety course prior to working with the live animals and laboratory equipment. All procedures employed in the experiments included in this study were approved by the Italian Ministry of Health (permit number: 196/2017-PR granted on 24/02/2017).

### 3. Results

These findings offer insight into how chicks' preferences for different numerosities and how they adapt to environmental exposure, as hypothesized. Over the course of 2 months (October 2023 and November 2023) 36 chicks (18 females and 18 males) were tested for this study. The data from two of the examined chicks could not be used for that category since they were unable to make a decision during imprinting. This refers to the previously explained guideline in 2.3 *Free-choice test following Habituation* of when a choice is made, stating that the chick needs to cross the threshold where they can no longer see the other set group of 3D stimuli. Overall we found a significant interaction between the stimulus (5 or 9), and experimental condition (habituation or imprinting), ( $\chi^2 = 5.36$ ,  $p = 0.02$ ). Following habituation, the chicks spent notably more time near the novel set of 5 elements, (estimated mean difference (5-9) = 64.7, SE = 22;  $t = 2.94$ ,  $p = 0.01$ ). This findings shows a consistent preference for the novel set with lower numerosity after exposure to the 2D stimuli. Following imprinting, the chicks' preference for the set of five items vanished, and there was no discernible difference in time between the two sets (estimated mean difference (5-9) = -7.4 seconds, SE = 22;  $t = -0.34$ ,  $p = 1$ ). When comparing the two conditions, chicks spent significantly more time near the set of five elements following habituation than following imprinting, with a mean difference of 60.84 seconds (SE = 22;  $t = 2.76$ ,  $p = 0.03$ ). Analysing the first approach choice revealed that, following habituation, the chicks' initial approach was primarily towards the set of five components ( $P(5) = 0.72$ ,  $p = 0.01$ ), indicating a strong initial attraction to the lesser, but novel, numerosity. Following imprinting, chicks showed no consistent preference ( $P(5) = 0.53$ ,  $p = 0.73$ ). These findings suggest that their response to the numerosity of element sets (5 vs. 9) depend on whether they were in the habituation or imprinting phase.



**Figure 5. Results.** In this box plot graph, two testing conditions are being shown in the X axis, (Habituation and Imprinting), depending on which one the chicks underwent. The unit of measurement is in the Y axis, which is the duration in seconds chicks spent close to each numerosity. The triangles in each box plot represent each mean (average time), the gray line the median (the ‘middle’ of the data), and the small squares represent individual performance for each of the subjects. In orange, the time close to the prime (5) stimuli during the Habituation condition, and in yellow time close to the prime stimuli in Imprinting. Dark gray represents time spent near the non-prime (9) in Habituation, and light gray represents the non-prime stimuli in Imprinting.

Domestic chicks spent longer in the novel set 5 in the left column (data collected in test after habituation), but in the right column (imprinting data after prolonged exposure), the preference vanishes and the results are similar, with no discernible difference in the amount of time spent near either set. The data reveals that even though day-old chicks initially exhibited a preference for the prime number numerosity after habituation, this bias was significantly reduced

to produce even results following a full day of exposure and familiarisation with the stimuli. This decrease in preference would suggest that extended exposure modifies the chicks' initial reaction to numerosities, either as a result of equal familiarity with both sets or a process of desensitisation. For information about the raw dataset collected and used to create the box plot of this study, contact Professor Lucia Regolin ([lucia.regolin@unipd.it](mailto:lucia.regolin@unipd.it)) or Maria Loconsole ([maria.loconsole@unipd.it](mailto:maria.loconsole@unipd.it))

## **4. Discussion**

### **4.1 Analysis of Results**

While the results show some concordance with past research, the most interesting part is where they differ. The results showed that, even though these conditions (habituation and imprinting) separately have distinct effects on chicks' preferential choice, combining both strategies within the same subjects gave us a perspective of the interaction between both conditions. The habituation testing with 2D models in this experiment showed results that aligned with past research, as evidenced by the fact that they spent a lot more time near the set of five elements than the set of nine. However, when the imprinting test was conducted, the initial preference was diminished or eliminated altogether, as a result of the 24-hour exposure to 3D, salient objects. Even while longer exposure did not result in a particular preference for either group, as opposed to previous research where chicks showed a preference for the greater numerosity set, these results suggest that exposure duration may have an impact on preference as it shows a weakening of the initial preference for the set of 5 after prolonged exposure. Following imprinting, however, chicks showed no consistent preference, which lends more

credence to the notion that the initial preference was weakened by the extended exposure. These findings suggest that their response to the numerosities (sets of 5 vs. 9) depended on the interactive effects of habituation and imprinting conditions, and thus affected the choice mechanisms of newborn chicks.

These results point to the following findings. First of all taking into account novelty vs. magnitude, the preference for novelty in habituation highlights the importance of perceptual differences in early cognitive strategies. This suggests that novelty may serve as a mechanism for exploration of unfamiliar stimuli. On the other hand, the results following the imprinting interference showing this preference vanished could be interpreted as the familiarity and salience of a 3D object making baby chicks more likely to pick the larger numerosity than in the previous condition, but still doesn't equal to a unequivocal inclination. After combining both conditions, the results suggest that cognitive mechanisms underlying numerical and perceptual processing may interact in complex ways, with potential to change according to what the circumstance the chicks are in. These mechanisms are flexible and influenced by exposure, as they adapt depending on what the circumstance requires; it is not a rigid behavior. Chicks' cognitive preferences are highly adaptable and can be shaped by specific experiences, indicating an ability to toggle between perceptual and numerical processing based on environmental cues.

## **4.2 Limitations of the Research**

A possible limitation is that these conditions have not yet been tested by reversing paradigm order, to control for other potential carryover effects. Meaning there is no research showing what effect would occur if the same subject groups underwent a repeated measure test

in which they first were prolongedly exposed to 3D objects and then 2D ones. Switching the conditions could give further information on how the mechanisms reacting to novelty vs composite numerosities interact when a subject group undergoes different stimuli. This would entail going instead, from a more salient condition (3D objects) to a less salient one (2D), to observe whether there is a response, or other-way effect. This could be an example of future research. It would also be interesting to change the prolonged exposure, instead making it the 2D objects. This could show whether, even though not as salient, they will still trigger an attachment, and go for the larger set (Rugani et al. 2011, 2013, 2020), or if this would reflect, instead, the inherent exploratory behavior of the novel number we have seen in previous research (Jones et al., 1996).

Regarding imprinting, there is a chance that in early stages, the trend towards a highly restricted preference is temporarily reversed and some birds may even prefer the unfamiliar object, in order to explore, so this could be a phenomenon affecting the decision making (Jackson et al., 1974).

### **4.3 Future Research**

As mentioned in section 4.2, future research could reverse testing order, beginning with an exposure to 3D objects, followed by 2D stimuli. This could clarify whether they would affect each other, by noting a change in the preferential outcomes, or no effect, failing to change the intrinsic properties of each condition. It could also be interesting to, in the future, test how much familiarity plays a role in quick and efficient decision making between two sets. This could be done by having a different set of stimuli, this time more salient (for example making the 3D objects resemble baby chicken) or improving the ecological validity by further resembling

outdoors or making the set groups other real-life chicks. Research has shown that chicks will go for artificial objects in controlled conditions in which they do not have any other features to rely on (Rugani et al., 2010) thus, it would be interesting to provide them with different, more ecological options.

It could also be incredibly interesting to relate such results to humans' early experiences. The findings of this study reveal a non-numerical, symmetry-based mechanism for grouping discrimination in chicks, mirroring similar cognitive strategies observed in humans, including individuals with Autism Spectrum Disorder (ASD) who can identify prime numbers without formal training (Loconsole & Regolin, 2022). This innate ability in *Gallus gallus domesticus*, a non-mammalian species, suggests a ubiquitous cognitive mechanism in vertebrates for early detection of symmetrical and asymmetrical patterns. We can discover that human youngsters have genuinely exhibited comparable behaviour, for instance, “groupitizing” was introduced as a way to describe how children may capitalize on grouping information to facilitate enumeration processes, grouping items into subsets to speed up number configuration (Starkey & McCandliss, 2014). This would attest to non-prime, symmetric numbers, being easier for humans to leverage with cognitive reasoning. Utilising intrinsic perceptual systems may enhance mathematics learning and educational achievements, according to the findings in chicks, which also indicate ways for improving numerical thinking in people (Rugani, et al. 2020).

#### **4.4 Conclusion**

In closing, this study aimed to bridge the gap between previously known research on chick's numerosity preferences and choice mechanisms. Using a repeated measures experiment, we examined how chicks responded to sets of novel (5) and composite (9) objects, influenced by sequential exposure to habituation (2D elements) and imprinting (3D elements) conditions. Chicks initially showed a preference for the novel set, in this case 5, following habituation, which aligned with prior literature on explorative learning. The chicks' initial preference, however, decreased with extended exposure to 3D stimuli, following imprinting, resulting in no significant difference in the amount of time spent near either set. This change shows that chicks' numerical cognition is flexible and changes depending on the circumstance they are in. These results also indicate that familiarity and prolonged exposure can shift initial choices, and cognitive mechanisms underlying processing may interact to make such a change. Overall, this study offers a new perspective on how various experimental settings affect chicks' innate decision-making processes, as well as emphasises the significance of experience in forming numerical decisions, and lastly opens more ideas for further research on animal numerical processing.

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