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# **Session 1**

Wednesday 20th

15.00-17.00

# Talk 1

## A Supernormal Song Stimulus Enhances Behavioral and Neural Responses in Nightingales

Giacomo Costalunga<sup>1</sup>, Daniela Vallentin<sup>1</sup>

1. MPI Biological Intelligence

neuroethology, electrophysiology, acoustic communication, songbird, behaviour

Coined by Niko Tinbergen, a 'supernormal stimulus' indicates an exaggerated version of a natural stimulus that elicits a more intense behavioural response. Supernormal stimuli have been behaviorally investigated in several species mostly for visual stimuli, but other sensory modalities and the neural representations of supernormal stimuli are largely unknown. Here, we identified a supernormal song stimulus for nightingales and explored the neural correlates that might explain the enhanced behavioural response. Nightingales, songbirds with a repertoire of up to 200 different songs, perform sophisticated, night-long song duels against conspecifics. During these duels, they perform song-matching, copying repertoire-shared songs of their rivals. To test whether the bird's own song (BOS) can act as a supernormal song stimulus, we conducted field experiments exposing wild nightingales to playbacks simulating different opponents. We found that both temporal and spectral aspects of song duels were adjusted depending on different playbacks. Specifically, nightingales reduced song duration, took longer gaps in between songs and reduced the amount of responses overlapping with playbacks when counter-singing against their own songs. Using a deep learning approach, we found that BOS playbacks evoked more song-matching compared to conspecific song indicative of a supernormal song stimulus which intensifies preexisting singing patterns. Next, we investigated the neural substrates of song-matching behaviour of song stimuli of normal versus supernormal stimuli by performing intracellular recordings in hand raised and song-tutored nightingales. Premotor neurons in the nightingale's premotor area HVC selectively responded to the presentation of song playbacks, with enhanced responses to playbacks of the bird's own songs. Taken together, these results show how nightingales' own songs act as supernormal stimuli, eliciting behavioural responses in wild animals and evoking precise auditory integration necessary for song-matching in a premotor circuit in the nightingale's brain.

## Talk 2

### *In vivo* Investigation of Putative Neuronal Biomarkers for Recovery in a Mouse Model of Stroke

Livia Vignozzi<sup>1</sup>, Stefano Varani<sup>1</sup>, Emanuela Beretta<sup>1</sup>, Gabriele Deidda<sup>1,2</sup>, Manuela Allegra<sup>2,3,4</sup>

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behaviour, electrophysiology, stroke, optogenetics

After an ischemic event, neural circuits in the peri-infarct area undergo plastic changes aimed at the recovery of the impaired neurological function. However, the degree of spontaneous recovery varies significantly among patients. Consequently, the identification of neurophysiological biomarkers that can predict the outcome of recovery early on is crucial for effective treatment strategies in this pathology. To address this aim, we make use of a mouse model of adult stroke, the middle cerebral artery occlusion (MCAO), and study the correlation between the impaired neurological function and neuronal activity changes in the peri-infarct area.

Given that the motor cortices are perilesional to the lesion, we assessed the evolution of the motor spontaneous recovery after stroke, by performing a battery of behavioural tests, including the Gridwalk, Rotarod and Grip Strength test. Among these, only the Gridwalk test proved sensitive enough to differentiate between stroke and sham-operated mice. Moreover, to detect and discriminate finer movements of the impaired forelimb, we also explored a more specific behavioural task, the Water Grasping test.

To investigate potential correlations between behavioural impairments and neural changes in the peri-infarct area, we performed spontaneous and evoked *in vivo* local field potential (LFP) recordings in anaesthetised Thy1-ChR2-YFP transgenic mice at D9 and D30, the subacute and chronic phases of stroke, respectively. We found higher amplitude responses to intra-hemispheric photostimulation of the premotor cortex in stroke mice with good recovery outcome compared to those with low motor recovery at D30 after stroke. Furthermore, we performed preliminary longitudinal single-photon calcium imaging experiments in freely moving mice during Gridwalk and Water Grasping.

By integrating behavioural assessments with neuronal activity recordings, our study aims at providing a comprehensive understanding of the relationship between motor deficits and cortical changes after stroke.

## Talk 3

### Unraveling the link between dopaminergic functioning and social behavior in a chick model of Autism Spectrum Disorder

Alice Adiletta<sup>1</sup>, Alessandra Pross<sup>2</sup>, Nicolò Taricco<sup>1</sup>, Samantha Pedrana<sup>1</sup>, Orsola Rosa-Salva<sup>1</sup>, Paola Sgadò<sup>1</sup>

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domestic chicks, early social behaviors, autism, dopamine

Thanks to the accessibility of their embryos, domestic chicks have been traditionally exploited in the study of developmental biology. Nevertheless, as an extremely precocial species, domestic chicks could also represent an ideal model to study early spontaneous behaviors. This peculiarity has allowed researchers to define the domestic chick as an extremely social animal, equipped already after hatching with the ability to properly orient towards social features (1,2,3). This social orienting ability, largely described also in humans (4,5,6,7), is believed to rely on specific brain circuits, which do not only orient newborns' attention to social stimuli, but whose activation is fundamental for the correct development of the social brain network (8,9). Therefore, defining the aberrant conditions that underlie alterations of such preferences represent a powerful tool in further understanding social disorders such as Autism Spectrum Disorders (ASDs). In this work, we exploit the evolutionary conservation of early social behaviors to study the mechanisms that underlie social preferences in domestic chicks, investigating both typical and atypical development. Precisely, by using a pharmacological model of ASD based on the embryonic administration of Valproic Acid (VPA), we aim to observe the social behavior of these animals and to characterize its underlying dopaminergic functioning. Up to now, we demonstrated that embryonic exposure to VPA impairs chicks' innate preference for face-like configurations. Moreover, we also found that VPA administration affects the rostro-caudal distribution of dopaminergic (DA) neurons in the mesencephalon and alters the expression of genes involved in DA signaling in the septum. Taken together, the disruption of the DA neuronal distribution and its altered signaling pathway in areas belonging to the social behavior network could further support the emerging hypothesis of a prominent role of DA in social cognitive development, and provide insights into a possible link between DA dysfunctions and social cognitive deficits.

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## Talk 4

### Innovation across 13 ungulate species

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3. Zoo Leipzig
4. Zoo Nürnberg

innovation, problem-solving, neophobia, fission-fusion, social integration

Innovation is the ability to solve new problems or find new solutions to familiar problems, and it is known to provide animals with crucial fitness benefits. Although this ability has been extensively studied in some taxa, the factors that predict innovation within and across species are still largely unclear. In this study, we used a novel foraging task to test 111 individuals belonging to 13 ungulate species - a still understudied taxon. To solve the task, individuals had to open transparent and opaque cups with food rewards, by removing their cover. We assessed whether individual factors (neophobia, social integration, sex, age, rank) and socio-ecological factors (dietary breadth, fission-fusion dynamics, domestication, group size) predicted participation and performance in the task. Using a phylogenetic approach, we showed that success was higher for less neophobic ( $p < 0.001$ ) and socially less integrated individuals ( $p = 0.047$ ). Moreover, less neophobic individuals ( $p = 0.001$ ), individuals of domesticated species ( $p = 0.005$ ) and having higher fission-fusion dynamics ( $p = 0.010$ ) were more likely to participate in the task. These results are in line with recent literature suggesting a central role of sociality and personality traits to successfully deal with novel challenges, and confirm ungulates as a promising taxon to test evolutionary theories with a comparative approach.

# **Session 2**

Thursday 21st

09.00-10.40



## Talk 1

### Using temperature to analyze the neural basis of a time-based decision

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behaviour, temperature, electrophysiology, striatum, interval timing

The basal ganglia are thought to contribute to decision-making and motor control. These functions are critically dependent on timing information which can be extracted from the evolving state of neural populations in their major input structure, the striatum. However, it is debated whether striatal activity underlies latent, dynamic decision processes or kinematics of overt movement. Here, we measured the impact of temperature on striatal population activity and the behaviour of rats, and compared the observed effects to neural activity and behaviour collected in multiple versions of a temporal categorization task. We found that temperature affected overall activity levels non-monotonically, with both warming and cooling producing lower baseline firing rates. In contrast, temperature manipulations caused monotonic and graded changes in the temporal scaling of neural activity, mimicking endogenous decision-related variability in this feature of striatal dynamics. Temperature manipulations also caused bidirectional and graded changes in animals' timing judgments, mirroring the temperature-dependent modification of temporal scaling of neural activity as well as the observed relationship between temporal scaling of activity and animals' judgments. Strikingly, movement kinematics were not similarly affected by temperature. Instead, the patterns of animals' reaction times and average movement speeds were best described as non-monotonic functions of temperature, similar to the observed non-monotonic effect of temperature on baseline firing rate. Together, these data strongly suggest that the time course of evolving striatal activity dictates the speed of a latent process that is used by rats to determine when to do what action, but that this feature of neural activity is not what determines the moment by moment kinematics of movement execution. Instead, the striatum may additionally provide a lower dimensional gain signal coded in the firing rate of neurons that controls overall vigour. More broadly, they establish temporal scaling of population activity as a likely neural basis for variability in timing behaviour.

## Talk 2

# Acute and Chronic Low Oxygen Compromise The Survival and Cognition of a Coastal Cephalopod

Mélanie Court<sup>1</sup>, Marta Macau<sup>1</sup>, Tiago Repolho<sup>1</sup>, Vanessa Madeira Lopes<sup>1</sup>, Rui Rosa<sup>1</sup>, José Ricardo Paula<sup>1</sup>

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hypoxia, deoxygenation, behaviour, cuttlefish, *Sepia officinalis*

The ocean is undergoing deoxygenation and the spread of hypoxic areas. Ocean deoxygenation and standing levels of hypoxia are shrinking fundamental niches, particularly in coastal areas, yet documented repercussions on species development and behaviour are limited. Here, we tackled the impacts of deoxygenation (7 mg O<sub>2</sub> L<sup>-1</sup>), mild hypoxia (nocturnal 5 mg O<sub>2</sub> L<sup>-1</sup>), and severe hypoxia (2 mg O<sub>2</sub> L<sup>-1</sup>) on cuttlefish (*Sepia officinalis*) development (hatching success, development time, mantle length) and behaviour, i.e., ability to learn (individual- and socially), to camouflage, and to explore its surroundings spatially. We found that hypoxia yielded lower survival rates, smaller body sizes and inhibited predatory (increased latency to attack the prey) and antipredator (camouflage) behaviours. Acute and chronic exposure to low oxygen produced similar effects on cognition (inability to socially learn, increased open-field activity levels, no changes in thigmotaxis). It is thus expected that, although cuttlefish can withstand oxygen limitation to a certain degree, expanding hypoxic zones will diminish current habitat suitability.

## Talk 3

### Does social hierarchy affect executive function abilities and brain morphology?

Angelo Guadagno<sup>1</sup>, Zegni Triki<sup>1</sup>

1. University of Bern

social complexity, flexible learning, inhibitory control, working memory, fish

We know from comparative studies, but also from research on inter-individual variation, that the social environment, like group size, to some extent, affects brain complexity, such as brain size and the size of brain areas (the Social Brain Hypothesis). However, there is limited knowledge about how other social factors, such as hierarchy and roles, may affect brain and cognitive abilities. To explore this, we investigated whether reproductive dominants have any cognitive and brain advantages compared to helper subordinates. To test our hypothesis, we evaluated the cognitive capacities of breeders and helpers of a highly social fish species, the African cichlid *Neolamprologus pulcher*. We tested the fish in reversal learning, detour task and object permanence tests to evaluate cognitive flexibility, inhibitory-control and working memory abilities, respectively. Afterwards, we sampled their brains for brain morphology analyses. We also controlled for potential sex differences by testing both males and females. Our preliminary findings show that fish differently perform during the same task and across different tasks, suggesting both individual and cognitive capacity differences. However, data analyses are currently still in progress and hopefully we will present our findings at the ESCBC meeting. Together, our study will help understand better how social roles may contribute to the social brain hypothesis and whether different roles have distinct cognitive advantages.

## Talk 4

### Interspecific competition between the Great spotted woodpecker (*Dendrocopos major*) and the Syrian woodpecker (*Dendrocopos syriacus*)

Alena Fišerová<sup>1</sup>, Michaela Syrová<sup>1</sup>, Ladislava Krausová<sup>1</sup>, Kateřina Antonová<sup>2</sup>, Ondřej Fišer<sup>1</sup>, Jakub Ondruch<sup>1</sup>, Jan Špička<sup>1</sup>, Petr Veselý<sup>1</sup>

1. University of South Bohemia

2. Charles University

interspecific interactions, competition, playback experiment, behaviour ecology, aggression

The Great Spotted Woodpecker (GW) is a common species throughout Europe inhabiting all forest types while the Syrian Woodpecker (SW) is common in south-eastern Europe, preferring river margins, orchards and urban parks. In Czech Republic, these two species recently encounter, while in Finland, an allopatric population of GW unfamiliar with SW occurs. Our aim was to find out how GW and SW are able to coexist in Czech Republic and what are the responses of allopatric GW to SW as a potential unfamiliar competitor. Both species are strongly territorial intraspecifically and may compete even interspecifically, as there is a presumed niche overlap. At the beginning of their breeding period (March and April 2022 and 2023), we identified particular territories of both species and subsequently conducted a dummy experiment in the centre of each territory. A mount of particular species associated with a playback was presented to the focal birds and several behavioural responses were recorded. The observed behaviours were scored using software BORIS and subsequently analysed using multivariate analysis (Principal Component Analyses). We obtained canonical scores describing two axes. First axis was loaded by active behaviour like attacking and flying over the mount, the second axis was loaded mainly by the distance, where the tested individual spent most of the time. We showed that both species are territorial especially intraspecifically, but SW also showed quite high aggression towards GW. Allopatric GW population from Finland showed also quite increased aggression towards SW compared to sympatric population. Our results suggest that 1) there is a certain level of competition between SW and GW, 2) that this aggression is asymmetrical as GW is more tolerant towards SW than vice versa, and 3) that allopatric population has a certain level of precaution towards unfamiliar, though similar species.

## Talk 5

# Probing the mechanistic implementation of temporal control using behavior

Sofia Freitas<sup>1</sup>, Alfonso Renart<sup>1</sup>, Joseph J. Paton<sup>1</sup>

1. Champalimaud Centre for the Unknown

mechanistic description, temporal control, timing, vigor, behavior regularities

Animals extract temporal structure from the environment and use it to anticipate, plan, and adjust their behavior toward maximum benefit while mitigating costs. Despite the fundamental importance of temporal control in behavior, we lack a principled framework for understanding how the multiple features that characterize the temporal control relate to each other and the environment. How do we determine when to engage in a behavior, and with what intensity?

To address these questions, we trained rats using an operant lever pressing paradigm under fixed interval (FI) schedules of reinforcement. Unlike previous studies, we systematically varied FI, reward rate, and the presence or absence of a stimulus that signals FI termination.

The animals' behavior follows temporal regularities. In particular, rats display a characteristic pattern of behavior within a trial, transitioning from low to high rates of pressing and maintaining a constant rate thereafter. Interestingly, the dynamics of the transition and the characteristic high pressing rate change selectively according to the experimental variables, suggestive of distinct mechanisms operating at different temporal scales.

Our behavioral results serve as constraints for the development of a computational framework to explain rats' behavior. Our goal is to specify a mechanism for determining when to transition to high rates of responding and with what frequency to respond as a function of reward, FI duration, and the amount of information available regarding FI termination. Overall, our work represents an important step towards a more comprehensive and principled understanding of temporal control.

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## **Session 3**

Thursday 21st

11.00-13.00

## Talk 1

### The Neural Control of Gait Switching in Larval Zebrafish.

Elena M. D. Collins<sup>1</sup>, Edite Figueiras<sup>1</sup>, Alexandre Laborde<sup>1</sup>, Adrien Jouary<sup>1</sup>, Aaron Ostrovsky<sup>1</sup>, Michael B. Orger<sup>1</sup>

1. Champalimaud Neuroscience Programme

Animals need to perform a diverse range of behaviours to navigate their environment successfully. Larval zebrafish swim using discrete episodes of propulsion called bouts that can be classified into 13 categories of movement. We aim to elucidate the mechanism of transition between different movements, particularly in the context of forward swimming, on both a behavioural and neural level. To characterise gait-switching behaviour, larval zebrafish were presented with gratings moving from tail to head at different speeds eliciting forward swimming, known as the optomotor response. Collecting various kinematic parameters, through tracking fish position and tail angle, allowed swims to be classified into different categories of movement. In response to slow gratings, larval zebrafish predominantly exhibited slow swims, whereas fast gratings elicited a rapid transition from slow to sustained trains of fast swims. To identify the neural correlates of slow and fast swims and elucidate the population dynamics underlying gait transitions, we recorded activity from genetically labelled neural populations in the brainstem of head-fixed larval zebrafish while they performed the optomotor response in a closed-loop configuration, utilising a 'SCAPE' light-sheet microscope. Head-fixed fish showed differences in bout kinematics from freely swimming fish, with longer movements that included switches in frequency within a single bout. We therefore use regression analysis, based on kinematics of individual half-beats within each bout to identify neuronal populations associated with different modes of swimming. By showing how the brain selects dynamically between two distinct motor outputs we strive to understand fundamental principles in the supra-spinal control of locomotion.

## Talk 2

# A theory of magnitude: Insights from honeybees

Maria Bortot<sup>1</sup>, Giorgio Vallortigara<sup>1</sup>

1. CIMeC University of Trento

honeybees, ATOM theory, insect cognition, quantity representation, magnitude processing

The capacity to process discrete and continuous dimensions of a stimulus (e.g., numerosness, size, temporal duration) is thought to be supported by a general cognitive system estimating quantities irrespective of their dimension (e.g., number, space, time) and format (i.e., discrete, continuous). Studies in humans and other vertebrates have demonstrated the existence of symmetrical interactions between dimensions, supporting this theory (i.e., A Theory Of Magnitude, ATOM).

Honeybees process both continuous (e.g., relative size of visual elements) or discrete quantities (i.e., numerosness of an array of visual stimuli). However, whether this ability is supported by an underlying magnitude system is unknown. We investigated whether bees could transfer a quantity discrimination (i.e., to choose the larger/smaller stimulus) between a discrete (i.e., number) and a continuous (i.e., size) dimension. In a first experiment, we trained free-flying foragers to discriminate between different numerical comparisons having either a 0.5 (2 vs. 4; 4 vs. 8) or 0.67 ratio (2 vs. 3; 4 vs. 6). Then bees were tested for spontaneous choice using comparisons with identical numbers but different sizes. Irrespective of the ratio of stimuli, bees trained to select the smaller numerical quantity chose the congruent smaller size, and bees trained to select the larger numerical quantity chose the congruent larger size. In a second study, we trained bee foragers to discriminate between dimensionally larger and smaller elements with the same numerosness. At test, bees were presented with a comparison between relatively larger and smaller numerosities controlled for covarying continuous variables (i.e., total area, total perimeter, convex hull, element size). Results showed that bees generalised from the size to the numerical dimension, providing evidence of a bidirectional property of the association. The presence of this cross-dimensional transfer supports the hypothesis of a universal mechanism for the encoding of abstract magnitudes in vertebrate and invertebrate species.

## Talk 3

### Problem Solving and Physical Cognition in Two Passerine Species



Anita Szabo<sup>1</sup>, Alice Exnerova<sup>1</sup>

1. Charles University, Prague

behaviour, avian cognition, problem solving

In recent years, avian cognition has become a subject of intensive research. Most studies are, nevertheless, focused on cognitive abilities of a few bird taxa known for their high cognitive capacities, especially parrots and corvids, and comparative studies of other avian taxa are still scarce. Lever-pulling and the trap tube task are well-known paradigms to investigate problem solving and physical cognition in various species. The trap tube task was originally designed for tool-using species, as subjects had to insert a tool to reach the reward. However, with a modified setup – the tool pre-inserted - it is possible to test the species that do not use tools in the wild. Therefore, we can compare a wider diversity of avian taxa and ascertain if there is a link between tool use and the ability to understand physical properties of objects. As tits (*Paridae*) are known for their innovative behaviour and occasional tool use, they represent promising candidates for problem solving tasks. We tested wild-caught adult great tits (*Parus major*) and blue tits (*Cyanistes caeruleus*) as well as hand-reared juveniles of both species in lever-pulling and trap tube tasks. Both species and age categories of birds performed similarly well in the lever-pulling task. We found species-specific differences in performance of birds in the trap tube task, with only blue tits solving it successfully. Great tits were more cooperative but less successful compared to blue tits, irrespective of age and rearing conditions. To our knowledge this is the first study testing small passerine species in the trap tube paradigm, and we hope that future research will include a wider variety of species, which will allow for a better understanding of avian cognition.

## **Talk 4**

### **Signatures of Control-Limited Perceptual Decision Making in Freely Moving Rats**

Juan R. Castiñeiras<sup>1</sup>, Alfonso Renart<sup>1</sup>

## 1. Champalimaud Foundation

perceptual decision making, reaction time, normative models, optimal control, behaviour

Periods of disengagement with associated poor performance are generally observed during perceptual decision-making tasks, but a normative understanding is lacking. Here, we develop a theory that frames disengagement as a problem in cognitive control[1]. Good performance through task engagement requires control, which is costly, establishing a performance-control tradeoff. We derive decision policies that optimise this tradeoff as a function of the agent's cognitive control capacity. When it is sufficiently low, agents lapse. For intermediate control limitations, a new decision-making regime appears where agents do not lapse, but their behaviour is still shaped by control. We identify hidden signatures of control-limited behaviour at the level of accuracy, reaction time and decision confidence which are often observed experimentally, but had not been normatively explained[2].

In particular, we leveraged a specific yet simple prediction of our theory: control-limited agents should display, on average, faster errors than correct responses, while for agents with high control capacity, the pattern reverses and correct responses become faster than errors. To test this, we employed a sound lateralization task with freely moving rats developed in the laboratory[3], adapting it to match closely the statistical assumptions of the theory.

We reasoned that if the animals are sensitive to the available reward rate of the environment, they would be willing to mobilise more control when the reward rate is low, as this context incentivizes good performance, while they would be satisfied with lower levels of control when the reward rate is high[4]. We implemented this by manipulating the inter-trial intervals and error time-penalties in the task and making them either long or short, therefore controlling the maximum reward rate achievable. Preliminary results indicate that the rats respond to this manipulation, and so far the trends in the reaction times are compatible with the theoretical predictions.

1. Castiñeiras, J. R., & Renart, A. (2022). Control limited perceptual decision making. *bioRxiv*, 2022–06. 2. Pleskac, T. J., & Busemeyer, J. R. (2010). Two-stage dynamic signal detection: a theory of choice, decision time, and confidence. *Psychological review*, 117 (3), 864. 3. Pardo-Vazquez, J. L., Castiñeiras-de Saa, J. R., Valente, M., Damião, I., Costa, T., Vicente, M. I., ... & Renart, A. (2019). The mechanistic foundation of Weber's law. *Nature neuroscience*, 22(9), 1493-1502. 4. Simon, H. A., et al. (1990). Invariants of human behavior. *Annual review of psychology*, 41(1), 1–20.

## Talk 5

### Beware of My Face: The Role of Facial Configuration in Predator Recognition

Kateřina Antonová<sup>1</sup>, Ondřej Fiřer<sup>2</sup>, Petr Veselý<sup>2</sup>, Michaela Syrová<sup>2</sup>, Roman Fuchs<sup>2</sup>

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inversion effect, holistic processing, antipredator behaviour, predator-prey interactions, great tit

The important role of facial elements (hooked beak and conspicuous eye colour) in the recognition of avian predators has been repeatedly demonstrated. However, no attention has yet been paid to the importance of their canonical configuration i.e., the relative position of the eyes and beak. In our study, we tested the ability of untrained wild tits to recognise a dangerous predator (a sparrowhawk) with inverted eye and beak positions (invert dummy) and with one eye above and one below the beak (inline dummy) in aviary experiments. A dummy of a sparrowhawk with its head devoid of eyes and beak (empty dummy) served as a baseline alongside dummies of an unmodified sparrowhawk and a pigeon (as a harmless control). The experiment was carried out in two variants, with the complete dummy of the sparrowhawk visible and with the body of the dummy hidden in the bushes, from which only the head was visible. The tits showed no more fear towards the eyeless and beakless dummy than they did towards the pigeon. Towards the dummy with the modified configuration, the tits showed no less fear than towards the unmodified sparrowhawk, but in the case of the variant with an eye above and below the beak, their behaviour was different and can be interpreted as increased fear but also as surprise. Thus, the tits perceive a disturbance in the configuration of the predator faces, but do not show unambiguous signs of holistic processing i.e., impaired recognition of the inverted face.

# Session 4

Friday 22nd

11.00-13.00

## Talk 1

Problem-solving through individual cognition in invasive social insects

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confocal microscopy, decision-making, experience, patterns, supercolony

Biological invasions currently pose one of the major threats to ecology and economics, but they also offer valuable insights into evolutionary processes in a short time because species must adapt to solve new problems in the introduced environment. Problem-solving is usually addressed by behavioural plasticity, but it was proposed that in social species such as ants, the costs of individual plasticity might outweigh its benefits. We analysed behavioural and neuroanatomical variability in one of the most widespread invasive ant species, the Argentine ant (*Linepithema humile*), to unravel whether individual behavioural differences might be sufficient to reach colony-level solutions or exposure to problems is the factor shaping individual behaviour and brain. We exposed 173 age-controlled callow workers from five different colonies to sets of three behavioural tests conducted every day over five days. We tested ants' exploration in an empty arena, reaction towards a neophobic element (encounter with an unfamiliar object) and maze-solving ability. The maze was composed of six bilateral choices branching one after the other in a chiral manner. Repeated behavioural testing allowed us to analyse personality, predictability, repeatability, memory and learning. The brains were extracted with a fixed protocol; subjected to antigen staining to focus on the micro glomeruli density of the Mushroom Bodies calix and then correlated with the behavioural variables. Initial results suggest callow workers' personalities when considering exploration, time spent moving, and use of the central part of the arena (most vulnerable). Surprisingly, our results showed that the individuals' explorative value, the use of the new item, and the number of errors avoided in maze solving increased with each successive test with the time spent on exploration remaining constant. Our results suggest that ants can use new resources and solve new problems without modifying their behavioural patterns. Future analyses will unravel how *Linepithema humile* behaviour is linked to neuronal development.

## Talk 2

### The hybrid delay task reveals self-control individual variability in bumblebees (*bombus terrestris*)

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behaviour, learning

Self-control (i.e., the ability to control impulsive behavior in a tempting situation) can be beneficial in a variety of contexts, as this ability allows goal-directed behaviors and optimal foraging. However, choosing to wait is not necessarily paired with the ability to maintain this choice. We developed a hybrid delay task paradigm combining delay choice and delay maintenance to investigate self-control in bumblebees. bees were first individually trained in a foraging arena to associate two differently colored artificial flowers either with an immediately accessible small reward or a delayed-large reward (15s delay). bumblebees that reached the learning criterion were then offered 10 delay choices between the two flowers/rewards. in this delay choice test, once the choice was made by the bees the decision could not be reversed. Finally, in the delay maintenance test we removed the 15s delay limit and recorded the time spent by individuals on the delayed-large reward option before they defected for the smaller one. In a final test, we investigated whether bee's self-control capacity correlated with their ability for reverse learning. To this end, bumblebees were tested to see if and how fast they could modify a previously learned association between two elements (flower color and reward). Our results show that some bumblebees can choose to wait for a better reward and then maintain their choice. However, this ability does not appear to help individuals to solve the reversal learning task, and thus cope with the transient uncertainty of resource availability when foraging.

## Talk 3

### Early Life Sociality and Social Learning in Piglets (*Sus scrofa*)

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social cognition, pigs, learning, early life development, social behaviour

Young animals need to learn quickly in order to survive, and therefore need to develop social cognitive skills early on. Young learn from their mother and peers, but in many species information about early life social skills is limited. The aim of this study was to look at the relationship between social interactions of piglets towards their mother (as measure of sociality) and social learning. Piglets need to learn where to find food within the first weeks of life, and mostly learn this from the sow. We observed 29 domestic

piglets for their interactions with the sow within the first 3 weeks of life (48h of continuous observation per piglet, across 6 days), as well as in a 3-minute social foraging test. In the foraging test, a sow with one piglet was placed in an arena with two different food items that were known to her but unknown to the piglet. Data were analysed in generalised linear mixed models to account for sow effects. Social behaviours towards the sow in the first 3 weeks of life were summed to give a measure of sociality. Nose-to-nose contact was the most common behaviour (38% of all piglet-to-sow behaviour). During the foraging test, piglets interacted with the sow  $12.8 \pm 4.82$  times (mean  $\pm$  SD) and equally explored food items ( $14.7 \pm 9.72$  times). Piglet-to-sow behaviour in the foraging test was unrelated to the piglet's sociality ( $p=0.85$ ), sex ( $p=0.89$ ) or body weight ( $p=0.93$ ). Similarly, the frequency in which a piglet explored the food was unrelated to the sow's feeding behaviour ( $p=0.91$ ) and the piglet's sociality ( $p=0.63$ ), sex ( $p=0.61$ ) or body weight ( $p=0.65$ ). In conclusion, piglet's sociality towards their mother did not influence their social and food directed behaviour in a foraging test. This suggests that social learning occurs irrespective of the frequency of social contact in other contexts.

## Talk 4

### The Role of Auditory Cortex in a Sound Lateralisation Task

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1. Champalimaud Foundation

behaviour, decision-making, lateralisation, optogenetics, auditory

Our lab recently proposed a mechanistic explanation for Weber's Law through a mathematical regularity relating choice and reaction time (RT) during performance of a sound lateralisation task. Through this regularity and a mathematical model based on bounded accumulation of evidence - and strongly constrained by these experiments - we are able to accurately describe the animals' behaviour [1] and investigate the necessary computations for perceptual decisions. This model also allows for the simulation of the effect of manipulations on its different functional elements. In the current work, we focused on the role of the auditory cortex (ACx). Lesions in ACx did not lead to changes in performance. We then performed optogenetic silencing of the ACx with StGtACR2. This opsin was expressed in ACx and blue LEDs were implanted bilaterally with cranial windows. Results show that bilateral inactivation of

the ACx during the RT severely disrupts the animals' behaviour (from ~80% correct in LED-OFF trials to ~55% in LED-ON trials; n=9). RTs were also strongly affected, being much shorter for LED-ON trials. Inspired by our computational modeling, and in order to explore a more restricted perturbation, we performed brief (30ms) bilateral silencing of the ACx. This brief silencing still has an effect on performance, but the effect on RT is variable across rats. To investigate whether the observed impairments are specific to auditory processing, we developed a visual brightness discrimination task and are currently silencing the ACx during this task. Our results so far suggest that ACx is important for discriminating the lateralisation of sound.

1. Pardo-Vazquez, J. L., Castiñeiras-de Saa, J. R., Valente, M., Damião, I., Costa, T., Vicente, M. I., ... & Renart, A. (2019). The mechanistic foundation of Weber's law. *Nature neuroscience*, 22(9), 1493-1502.

## Talk 5

### Nestmate Corpses Act as Negative Stimuli for Ants: Implications for Ant Behaviour and Control Strategies

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behaviour, memory, negative stimulus, invasive ants, pest management,

Invasive ants like *Linepithema humile* (the Argentine ant) pose a significant threat to ecosystems and economies worldwide, making it imperative to understand their behaviour and find effective management strategies. However, traditional eradication methods such as baiting have had limited success due to low bait consumption rates over time, driven in part by the abandonment of toxic baits. We propose that ants learn to avoid the odour of toxic baits because they are associated with the corpses of ants which fed on them. We investigated the effect of corpse-scent association on the odour and flavour preferences of ants by using scented corpses. Ants were tested in a Y-maze or a dual-choice feeder after exposure to scented corpses or dummies. When focal ants encountered scented freeze-killed ants, 69% (n = 64) avoided a branch of a Y-maze bearing the same scent. At a collective level, when colonies were given access to two differently scented food sources via a path laden with scented ant corpses, the matching



food source was neglected compared to an alternative food source. However, ants exposed to scented corpses did not feed for less time on food scented to match the corpses compared to a control food in a dual-choice feeder. This study demonstrated that nestmate corpses act as a negative stimulus for *Linepithema humile* ants, leading to avoidance of odours associated with the corpses and potential avoidance of toxic bait at both individual and collective levels, but not a reduction in feeding once the food source is encountered. Nonetheless, this study demonstrates that conspecific corpses act as a negative stimulus for ants, and this should be kept in mind when planning control efforts.