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SA01

The Predictive Coding of Voluntary Self-Motion: Vestibular Circuits for Action and Perception

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Integrating sensory with motor signals during voluntary behavior is essential for distinguishing stimuli that are a consequence of intended actions from those that are externally generated. This ability enables the brain to flexibly fine-tune motor actions based on sensory feedback, a computation necessary for subjective awareness of the effects of movements. The lecture will explore the neural circuits that perform this computation with a focus on vestibular pathways and highlighting the cerebellum's role in building predictive models of self-generated movement as individuals explore the world.

SA02

Manipulation of neuronal activity in a genetically diverse context

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Animals exhibit significant variation in their behavior among individuals. Several factors contribute to this variability, including genetic variation that impact the development and function of the nervous system. Therefore, to understand the neuronal circuits that influence behavior in a natural context, it is necessary to manipulate neuronal activity within a genetically diverse context. Here, we present an approach that combines genetic diversity with transgenic tools to manipulate neuronal activity and measure its behavioral effects.

We used Drosophila melanogaster, a model organism with an extensive genetic tool set to manipulate neuronal activity and a variety of wild-derived lines with genetic variation amongst themselves. To obtain animals with different genetic backgrounds in a versatile and scalable process, we used heterozygote animals as test subjects. Half of the genome of these animals derive from a standardized genetic background, where the necessary genetics tools to manipulate neuronal activity are used. The other half derives from different genetic backgrounds of the Drosophila genetic reference panel (DGRP). This allowed us to survey the effects of genetic variants that act as dominant alleles.

We focused on the flies' response to a looming stimulus, which simulates an approaching object and typically triggers flight or freezing responses. Control flies, with no neuronal manipulation, from 14 different genetic backgrounds (~200 flies per genetic background), exhibited genetically dependent variation in their responses, validating the use of heterozygote animals. It has been previously described that the descending neuron DNp09 impacts freezing behavior. We inhibited DNp09 in the same 14 genetic backgrounds with Tetanus toxin (TNT, ~200 flies per genetic background). The targeting of TNT expression in this single pair of neurons was maintained in all tested genetic backgrounds. Behavioral assays show that inhibition of DNp09 reduces the probability of entering the freezing state, independently of genetic background. The probability of breaking from freezing is reduced in all genetic backgrounds, but in this case, dependent on the genetic background. These results suggest that DNp09 is an essential node in the neuronal network to maintain the freezing state in a natural context, but that freezing entry may be compensated by other neurons in the network.

SA03

Visually guided navigation in early primates

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Vision, the key sensory modality for primates, plays an important role in navigation. In the case of nocturnal primates, such as the mouse lemurs, the visual system has evolved to adapt to low-light environments. Despite the darkness, mouse lemurs navigate through dense forest by leaping across tree branches. This complex behavior heavily relies on visual guidance to estimate the distance and landing target. How mouse lemurs perform these incredible jumps in darkness, and how their ability to do so is affected by other environmental factors, remains an open question. To investigate this matter we designed a novel jumping framework for mouse lemurs using the latest version of the EthoLoop system (www.etholoop.org). EthoLoop is an optical animal tracking system designed for small animals, not only to track their spatial position (~800Hz), but also to provide continuous close-up views while freely roaming in their natural habitat. Taking advantage of the real-time position tracking we control multiple wireless feeding platforms and are able to reinforce behavior, such as jumping in a reproducible manner. This provides a rich dataset of the kinematics of individual jumps, enabling comparison across sessions with different illumination settings. In parallel, we have taken the first step to study the navigation of freely moving mouse lemurs in their natural habitat. Here we will present our preliminary result using the battery powered version of the EthoLoop system in the heart of Kirindy forest of western Madagascar.

SA04

Studying whisker movements to gain insights into the natural sensory behaviours of mammals.

Robyn Grant[1]

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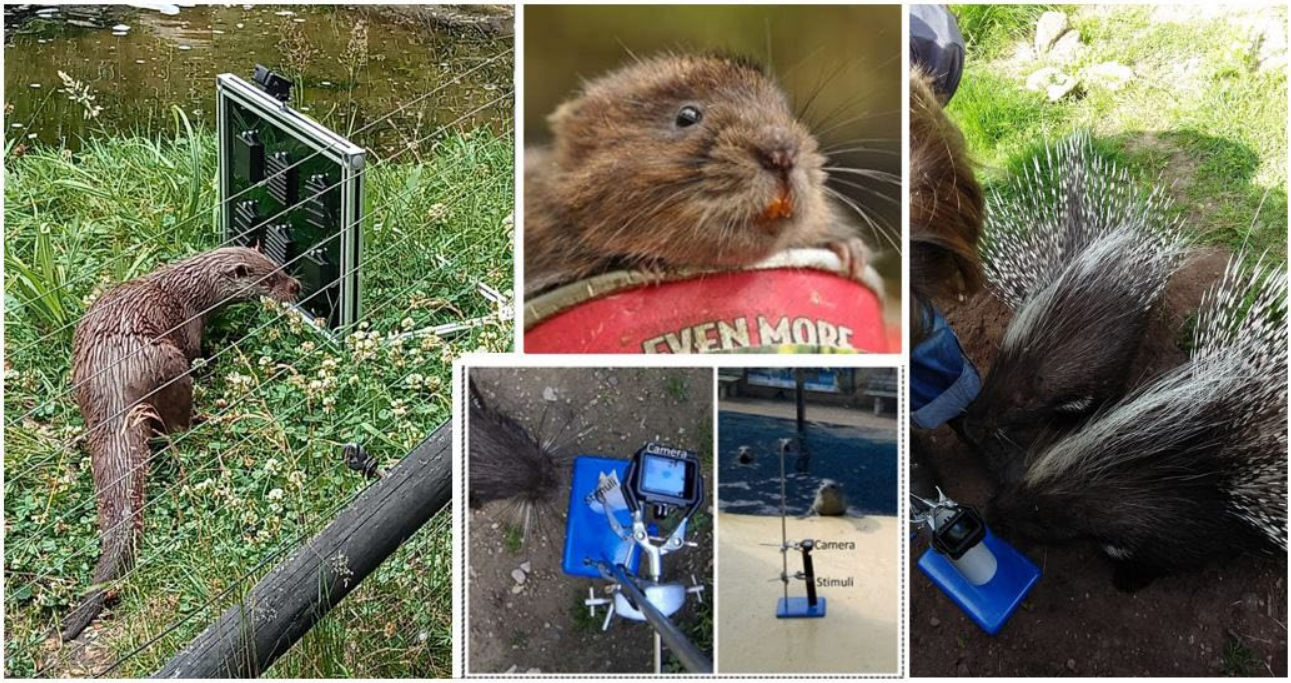
Nearly all mammals have whiskers – sensory tactile hairs, also known as vibrissae. In fact, whiskers are only truly absent in a handful of species, including humans. However, much of what we know about whiskers comes from studying just a few species, such as rats and mice, and often in quite constrained laboratory settings. My research focusses on describing whisker movements and behaviours in many different species while the animals are freely moving and taking part in naturalistic whisker exploration tasks. In this presentation I will showcase some of the tasks that I have designed for laboratory and zoo studies to examine whisker movements during whisker-object exploration and whisker-guided locomotion. All studies have been ethically approved by the committee at Manchester Metropolitan University, and the local panels of each collaborating lab and zoo institution. I will describe commonly observed behaviours, such as contact-induced asymmetry, head-turning asymmetry and look-ahead, and reflect on how these might be associated with attention. I will also identify some species-specific behaviours, such as spread reduction and task-specific behaviours that I have observed in rodents and pinnipeds, respectively. I will go on to reflect on my current methods and demonstrate new protocols that I am developing in the field. Overall, studying whiskers comparatively can give us important insights into mammalian health, welfare, ecology and evolution.



SA05

State-dependent neural processing of dark flash stimuli in the larval zebrafish

Charles Heller[1]

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Sensory computations evolve as a function of internal state: are we hungry or satiated, sleepy or alert? To investigate how the brain implements this flexible processing of sensory information, we chose to study the larval zebrafish because of its well-defined behavioral repertoire and its optical accessibility for brain imaging during natural behavior. In this study, we recorded brain-wide activity at single cell resolution in freely moving fish while simultaneously presenting whole-field, dark flash visual stimuli. Behaviorally, dark flashes elicited high amplitude turn responses in awake, alert fish. Notably, these responses were abolished during quiescence. Neurally, dark flashes drove robust visual responses in single neurons spanning multiple brain regions. Despite the behavioral variability, neural responses to the stimulus remained stable across waking and quiescent states. However, we found that the inferred functional connectivity between dark flash responsive neurons and their downstream, motor neuron targets was suppressed by quiescence. This led us to hypothesize that changes in inter-brain region connectivity might underlie the gating of behavioral responses. To investigate this further, we used Reduced Rank Regression to identify the dimension of visual neuron activity that was most predictive of activity in downstream motor neurons. We discovered that the weights defining this dimension were highly dependent on internal state. Specifically, the projection of dark flash evoked activity onto these weights changed significantly between quiescent and non-quiescent trials, despite the visually evoked response being stable. Thus, state-dependent changes in connectivity weights between brain regions may underlie sensorimotor gating of an innate, natural behavior in the zebrafish.

SA06

Foraging as a lens onto dopamine signalling at different timescales

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A fundamental requirement for all organisms is to determine how to adapt to changes in their environment or internal needs. Dopamine has long been linked with regulating adaptive decisions through its implication in reinforcement learning, yet there is still little consensus as to how these signals guide adaptive decisions. One potential reason for this is that, while laboratory studies have commonly focused on situations where animals are required to learn and compare the values of simultaneously presented options, as happens during reversal learning or 'bandit' tasks, in more naturalistic foraging settings, potential sources of reward are more often encountered sequentially. A key computation is then how long to persist in working for reward in the current location and when to switch to an alternative based on estimates of the expected future reward rate in the current location against the average reward rate in the broader environment. Importantly, increasing evidence suggests dopamine represents reward information over different timescales, making it potentially ideally placed to regulate such foraging decisions. I'll describe how our ongoing work, combining techniques to monitor and manipulate dopamine over prolonged periods in freely-behaving animals performing foraging tasks, is helping us unravel how dopamine signalling at different timescales helps promote efficient adaptive foraging.

SA07

Identifying Neural Mechanisms for Natural Behavior Through Computational Ethology

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The brain allows animals to successfully interact with the world through behavior. But how does the brain compose natural behaviors — the kinds of behaviors that are expressed by animals when they are unrestrained and free to act based upon their own motivations? And how do sex, age, internal state, individual identity coalesce into a context-appropriate pattern of behavior at any given moment? Here I describe recent developments in computational ethology, and highlight how these emerging approaches can shed new light on how the brain endows natural behavior with meaning.

SA08

From goals to actions: neural circuits for instinctive navigation

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When faced with predatory threats, escape towards shelter is an adaptive action that offers long-term protection against the attacker. Animals rely on knowledge of safe locations in the environment to instinctively execute rapid shelter-directed escape actions. Although previous work has identified neural mechanisms of escape initiation, it is not known how the escape circuit incorporates spatial information to execute rapid flights along the most efficient route to shelter. In my talk I will show that the mouse retrosplenial cortex (RSP) and superior colliculus (SC) form a circuit that encodes the shelter-direction vector and is specifically required for accurately orienting to shelter during escape. Shelter direction is encoded in RSP and SC neurons in egocentric coordinates and SC shelter-direction tuning depends on RSP activity. Inactivation of the RSP–SC pathway disrupts the orientation to shelter and causes escapes away from the optimal shelter-directed route, but does not lead to generic deficits in orientation or spatial navigation. We find that the RSP and SC are monosynaptically connected and form a feedforward lateral inhibition microcircuit that strongly drives the inhibitory collicular network because of higher RSP input convergence and synaptic integration efficiency in inhibitory SC neurons. This results in broad shelter-direction tuning in inhibitory SC neurons and sharply tuned excitatory SC neurons. These findings are recapitulated by a biologically constrained spiking network model in which RSP input to the local SC recurrent ring architecture generates a circular shelter-direction map. We propose that this RSP–SC circuit might be specialized for generating collicular representations of memorized spatial goals that are readily accessible to the motor system during escape, or more broadly, during navigation when the goal must be reached as fast as possible.

SA09

Flexibility of aversive behaviours and circuits

Katja Reinhard[1,2,3], Lorenza Tortella[1], Rani Vastenavondt[2], Benjamin Crevits[2], Sybren De Boever[2], Po-Yu Liao[1], Lucia Zanetti[1], Karl Farrow[2,3]

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Avoiding danger and approaching food or mates are one of the most essential and conserved set of behaviours, observed in most species from crabs to primates. Nevertheless, in an ever-changing environment, the type and kinetics of these innate behaviours need to be flexible and context-specific to optimize an animal's survival. However, it remains unclear to which degree innate behaviours can be influenced by the environment of an animal; as well as how brain circuits are organized to allow reliable and fast, yet flexible and context-specific behaviours. We are addressing these questions by studying the effect of ambient light on innate reactions of rodents. In nature, animals will encounter predators and prey under a variety of different light conditions, which have intrinsically distinct levels of danger and opportunity. However, most studies are performed under daylight conditions, and it is unknown how different light conditions affect predator avoidance and prey capture behaviour.

To induce innate avoidance and approach in freely moving animals, we presented standardized visual stimuli mimicking predators and prey on the top and side monitors of a behavioural box with a Plexiglas shelter in one corner. Rodents were filmed during those experiments and behaviour type (skittish darts, escape, running, stopping, freezing) as well as their kinetics were analysed offline. While similar paradigms are used by many different research groups, it remains unclear what parameters enable fast and reliable data collection while preventing habituation of the animals to the behaviour-eliciting stimuli. We hence set out to first test various combinations of session intervals and shelter locations, and their effect on visually induced behaviour in Mus musculus. We found that shelter location impacted session duration since mice spent more time foraging if the shelter was placed opposite of the setup entrance, providing more opportunities for stimulus presentations. Furthermore, while the time between early session (3 or 7 days) had little effect on behaviour, short time intervals led to switches between escaping and stopping.

We then used the same experimental paradigm with a 7-day session-interval to test the effect of ambient light on innate behaviour of American (Peromyscus) and Eurasian (Mus) rodent species. We found general and species-specific adaptation to different ambient light levels. For example, a looming stimulus mimicking an attacking predator induced a faster onset and more vigorous reaction in all species under moonlight than under brighter light conditions. For cricket-mimicking stimuli and non-threat stimuli, on the other hand, different species changed their behaviour differently depending on the light level. In addition, pre-stimulus behaviour could predict some reaction to threat, but only in a light-specific manner. Taken together, we found a strong and species-specific effect of ambient light on various innate reactions, suggesting that successful context-specific avoidance and hunting strategies are passed on to offspring through inheritance rather than learning. Finally, ongoing Neuropixels recordings of the underlying behaviour-driving circuits through the superior colliculus suggest ambient light specific encoding of ethologically relevant visual stimuli.

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SA10

Unraveling multiscale plasticity and variability in behavior across species

Antonio Carlos Costa[1]

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Animals chain movements into long-lived motor strategies, exhibiting variability across scales that reflects the interplay between internal states and environmental cues. Understanding how these complex patterns in behavior emerge requires a comprehensive approach that embraces variability across scales. We build high-fidelity Markov models from maximally-predictive sequences, bridging across scales from posture dynamics to long-lived navigation states across species. In C. elegans and larval zebrafish we find that behavioral plasticity leads to heavy-tailed statistics in behavior. Additionally, we introduce new approaches for measuring phenotypic variation across behavioral timescales, and find structure to inter-fish variability that reveals how sensory inputs and motivational states drive behavioral variation along an exploration-exploitation axis.

SA11

Body state encoding in primary and secondary somatosensory cortex of freely moving mice

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Sensory processing has conventionally been studied on restrained or anaesthetised animals. However, in natural settings, sensory perception is not as controlled or restricted; instead, it is a dynamic process involving continuous interactions between an animal's movements and sensory inputs. This process of active sensing and sensorimotor integration is vital for adaptive behaviour. Thus, studying animals in more naturalistic settings, where they are freely moving, is crucial for better understanding sensory processing. The overall aim of our lab has been to do this by combining electrophysiological recordings from freely moving mice with advanced methods for 3D tracking of body state. Here, we specifically focus on comparing the function of primary vs associational sensory areas, using the mouse whisker system as a model. Previous work on immobilised animals has shown that neurons in the secondary somatosensory cortex (S2) have larger and more complex receptive fields compared to the primary somatosensory cortex (S1) but no previous study has compared their function in freely moving animals. To investigate this, we chronically implanted Neuropixels probe into the S1 and S2 and recorded the neural activity in both regions simultaneously while the animal was freely moving and exploring objects in an open-field arena.

All the behavioural experiments were conducted in darkness under infrared illumination, with four cameras capturing the freely moving mouse behaviour. We tracked 11 landmarks on the head and body using DeepLabCut and reconstructed their 3D coordinates using a custom triangulation algorithm. Snout-to-surface distance (SSD), or the distance between the snout and the closest point on any of the arena surfaces, served as a proxy for whisker-surface touch. We also characterised the body state by extracting parameters such as 3D whole-body velocity, 3D allocentric head angles, their temporal derivatives, and principal components of the body shape from the 3D landmarks.

A supervised learning algorithm (XGBoost) was trained to predict the firing rates of S1 and S2 neurons based on SSD and body state parameters. To assess the extent of body state encoding in S1 and S2 and to exclude the possible correlation between whisker touch and body state, all the experiments were repeated after severing the infra-orbital nerve to eliminate whisker afferent input. We found that SSD was a statistically significant predictor of firing rate for the majority of S1 and S2 neurons. Incorporating body state into the prediction significantly improved the accuracy in both areas. These results suggest that body state modulation is widespread, affecting both primary and associative sensory cortices.

SA12

Cortical integration of body posture and the vibrissae in freely exploring rats

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Nervous systems continually integrate sensory and motor signals to coordinate behaviour, whether to enable controlled flight in an insect or a monkey plucking up a grain of food. To better understand the neural mechanisms that support movement coordination in cortex, my lab studies the neural representation of body posture in freely moving rats. We developed methods to integrate 3D motion capture with high-density neural recordings to visualize neural spiking in relation to 3D pose kinematics, which revealed that head and back kinematics are precisely encoded in posterior parietal cortex (PPC) and secondary motor cortices. More recent recent work suggested that posture and movement signals appear to be a general feature of primary sensory and motor coding, including in auditory, visual and somatosensory areas, during unrestrained behavior. The ubiquity of kinematic tuning in sensory cortices prompted us to expand our framework to include head-mounted high speed (200 FPS) tracking of the whiskers and eyes, and to ask how vibrissal deployment integrates with head kinematics and whole-body movement. This was combined with Neuropixels recordings spanning the PPC and neighboring S1 barrel fields. Ongoing work indicates that single neurons in S1 and the PPC alike conjunctively encode whisker posture and head kinematics, and that this can be further gated by locomotion. Our observations indicate that whisker deployment and head movements are fully interwoven during naturalistic behavior, which is reflected in cortical coding in both primary and associative areas.

C01

Development and Multisensory Influences on Huddling Behaviour in Rat Pups

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Early life experiences involving multisensory interactions with social conspecifics influence brain development and social behaviour in rat pups. Rat pups, reared with their kin, interact through contact, olfaction, and ultrasonic vocalisations, with the predominant multisensory social interaction pre-weaning being huddling behaviour. We investigated how rat pups develop huddling behaviour through these interactions, hypothesising that kin status and developmental stage influence huddle organisation and formation.

To test this, we used an open arena while recording video and ultrasonic vocalisations, both synchronised to observe huddle formation in groups consisting of three animals at three different group ages. We adhered to the regulations outlined by the University of Edinburgh Animal Welfare Committee, and all experimental tests were conducted under a United Kingdom Home Office project licence. Observing development from P6-8 (n=22 groups: kin=14 and non-kin=8), P11-14 (n=22 groups; kin=13; non-kin=9), and P18-20 (n=23 groups: kin=14 and non-kin=9), we found that groups formed more triad aggregons as they aged ($H(3)=22.5$, $p<0.0001$, non-parametric Kruskal-Wallis test), and these groups remained intact once formed.

Contact quieting, where vocalisations decrease with huddle formation, was noted especially during P6-8, but also in the P11-14 age groups. In the youngest age group (P6-8), non-kin groups exhibited more vocal behaviour compared to kin groups (number of vocalisations per 20-minute session: kin groups = 86.9 ± 6.2, non-kin groups = 138.0 ± 6.2; mean ± SEM; $U = 40.0$, $p < 0.001$, non-parametric Mann-Whitney U test). Exposure of kin groups to foreign bedding increased vocalisations, indicating that olfaction and contact interact to produce contact quieting, and that kin detection through olfaction leads to quieter vocal states in kin/familiar settings.

Overall, we observed that huddling behaviour is highly regulated by development, relies on the integration of multisensory cues, and may depend on the kinship composition of the huddling group.

C02

Passive movements evoke fast modulation of visual response in mouse visual thalamus

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Vision guides active movements such as walking or climbing. Execution of such movements is accompanied by predictable changes in animal's posture required to maintain balance and prevent falls. However complex environments also impose passive movements that cause unpredictable changes in animal's balance. Slips due to unstable terrains, movements imposed by vehicles' acceleration, suspended scaffolding or tree branches are all examples of postural challenges requiring fast motor adjustments.

Previous studies have shown that visual processing is modulated by active movements such as locomotion. However, it is currently unknown how visual processing is affected during passive movements imposed by the environment. We address this question by recording neuronal activity from the thalamic dorsal lateral geniculate nucleus (dLGN) in anaesthetized mice, positioned on platform tilting along pitch, roll, and yaw axes, and in freely moving mice placed on motorised tilting arena. In both anaesthetised and awake animals, we compared responses to light onset and offset when animals were tilted and when stationary.

Our findings reveal that 20%-30% of LGN neurons showed modulated activity in both anaesthetized and freely moving mice. Clustering analyses revealed that these effects encompass both ON and OFF cell types. In darkness, few cells responded directly to tilts in anaesthetised animals but ~25% cells were responsive in awake animals. Both modulation of light responses and direct responses occurred with remarkably short latencies (~50ms).

These results indicate that visual responses undergo fast modulation during passive movements of the tilting platforms. These effects could be functionally relevant to control balance during postural challenges.

C03

Melanopsin-Mediated Low Spatial Frequency Vision in Mice

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Keywords: low spatial frequency; preference choices; elevation range

Background: Behavioural and electrophysiological studies of pattern vision in mice typically address responses to high spatiotemporal frequency stimuli, but natural scenes contain patterns over a wide spatial frequency range. Coarse patterns contain information about important features of the local environment and yet we know little about how they are detected or how they impact mouse behaviour.

Aims: To establish behavioural and electrophysiological protocols for studying low-frequency vision in mice and to employ them to determine the contribution of the inner retinal photopigment melanopsin to this sensory modality.

Methods

We constructed a lighting arena capable of generating and quantifying low spatial frequency light patterns. This lighting arena consisted of two compartments, where the spatial distribution of light could be quantitatively controlled in each compartment. The light distribution across a -180° to 180° elevation range was quantified using the ELF method developed by Nilsson & Smolka (2021). Mice were placed into the lighting arena and we quantified time spent in each compartment as an indicator of preference. We used mice lacking cone ($Cnga3^{-/-}$) or melanopsin ($Opn4^{-/-}$) phototransduction and receptor silent substitution methods to separate the contribution of cone and melanopsin photoreceptors to pattern preference. Additionally, neural activity in the lateral geniculate nucleus of freely moving mice was recorded under these low-frequency spatial patterns.

Results

Our lighting arena is capable of generating various low-frequency spatial patterns, which we used in a series of behavioural trials. We found that these low-frequency spatial patterns could be detected and influence mouse choice at least as much as overall scene brightness. Local radiance around the horizon (-20° to 20°) primarily influenced mice's preference choices. Mice preferred to spend time in the compartment where the horizon was darker. This preference was retained in $Cnga3^{-/-}$ but disrupted in $Opn4^{-/-}$ mice. Correspondingly, in silent substitution experiments, visually intact mice specifically preferred horizons and grounds darker for melanopsin, suggesting that melanopsin plays a primary role in detecting low-frequency spatial patterns. Electrophysiological recordings revealed increased firing rates and altered power spectra in response to low-frequency

patterns, compared to brightness-matched uniform light. Further analysis of the electrophysiological data is ongoing.

Significance

Our findings demonstrate that mouse exploratory preferences are modulated by low-frequency patterns and especially by light intensity around the horizon. The detection of low-frequency spatial patterns appears to be mediated by melanopsin's sampling function.

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C04

Multiple locomotion modes emerge from reconfigurations of neural population dynamics in Aplysia

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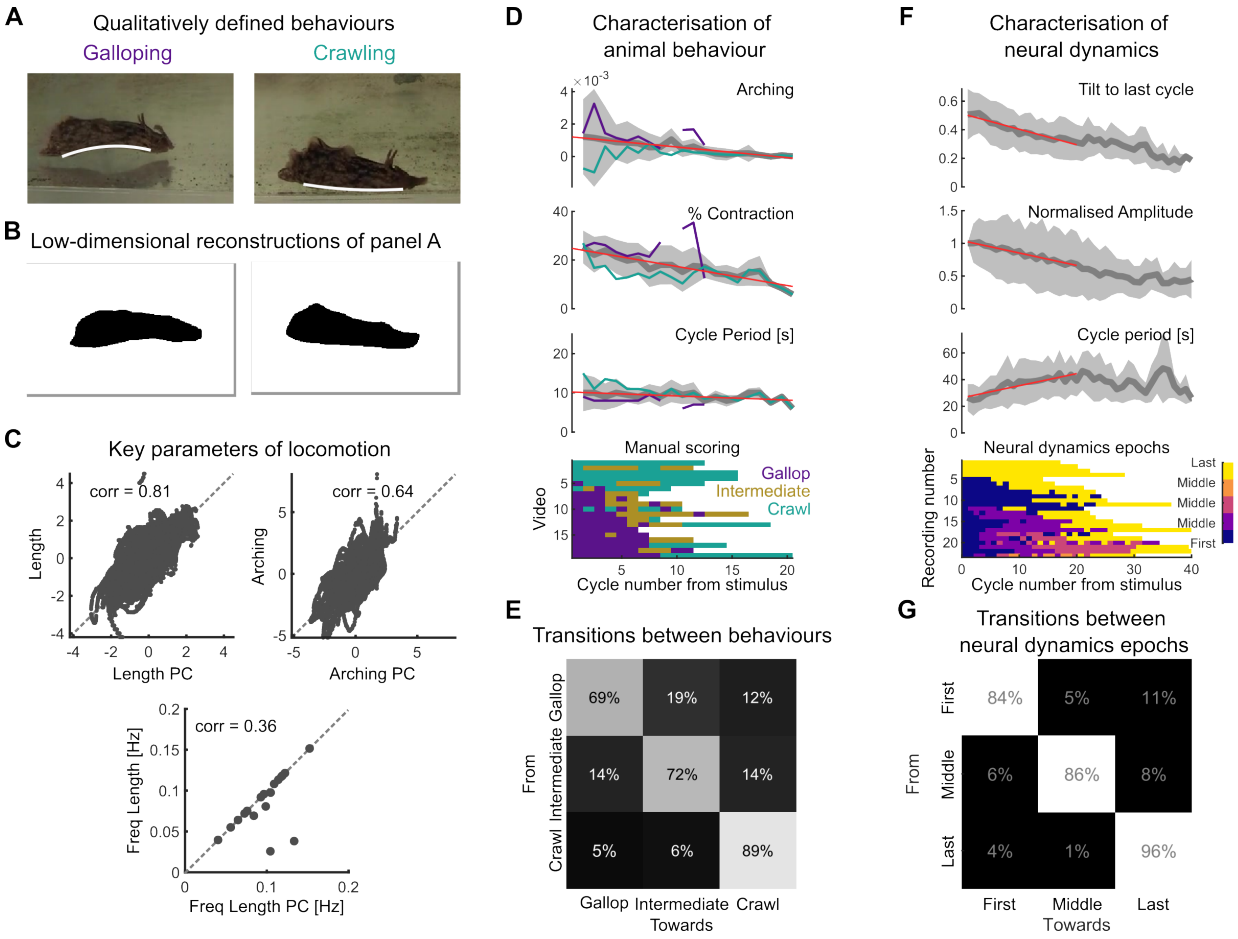
One of the key goals of neuroscience is to uncover the neural mechanisms that generate behaviour. Although much progress has been made in the development of tools that describe animal behaviour and the neural circuits that generate them in the last decade, how neural dynamics can give rise to multiple behaviours and the transitions among them remains unclear. Here we address this question for a ubiquitous behaviour such as locomotion in the sea slug Aplysia californica. We examine the neural dynamics underlying the transition between classically-defined locomotion modes: galloping and crawling (Fig 1A). These two modes have been classically defined based on qualitative changes in the animal shape (arching of the foot vs no arching) [1-4]. Yet, to date, no quantification of the animal locomotion escape has characterised these two modes and their transitions. Here we developed a customised tracker to characterise Aplysia's locomotor behaviours. Our unsupervised analysis of video-tracking data (N = 19) shows that both forms of rhythmic locomotion can be captured by a low-dimensional model (the eigenslug) (Fig 1B). The eigenslug reveals that locomotion is defined by three key parameters: the animal's length, the arching of its foot, and the period of each cycle of muscle contractions driving the movement (Fig 1C and 1D). These parameters are flexibly controlled to generate the specific mode of locomotion. Unlike in previous observations [4], we found that the escape response does not correspond exclusively to a gallop-to-crawl sequence, but transitions from crawl to gallop are also possible (Fig 1E). To understand the neural dynamics that give rise to these locomotion modes and the transitions between them, we imaged the population neural activity of the pedal ganglion using voltage-sensitive dyes during fictive escape locomotion for up to 20 minutes (N = 25 recordings). These recordings are an order of magnitude longer than previously reported experimental settings [5-6]. We found that population activity consistently forms a low-dimensional spiral whose parameters independently control each of the three locomotion parameters defined by the eigenslug: the spiral's tilt of rotation controls the arching of the foot, its amplitude controls the animal's length contraction, and its period controls the period of the animal's locomotion cycle (Fig 1F). Furthermore, unsupervised clustering of the neural dynamics based on the tilt of the neural trajectories recovers the statistics of the transitions between locomotion modes (Fig 1E and 1G). In this way, the locomotion modes emerge from continuous reconfigurations of the population's low-dimensional dynamics. These results suggest that flexible control of low-dimensional neural dynamics is a general principle for specifying and transitioning between multiple motor patterns.



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C05

Perceptual constancy for an odour is acquired through changes in primary sensory neurons

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The ability to consistently recognise an object despite variable sensory input is termed perceptual constancy. This ability is not innate, rather it develops early in life yet little is known about the neural processes underpinning its development.

We have taken advantage of the olfactory system of mice and using behavioural tests show that, when mice are naïve to an odour, perceptual constancy is not maintained for an odour across increasing concentration (n = 32). 2-photon imaging of neural activity in the olfactory bulb revealed that the perceptual change coincides with a rapid reduction in activity in a single glomerulus that is most sensitive to the odour (n = 9). This drop in activity is not a property of circuit interactions within the olfactory bulb; it is already present in the olfactory sensory neurons (n = 13). Computational modelling shows that the rapid adaptation at higher concentrations is due to a sensitivity mismatch of olfactory receptor neurons resulting in transmission failure from the nose.

We then show that upon forming an association of this odour with food, mice perceive the odour as the same object across the whole range of concentrations tested (n = 7). Correspondingly the sensitive glomerulus no longer displays rapid adaptation, due to a large sensitivity shift that matches its dynamic range to that of the food odour, when transmission failure is prevented, perceptual constancy is maintained. This work shows that the plasticity of the primary sensory organ enables learning of perceptual constancy.

C06

Measuring motivational switching in mice using open-design: the Switchmaze

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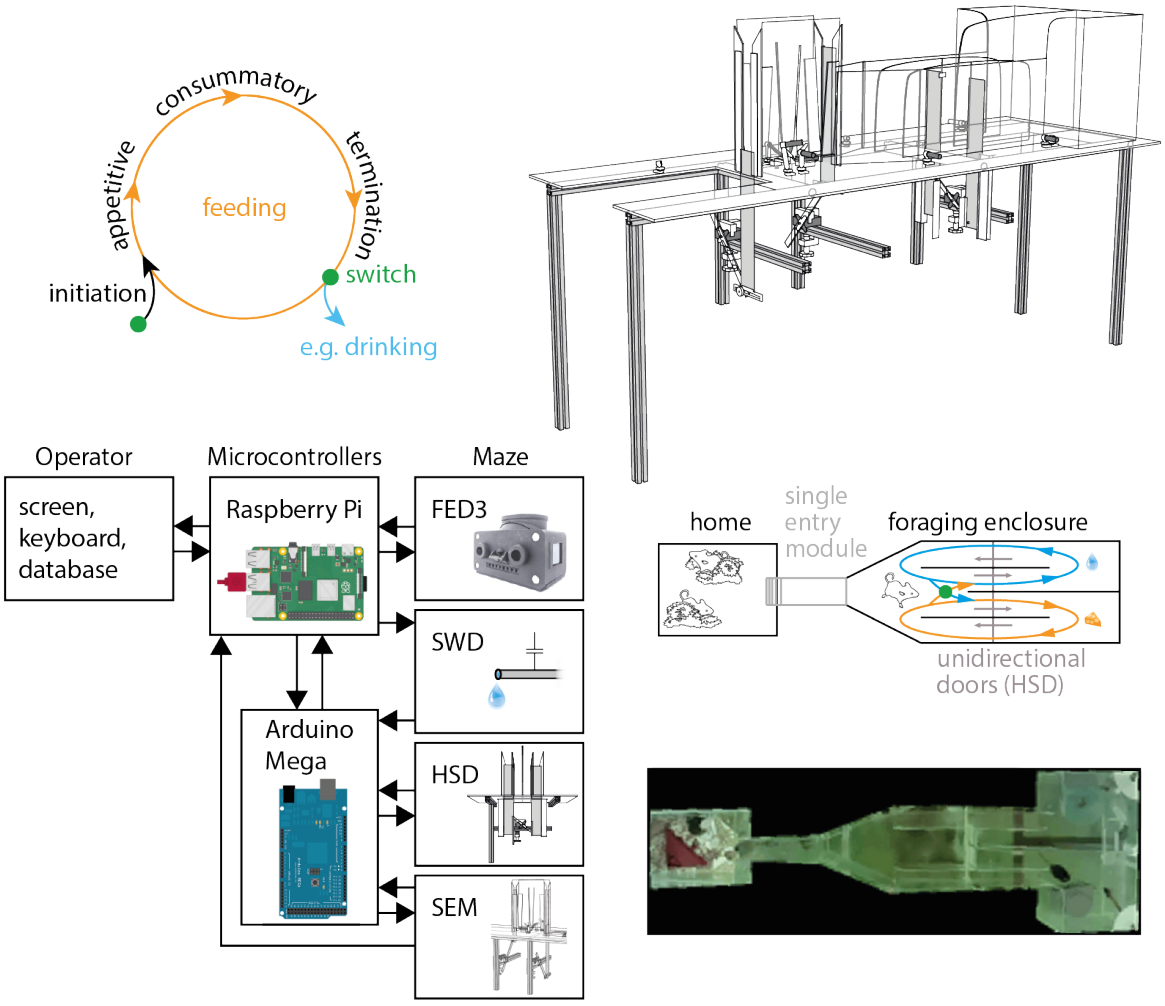
Animals need to switch between motivated behaviours, like drinking, feeding or social interaction, to meet environmental availability and internal needs. However, motivational switching is rarely studied, partly due to lack measurement systems. We designed an automated extended home-cage for measuring motivational switching in mice, the Switchmaze, using open source hardware and software (Figure 1). As proof-of-concept, we show environmental manipulation and targeted brain manipulation experiments which altered motivation switching without effect on traditional parameters.

Individual animals access the Switchmaze from the home-cage and choose between entering one of two chambers containing different goal objects or returning to the home-cage. Motivational switching is measured as a ratio of switching between chambers and continuous exploitation of one chamber. This motivation switching rate (MSR) was on average 1.4 ± 0.9 (mean ± SD, n=20 animals) after continuous habituation for >7 days. To test if motivational switching was different from random, we randomly shuffled the trial sequence and re-calculated MSR 1000 times for each animal. The arising distribution encompassed the actual MSR in most cases (only 4/30 animals had MSR higher than the 99th percentile of the shuffled data). This suggests that spontaneous motivation switching is optimized to appear random, which could conceivably function to decrease the predictive information available to competitors and predators.

To test the role of key neural populations in motivational switching with a loss-of-function manipulation, we expressed the inhibitory chemogenetic construct, hM4Di in either prefrontal cortex (PFC) output neurons to the hypothalamus (PFC-hM4Di) or in the perifornical hypothalamus (H-hM4Di). As a control cohort, we used wild-type mice that did not express a transgene, and which were interleaved in groups of hM4Di expressing mice that lived in the Switchmaze. We measured behaviour for 6 hours following a vehicle injection (i.p. saline) or C21, which activates hM4Di. Basic behavioural metrics were not changed in PFC-hM4Di or H-hM4Di mice upon activation of the inhibitory DREADDs with C21, as two-way repeated measures ANOVA tests showed no significant cohort-time interaction for food consumed ($F(2,27)=0.61$, $p=0.55$), block count ($F(2,27)=0.66$, $p=0.52$), trial count ($F(2,27)=0.50$, $p=0.61$), block duration ($F(2,27)=0.75$, $p=0.48$) or trial duration ($F(2,27)=0.69$, $p=0.51$). However, MSR was altered significantly (cohort-time interaction $F(2,27)=3.99$, $p=0.03$) and paired t-tests revealed the effect was a 46.5 ± 45.5% increase in the PFC-hM4Di cohort ($p=0.007$). The average switch rate exceeded the 99th percentile of switch rates expected from randomly permuted trial sequences.

Our results suggests that a behavioural role of the PFC is to regulate motivational switching in order to appear less predictable. The Switchmaze may be useful in scoring behavioural rigidity, which is a hallmark of many neuropsychiatric disorders. This work demonstrates the utility of open-design in understanding animal behaviour and its neural correlates.



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C07

Synaptic depression outperforms potentiation in learned stimulus discrimination under divisive normalization of opposing outputs

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Why are brains the way they are? Are their circuit architectures and synaptic plasticity rules in some sense 'optimal'? If so, in what sense, or in what contexts? We address these questions using olfactory associative memory in the fruit fly Drosophila. Flies can learn to associate a particular odour with a reward (e.g., food) or punishment (e.g., shock) and thereafter approach or avoid the trained odour. These associative memories are stored in Kenyon cells in the mushroom body, by weakening synapses from odour-responsive Kenyon cells onto mushroom body output neurons (MBONs) that lead to incorrect actions (e.g., odour+punishment weakens KC->Approach synapses). Why weaken incorrect actions rather than strengthening correct actions? Notably, synaptic depression is also used for learning in the vertebrate cerebellum, which has a remarkably similar architecture to the insect mushroom body, suggesting that using depression may be functionally advantageous.

We show both analytically and using simulations that depression outperforms potentiation for discriminating odours with overlapping KC representations, under a particular condition: if behaviour depends on the relative, not the absolute, difference between Avoid vs. Approach MBON activities (i.e. divisive rather than subtractive normalisation). To test whether behaviour depends on the relative difference, we measured aversive learning for a range of odour concentrations and punishment intensities, in an individual-fly T-maze (n=95-442 flies per condition). We automatically tracked the flies' decisions to enter or leave the side with the punished odour, and from the statistical distributions of these stochastic decisions, we inferred the mean and variance of the flies' underlying preference for/against the odour. We fitted these data to alternative mechanistic models for learned decision-making that used subtractive or divisive normalisation, and the fit was better with divisive normalisation. These results suggest that flies learn by synaptic depression because, in the mushroom body, it is computationally superior to synaptic potentiation. These results illustrate how quantitative analysis of natural behaviour illuminates neural mechanisms underlying learned decision-making.

N.A., K.G.-W., J.S.J. and M.W.T. contributed equally to this work.

C08

Mitochondrial origins of the pressure to sleep

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Introduction and aims

Sleep pressure, the process variable in sleep homeostasis, currently lacks a physical interpretation. Although prolonged waking is associated with numerous changes in the brain—of neuronal firing patterns, the strengths of synaptic connections, metabolite concentrations, and metabolic and gene expression programs —, it remains generally indeterminable whether these changes are causes or consequences of a growing need for sleep. Perhaps the only realistic opportunity for separating causation from correlation exists in specialist neurons with active roles in the induction and maintenance of sleep; in these cells, sleep's proximate (and maybe also its ultimate) causes must interlock directly with the processes that regulate spiking.

Results and methods

To obtain a comprehensive, unbiased view of molecular changes in the brain that may underpin these processes, we have characterized the transcriptomes of single cells isolated from rested and sleep-deprived Drosophila melanogaster flies. Transcripts upregulated after sleep deprivation, in sleep-control neurons projecting to the dorsal fan-shaped body (dFBNs) but not ubiquitously in the brain, encode almost exclusively proteins with roles in mitochondrial respiration and ATP synthesis (Figure 1). These gene expression changes are accompanied by mitochondrial fragmentation, enhanced mitophagy, and an increase in the number of contacts between mitochondria and the endoplasmic reticulum, creating conduits for the replenishment of peroxidized lipids (Figure 3). The morphological changes are reversible after recovery sleep and blunted by the installation of an electron overflow in the respiratory chain (Figure 3). Inducing or preventing mitochondrial fission or fusion in dFBNs alters sleep and the electrical properties of sleep-control cells in opposite directions: hyperfused mitochondria increase, whereas fragmented mitochondria decrease, neuronal excitability and sleep (Figure 4). ATP levels in dFBNs rise after enforced waking because of diminished ATP consumption during the arousal-mediated inhibition of these neurons, which predisposes them to heightened oxidative stress (Figure 2). Consistent with this view, uncoupling electron flux from ATP synthesis relieves the pressure to sleep, while exacerbating mismatches between electron supply and ATP demand (by powering ATP synthesis with a light-driven proton pump) promotes sleep (Figure 2).

Conclusions

Sleep pressure control has thus mitochondrial origins, and conditions that allow or prevent electrons flowing through the respiratory chain of its feedback control neurons determine its accumulation. Parallels with hunger-control neurons in the mammalian hypothalamus suggest similar mechanisms have been adopted for the control of both such metabolism-directed behaviours. Sleep, like ageing, may thus be an inescapable consequence of aerobic metabolism.

Ethical standards, statistical and methodological details

All the work presented here has been conducted with Drosophila melanogaster flies, for which ethical approval is not needed.

The investigators were blind to sleep history and/or genotype in imaging experiments but not otherwise. Sample sizes in behavioral experiments (typically n=32 flies per genotype) were chosen to detect 2-h differences in daily sleep with a power of 0.9. All behavioral experiments were run at least three times, on different days and with different batches of flies.

For further statistical and methodological details, see our preprint: **Sarnataro R**, et al., 2024 **Mitochondrial origins of the pressure to sleep**. bioRxiv, 2024.02.23.581770; doi: https://doi.org/10.1101/2024.02.23.581770

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C09

A novel modular maze for behavioural analysis in freely exploring mice

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Introduction & Aims

Animals in nature sense their surroundings by actively engaging with them and processing the resulting signals according to their utility. Traditionally, the neuronal circuitry of sensory signalling and behaviour has been primarily explored using head-fixed rodents [1]. However, head-fixation induces long-term stress [2], severely limits natural movements [3], and disrupts sensory perception and exploration strategies crucial for sensory-based tasks [4].

To address these limitations, we developed an experimental architecture for investigating these capacities in freely moving rodents. We aimed for the setup to allow for the exploration of context-dependent decision-making, flexible planning, and the abstraction of sequential rules. The setup enables study of foraging behaviour in freely moving mice without restricting their movement or access to nutrition. The maze includes automated animal tracking, stimulus presentation and reward delivery.

Methods

We built the modular labyrinth with 'Makerbeam XL' posts and PVC/acrylic panels, opaque in visible light but transparent under infrared, encouraging mouse exploration while enabling machine vision tracking [5]. The panels slot into the posts and can be replaced with devices such as 3D-printed food pellet dispensers and rotating tactile gratings, controlled by servo motors and activated by microcontrollers. Mouse entry into specific regions of interest (ROIs) is tracked using OpenCV and a Python state machine.

Results

We developed an automated sensory maze that combines allocentric and egocentric navigation. We found that mice readily habituate to this modular maze, are motivated to explore it with no need for water or food restriction, and perform object-in-place recognition as described in the literature.

Automation in the maze provides high levels of experimental control while enabling simple and flexible behavioural task design. Device (stimulus or reward) motion is triggered when the mouse enters specific user-configured ROIs. The system supports flexible maze reconfiguration and scalability. An animal can encounter multiple stimuli as it moves from the labyrinth's origin to any endpoint, permitting the experimenter to set up complex rules or conditions governing whether the mouse will be rewarded, involving chains or sequences of stimuli, such as tactile or auditory. The entrance connects to a home cage, permitting free movement between the maze and the cage.

The maze is being used with two different types of stimuli. (1) Sequences of auditory stimuli, delivered according to animal presence in an ROI, are used to investigate how sensory predictability, as evaluated through the learning and processing of sequential structure, influences the emergence of intrinsic reward and subsequent decision making. (2) Sequences of oriented tactile cues are controlled on a trial-by-trial basis and predict the location of rewards in different maze chambers.

Conclusion

Our maze offers a practical and cost-effective approach for studying a wide range of cognitive behaviours in laboratory settings. Through this automated maze we model foraging behaviour, allowing mice to perform abstract sensory-guided sequential tasks, demonstrating context-dependent decision-making and probabilistic rule learning. The platform is designed to be easy to modify and share, through its use of inexpensive and readily available components.

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C10

Amygdala as a retino-recipient target for assessing light's effects on mood

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Light is crucial for vision but its roles in driving non-visual functions are similarly important. Light-dependent regulation of mood is evident from both human and rodent research, however, the biological mechanisms that underlie these non-visual effects are unknown.

The amygdala is a major component of the limbic system, known for emotional processing and mediating stress-induced anxiety. Furthermore, the latest research has identified that medial and central nuclei within the amygdala receive sparse yet direct retinal projections including innervation by melanopsin-expressing ipRGCs (intrinsically photosensitive retinal ganglion cells) that govern non-visual functions, suggesting that the amygdala physiology may be influenced by photic information.

Consequently, we set to investigate what visual input the amygdala receives using in vivo electrophysiology in anesthetized mice while presenting a range of visual stimuli. For this purpose, we utilized red cone mice (Opn1mw^{R}), a genetically modified strain that expresses human L cone opsin in place of mouse M cone opsin. This enables the use of silent substitution approaches where individual responses from different photoreceptors can be isolated based on their divergent spectral sensitivities using polychromatic light sources. We designed a set of photoreceptor-specific stimuli targeting melanopsin, rods, and individual cone opsins which we tested and validated in the red cone mouse visual thalamus before applying to define photoreceptor contributions to light-responsive neurons within the medial and central nuclei of the amygdala.

C11

Investigating non-sensory neuronal responses across cell types in the somatosensory cortex.

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The somatosensory cortex has been predominantly considered as a brain region encoding sensory information, such as that related to touch, in a somatotopically organised manner. Recent studies have contradicted this established view, showing that excitatory neurons in the somatosensory cortex can respond to information related to non-sensory aspects of behaviour, such as motor actions or rewards. However, it is unknown whether different cell types within the circuit respond to distinct non-sensory aspects of behaviour when an animal attempts to discriminate between different tactile stimuli. Here, we performed two-photon calcium imaging of different types of inhibitory neurons from head-fixed mice trained on a whisker-based tactile sequence discrimination task, to identify the responses of those neurons during behaviour. We imaged parvalbumin (PV)-, vasoactive intestinal polypeptide (VIP)-, and somatostatin (SST)- expressing interneurons in the barrel cortex. In preliminary data we found interneurons that increased their firing rate to non-sensory aspects of behaviour relevant to the task goal, such as licks. Motor related behaviour we have seen in all three cell classes included either neurons increasing their firing rate before the onset of both correct and erroneous licks, or neurons responding only before the onset of correct licks. Sensory responses were unequally represented in all three classes, with SST interneurons showing the highest fraction of sensory responses. We also found neurons with more complex activity patterns. We are currently incorporating locomotion and arousal (via pupillometry) into our analysis of these more complex aspects of neuronal behaviour. In summary, our data so far has shown that not only excitatory but also inhibitory neurons in the barrel cortex can show a response preference to multiple task variables including motor behaviour during a tactile sequence discrimination task.

C12

Genetic Dynamics In An Interplay Between Micronutrient Plasma Values And Behavioural Responses To Stress In Undiagnosed Premenstrual Dysphoric Disorder

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Genetic Dynamics In An Interplay Between Micronutrient Plasma Values And Behavioural Responses To Stress In Undiagnosed Premenstrual Dysphoric Disorder

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ABSTRACT

Behavioral patterns are the regular ways in which individuals act or respond to their environment, influenced by a combination of internal and external factors. Premenstrual Dysphoric Disorder (PMDD) is a pathological spectrum of emotional and somatic symptoms observed during the luteal phase of menstrual cycle interfering with the physical and social life of the individual. The aim of the study was to investigate the Genetic dynamics in an interplay between some micronutrient plasma values and behavioural responses to stress in undiagnosed PMDD. The study was carried out in Benin city, Nigeria. A total of 200 apparently healthy young female adults were recruited for this study. The various phases of the menstrual cycle were determined via gynecological questionnaires. They were classed into 2 groups: Those without PMDD symptoms (control) and those with PMDD symptoms (test). Five (5.0) mls of whole blood was collected, dispensed into Lithium heparin containers and centrifuged. Resulting plasma was separated into plain bottles and frozen for cortisol and trace metals analysis. One (1.0) ml was of whole blood was collected and dispensed into 0.5ml DNA shield container for genetic studies. Analyses were carried out in the University of Benin, University of Benin Teaching Hospital (UBTH) and Federal University of Technology, Akure (FUTA), Nigeria. All data were presented as mean ± standard error of mean (SEM). Statistical analysis was done using graph pad prism 8.1. The data was evaluated using two-way analysis of variance (ANOVA) utilizing the F test. Data was expressed as the mean value ± SEM for the control and test groups. Differences within the groups were then assessed using least significant difference (LSD) and p-values less than 0.05 ($p<0.05$) was considered statistically

significant. Result showed that cortisol concentrations were significantly higher for the test participants during the luteal phase when compared with the menstrual phase ($p<0.05$). Plasma zinc concentration was significantly lower in the test subjects during the luteal phase compared to the menstrual phase ($p<0.05$). CYP 17 gene expression was significantly up regulated in the test subjects compared to control subjects during the luteal of the menstrual cycle ($p<0.05$). The ESC/E (z) genes were significantly down regulated in the test subjects when compared with the control subjects ($p<0.05$). In conclusion, The study demonstrated a significant association between reduced plasma zinc concentration and increased irritable and depressed-state behavior.

Key Words: Premenstrual Dysphoric Disorder (PMDD), Irritableness, Depressed-state, Extra sex comb/Enhancer of Zest genes, Cytochrome P450 -17 gene.

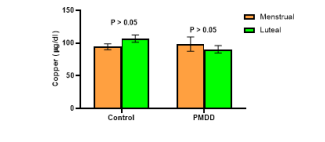

Fig.11: Showing Copper concentration during the menstrual and luteal phases of the PMDD and control participants.

There was no significant difference at luteal phase when compared with menstrual for control and PMDD participants.

GENETIC STUDIES

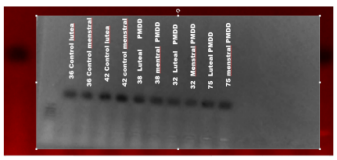

Fig.12: Showing Up regulation and down regulation of ESC/E(Z) gene expression in Premenstrual dysphoric disorder (PMDD) and in control participants. The thickness of the bands represents the mRNA expression of the gene. There was a decrease in the gene expression in the test participants during the luteal phase when compared with the luteal phase of the control participants.

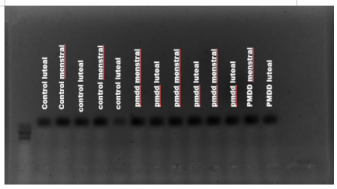

Fig.13: Showing the thickness of the bands represents the mRNA expression of the gene. There was significant increase in the CYP17 gene expression in PMDD participants meaning there was an increase in steroid hormones synthesis and in this case sex hormones (estrogen and progesterone) and glucocorticoids (cortisol) in the test participants compared to the control participants in both the luteal and menstrual phases.

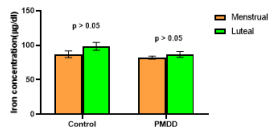

Fig.8 : Showing Iron concentration during the menstrual and luteal phases between test and control participants.

There were no significant difference during the luteal phase compared with menstrual for control and PMDD participants.

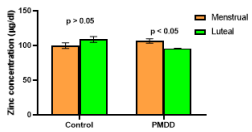

Fig.9: Showing Zinc concentration during the menstrual and luteal phases between test and control participants.

There was a significant difference during the luteal phase when compared with menstrual for control but no significant difference during the luteal phase compared with menstrual for PMDD participants.

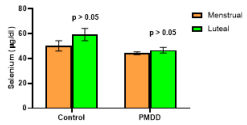

Fig.10: Showing Selenium concentration during the menstrual and luteal phases of the PMDD and control participants.

There were no significant difference at the luteal phase when compared with menstrual for control and PMDD participants.

RESULTS

Results were presented in graphs.

GRAPHS

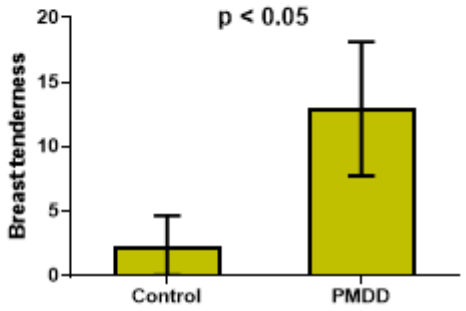

Fig.1: Showing breast tenderness between test and control participants.

There was a significant increase in the breast tenderness index during the luteal phase when compared with the menstrual phase for both control and PMDD participants.

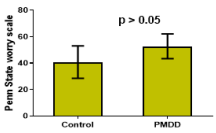

Fig.5: Showing the Penn state worry scale between test and control participants.

There was an increase in the uncontrollable worry index between control participants when compared with the PMDD participants but not statistically significant

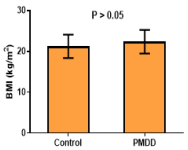

Fig.6 : Showing the Body mass index between test and control participants.

There was no significant difference between control participants when compared with the PMDD participants.

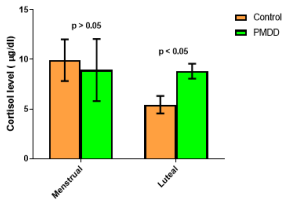

Fig.7 : Showing cortisol level during the menstrual and luteal phases between test and control participants.

There was a significant difference in the PMDD participants during the luteal phase when compared with the control participants.

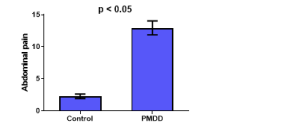

Fig.2: Showing abdominal pain between test and control subjects.

There was a significant increase in the abdominal pain index during the luteal phase when compared with the menstrual phase for both control and PMDD participants.

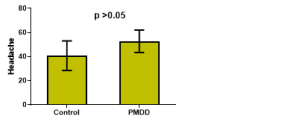

Fig.3: Showing the headache sensation between test and control subjects.

There was an increase in the headache sensation index in the control participants when compared to the PMDD participants.

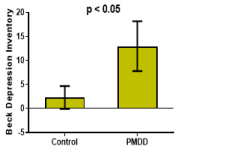

Fig.4: Showing the Beck's Depression inventory between test and control participants.

There was a significant increase in the depression index in the test participants when compared with the PMDD participants.

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C13

Investigating the Impact of Light Exposure on Mood in Everyday Life: A Naturalistic Study

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In the last decade, it has become apparent that light is not only important for vision, but that light responsiveness extends to fundamental aspects of mood, cognition, and performance. Light can be used as an effective and non-invasive therapeutic option with little to no side effects, to improve sleep, mood, and general well-being. However, to utilise the full potential of light to improve mood and well-being and design new interventions, further research is required, starting with determining the relationship between light and mood in human subjects in real-life situations.

This study's methodology will be the first to offer information into naturalistic light exposure situations and provide insight into its relationship with emotional bias in participants. In total, 25 healthy volunteers are recruited for a 1-week light monitor study. Participants will receive two devices—a light monitor (which measures the gold-standard unit of ambient light exposure, Melanopic-EDI) for daytime use and a Fitbit for continuous sleep and physiology tracking (activity and sleep patterns, heart rate). They are given access to an online questionnaire link. During the week, participants are asked to complete questionnaires, including a baseline questionnaire, which asks for information on sleep health, general health and demographics, in addition to a daily sleep diary and tasks aimed at assessing their current mood status. This includes both repeat subjective mood questionnaires and daily objective cognitive tests of emotional bias, based on using validated tests including the Emotional Categorisation Test (ECAT), Emotional Recall Task (EREC) and Emotional Recognition Memory Task (EMEM). This will also be the first time these objective mood tasks are used across the week, they have been adapted to be conducted on separate days, not just in one sitting.

Here, we describe the preliminary results of 5 healthy volunteers, which demonstrate the feasibility of the measurements we are collecting. We gathered on average 7.5 days of continuous melanopic EDI data from everyday life, proving the acceptability of our light monitoring protocol. Preliminary results suggest no significant differences between positive or negative bias across days. There were also no significant differences between days and number of incorrect words, suggesting no practice effect for the tasks. Future work will focus on analysing circadian and light exposure impacts on mood regulation. We anticipate that the analysis of the complete dataset will reveal daily rhythmicity of mood, efficacy of objective emotion tasks in detecting mood variations in real-world settings, and relationships between light exposure, physiological variables, and mood in healthy volunteers.

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C14

Inclusion of females in autism research; behavioural differences in valproic acid-induced rat model of autism

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Autism Spectrum Disorders (ASD), are characterized with impairments in communication, lack of interest in social interactions and repetitive behaviours. Even though the 4–5:1 male-to-female ratio of diagnosis is reported, the inclusion of both sexes in research is important, especially for the risk of misdiagnosing females. Prenatal valproic acid (VPA) exposure is used to model ASD-like symptoms in rodents (1). The prevalence of ASD in children exposed to VPA during pregnancy is reported to be 1:1 male-to-female ratio (2). This study aims to investigate how both males and females exhibit ASD-like symptoms after prenatal VPA exposure.

Pregnant Wistar Albino rats received either 400mg/kg VPA (n_{Mother}:4; $n_{Female\ Pup}$:8, $n_{Male\ Pup}$:8) or saline (n_{Mother}:4; $n_{Female\ Pup}$:8, $n_{Male\ Pup}$:8) on embryonic day 12.5 intraperitoneally. Pups were tested for olfactory discrimination (OD) on postnatal day 9 (P9). Latency to reach mother bedding was measured. Three-Chamber Social Interaction test (TCSI) was used to assess sociability on P25. Time spent in a chamber containing a stranger rat was measured. Locomotion was assessed in Open Field (OF) on P30. Total distance moved and immobility was measured. Anxiety was measured in Elevated Plus Maze (EPM) on P32. The frequency to entre open/closed arms was measured. Hole-Board apparatus was used to assess exploratory behaviour on P35. The frequency of head-dipping was measured. Statistical analysis was performed with Two-Way ANOVA test on Sigma-Plot. The study was approved by Bursa Uludağ University's ethics committee (numbered:2024-01/08).

Pups that were prenatally exposed to VPA showed differences compared to the control group in both sexes. Latency to reach mother bedding was significantly increased in both females and males of the VPA group ($p<0.001$; $p<0.001$). There was not a significant interaction between sex and VPA exposure ($F(1.31)=0.515$, $p=0.479$). In the TCSI test, both females and males of the VPA group spent significantly less time in the chamber containing a stranger rat ($p<0.001$; $p<0.001$). Interaction between sex and VPA exposure was significant ($F(1.31)=43.175$, $p<0.001$). Both females and males of the VPA group travelled significantly less ($p<0.001$; $p<0.001$) and spent significantly more time immobile ($p=0.011$; $p=0.026$) in OF. There was not a significant interaction between sex and VPA exposure in distance moved ($F(1.31)=0,189$, $p=0.667$) and immobile time ($F(1.31)=0.0734$, $p=0.788$). In EPM, VPA females had a higher ratio of entry to open arms compared to control females ($p<0.001$), whereas VPA males had a lower ratio of entry to open arms compared to control males ($p=0.002$). Interaction between sex and VPA exposure was significant ($F(1.31)=38.323$, $p<0.001$). The frequency of head-dipping was significantly lower in both females and males of the VPA group ($p<0.001$; $p<0.001$). Interaction between sex and VPA exposure was significant ($F(1.31)=34.300$, $p<0.001$).

This study suggests that prenatal exposure to VPA induces ASD-like symptoms in both male and female rats. Notable differences between control and VPA groups were reported in performed

behavioural tests, with some of these differences being sex-dependent. Expression of autism varies in females, which often causes underdiagnosis and leads to loss of effectiveness of treatment. Therefore, the inclusion of both sexes in research is crucial and should be emphasized.

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C15

Investigating the role of auditory cortex in interaural level difference perception in mice

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The environment is constantly changing around us, to provide accurate information upon which to base decisions and actions, the brain must be flexible and adapt to these rapidly changing surroundings. To deal with these changes, neurons can adapt their sensitivity or tuning to represent the most important aspects of the environment. Sound location is a fundamental aspect of understanding the auditory environment and is important for survival, after all one wants to run away from danger, not towards it. Neurons in the brain can adapt to sound location and sound location perception of humans can be altered by spatial adaptors[1]. However, the mechanisms underlying neuronal adaptive coding and how it directly alters auditory spatial perception remain unknown. We aim to perform simultaneous neuronal activity recordings and sound location perception testing in the presence of adapting stimuli to investigate the mechanism of spatial adaptation and the effects of adaptation on auditory spatial perception.

To study how adaptation to sound location affects perception we first designed a behavioural task to probe sound location perception in mice. Given the size of the mouse head, it is most likely that mice rely on interaural level difference to localize sounds in the horizontal plane[2]. Subsequently, head-fixed mice were trained in a lateralization task where they reported the location (left/right) of a sound based on the interaural level difference. There is a large body of work to suggest that the auditory cortex is necessary for sound localization ability[3], however, this has not been tested in mice. Furthermore, it has been shown that feedback from the auditory cortex (AC) to the inferior colliculus (IC) affects spatial tuning properties of neurons in the IC[4] and is necessary for adaptation to unilateral hearing loss[5]. To test the role of AC in sound location perception, we used optogenetics to modulate activity of AC or cortico-collicular feedback neurons during task performance.

We found that mice were able to perform the task, with 5/6 mice successfully completing training and testing. Sensitivity to sound location was assessed by finding the slope of a logistic regression fitted to the psychometric curves of mice with and without optogenetic manipulation. Inactivation of excitatory neurons in the auditory cortex during task performance reduced sensitivity to sound location (n = 2 mice), while neither inactivation (n = 2 mice) nor activation (n = 2 mice) of cortico-collicular feedback neurons during task performance affected sensitivity to sound location.

Mice successfully learned to report the location of a sound based on the interaural level difference. Preliminary results indicate that inactivation of excitatory activity in the auditory cortex affected task performance and reduced sensitivity of mice to sound location. Modulation of cortico-collicular feedback did not affect task performance or the sound location sensitivity of the mice. In future work mice will be required to perform the task in the presence of a spatial adaptor sound.

All experimental procedures were in accordance with NIH guidelines and approved by the IACUC at the University of Pennsylvania.

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C17

A blueprint for advancing neurethological research platforms for the generation of continuous multi-day neuro-behavior datasets

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Understanding the neural underpinnings of voluntary or self-initiated actions is crucial for advancing psychological and behavioral sciences and for understanding neurological diseases. Progress in neuroethology necessitates improvements in neural recording systems, enrichment paradigms, and data science methods. One significant challenge is the continuous recording of biometric or neural data from model animals, such as rodents, over extended periods. Current tethered neural headstages require commutators or periodic manual disentanglement, and available wireless data loggers are limited to a few hours of recording, necessitating constant human assistance.

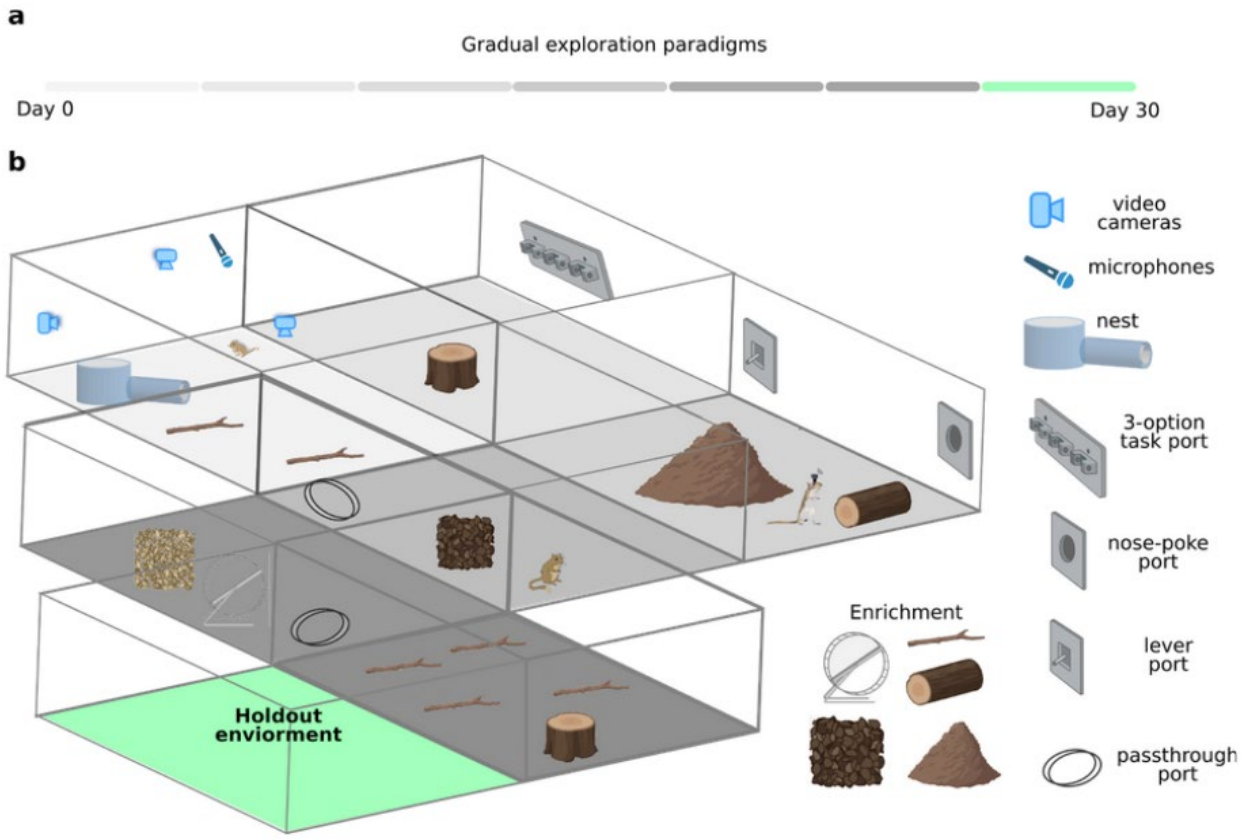
We propose a research framework and platform for automating neural and behavior recordings in dynamic open-field paradigms to facilitate the collection of large-scale datasets required for modern machine learning methods. Our proposal outlines three critical advancements: the personalization of low-cost wireless neural loggers, automation of large-scale neuro-behavioral data collection, and the use of sequentially accessible arenas.

For low-cost wireless loggers, we propose a blueprint for custom headstages using standard microamplifiers, such as those from Intantech and IMEC. Our blueprint includes a printed circuit board (PCB) design that can be easily modified and sent to an array of existing low-cost PCB printing companies. The design integrates optional devices, including microphones, 3-axis accelerometers, Wi-Fi capabilities, and memory card and battery pack integration.

For automating data collection, we present a paradigm where self-head-fixing rodents can facilitate the automated removal, recharging, and replacement of neural loggers. This paradigm relies on mechatronics approaches and low-cost robotic arms that have sub-millimeter spatial resolution and high-sensitivity to handle neural loggers while avoiding damage or miss-insertions.

Lastly, we propose multi-chamber open-field arenas (MOFA) designed to provide stochastic-causal enrichment - creating learning paradigms where rodents can learn a variety of stimulus priors as well as causal effects of their actions. The arenas include randomized sensory cues to measure psychometrics related to perception, motivation, and decision-making. The MOFA setup involves gradual exploration paradigms over several days, allowing mice to progressively explore different areas. This approach enables the evaluation of computational models' generalization based on specific sections of the environment, rather than traditional data hold-out methods.

Our blueprint aims to enhance and accelerate the study of neural dynamics and behaviors, generating large-scale datasets for machine learning and traditional research.


a
Gradual exploration paradigms
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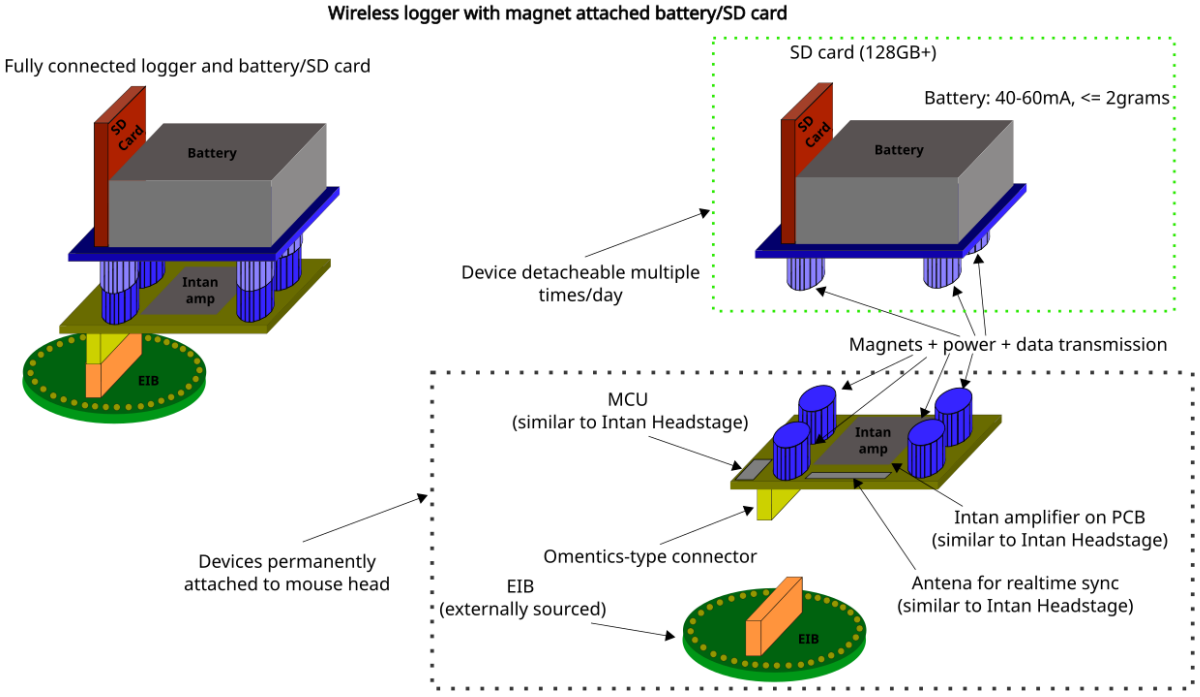
Wireless logger with magnet attached battery/SD card
Fully connected logger and battery/SD card
SD Card
Battery
Intan amp
EIB
SD card (128GB+)
Battery: 40-60mA, <= 2grams
Device detacheable multiple times/day
Magnets + power + data transmission
MCU
(similar to Intan Headstage)
Intan amp
Intan amplifier on PCB
(similar to Intan Headstage)
Devices permanently attached to mouse head
Omentics-type connector
EIB
(externally sourced)
Antena for realtime sync
(similar to Intan Headstage)
EIB

C18

Zinc deficiency: A harbinger of vestibular dysfunction?

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BACKGROUND

Role of zinc in the maintenance of redox homeostasis have been explored & is established to have antioxidant effects in the body. Presence of zinc in auditory system & its role in deafness & tinnitus is well established. Zinc supplementation has shown to not only improve tinnitus but also dizziness associated with it which marks the presence of zinc in vestibular system [4]. The possible role of zinc in modulating neurotransmission across the glutaminergic synapses in vestibulo cerebellar, vestibulo-occular pathways are established. So, this study aims to assess serum zinc levels in vestibular disorder patients & correlate with severity of the vestibular dysfunction

METHODOLOGY

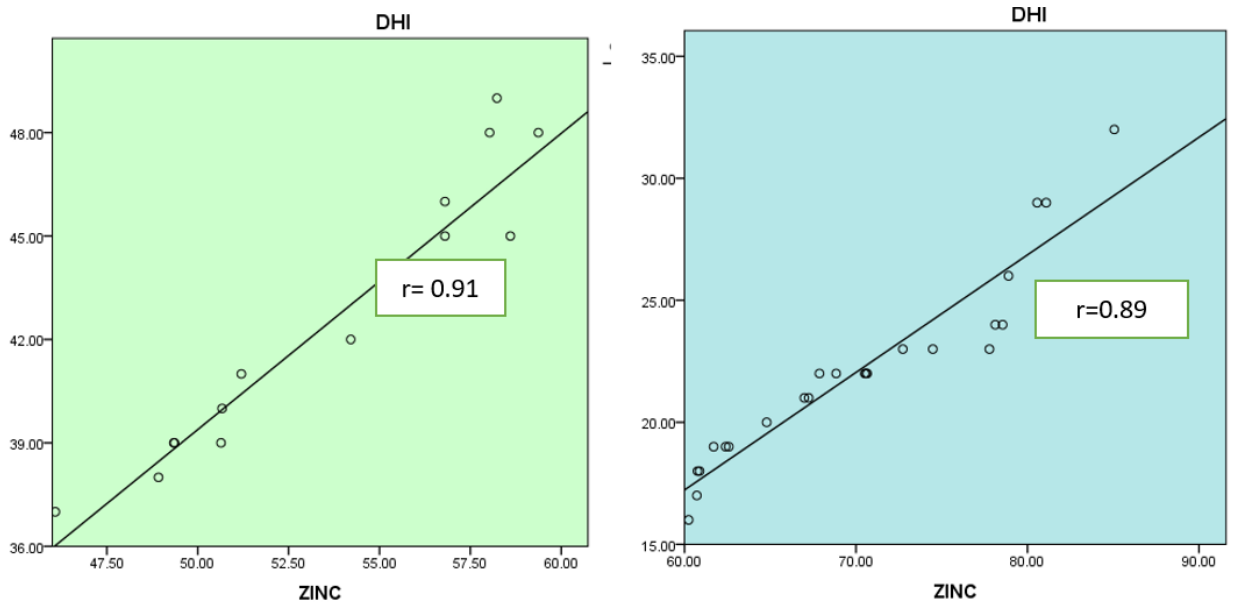
This was Cross sectional comparative study, 40 patients with vestibular disorders & 40 subjects without vestibular dysfunction were included in the study. After thorough history taking, these patients were subjected to Dix Hallpike test, Head Impulse test Romberg test on foam with eyes closed /The Clinical Test of Sensory Interaction and Balance (CTSIB) to confirm vestibular dysfunction. After that, Dizziness handicap inventory (DHI) to assess the severity of the vestibular dysfunction. Serum zinc levels along with other, micronutrients like magnesium, calcium & Serum vitamin B12 & vitamin D were also assessed. Serum zinc levels were compared with age matched controls.

Results:

Out of 40 patients, 24 had mild & 16 had moderate scores according to DHI. Serum zinc levels in study group was **60.63**±10.10 which was significantly ($p<0.005$) lower than compared control group **70.50**±19.1. Also serum zinc levels significantly($r:0.91, p<0.005$) reduced when the severity of the dysfunction increases.

Conclusion

zinc supplementation in vestibular disorder patients can be considered as add on therapy, which might have a beneficial role with respect to cognitive function as well.



A(MILD) B (MODERATE)

Fig 1: Correlation of s. Zinc levels with severity of vestibular dysfunction

Table 2: Micronutrients in both the groups

	Study group (Mean & SD)	Comparative group (Mean & SD)	T value	P value
S. zinc	**60.63**±10.10	**70.50**±19.1	**2.889**	**0.004 ***
S. Magnesium	**2.13**±0.34	**2.27**±0.19	**2.768**	**0.022**
Vitamin D	**22.53±12.2**	**16.86**±7.43	**2.923**	**0.014**
Vitamin B12	**216.38**±158.10	**198.3**±87.8	**0.6323**	**0.529**
Folic acid	**10.60**±5.59	**8.78**±6.43	**1.3510**	**0.1806**

*statistically significant

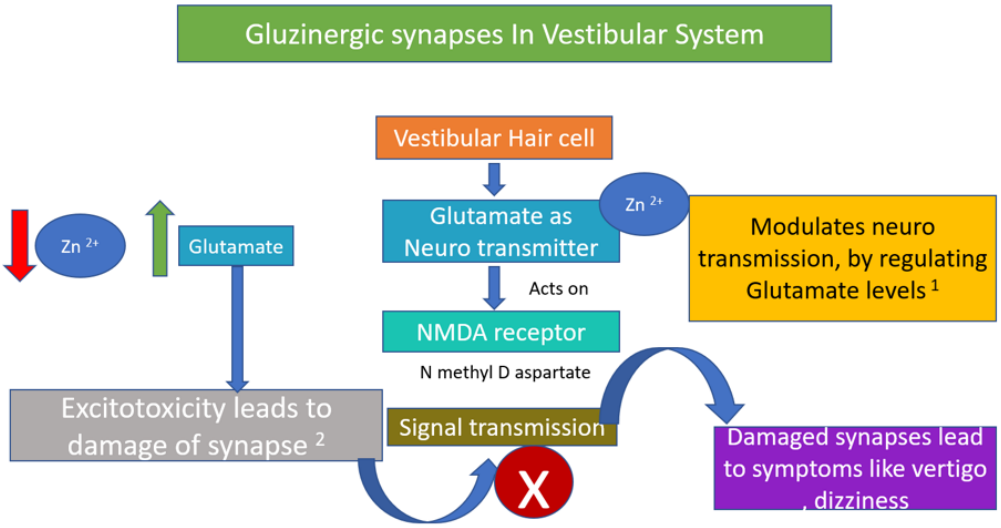


Fig 2: Events following zinc deficiency in gluzinergic synapse