Hexadirectional modulation of grid cell firing by firing rate adaptation

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Abstract

Grid cells are crucial for encoding spatial knowledge and supporting navigation. To non-invasively detect grid cell activity in humans, hexadirectional modulation (HM) methods have been developed. These methods look for macroscopic brain activity that varies with movement directions with six-fold rotational symmetry. Despite their widespread use with functional neuroimaging, the cellular origin of HM remains unclear. Here, we analysed over 1,000 grid cells recorded from freely foraging rats to evaluate two proposed mechanisms: (1) firing rate adaptation and (2) directionally tuned conjunctive grid cells. We observed significant HM after controlling for variability in speed and location using a Poisson multiplicative model. Consistent with the adaptation hypothesis, firing was reduced when grid fields were visited in close succession, a pattern more common during grid-aligned movement. These results offer a physiological explanation for HM and inform the design of future experiments aiming to detect grid cell activity non-invasively.

Keywords: grid cells, hexadirection, six-fold, adaptation, repetition suppression

Introduction

Grid cells fire at multiple periodic locations across the environment, supporting spatial navigation and memory. They have been implicated in a range of cognitive functions and deteriorate early in Alzheimer's disease (Ying et al., 2023; Fu et al., 2017).

To non-invasively detect grid cell activity in humans, hexadirectional modulation (HM) methods have been developed (Doeller et al., 2010). These measure six-fold rotational symmetry in macroscopic brain signals—such as BOLD in fMRI or theta power in MEG and EEG—modulated by running direction (Fig. 1A). Although widely used, the cellular origin of HM remains elusive (Bin Khalid et al., 2024).

Two main mechanisms have been proposed: (i) firing rate (FR) adaptation, where aligned movement direction causes grid fields to be visited in close succession, leading to suppressed firing; and (ii) the contribution of directionally modulated "conjunctive" grid cells, whose preferred directions might align with grid axes (Fig. 1).

Here, we examined the source of HM by analysing a large dataset of grid cells (1,400 cells from 167 recordings across 24 rats) recorded during free foraging (Barry et al., 2012; Gardner et al., 2022; Hafting et al., 2005; Krupic et al., 2015). To isolate genuine hexadirectional modulation, we accounted for confounding effects due to inhomogeneous sampling of location, direction, and speed during natural behaviour (Burgess et al., 2005).

Conjunctive cells don't explain HM

To test whether conjunctive directional grid cells contribute to hexadirectional modulation (HM), we examined the alignment between their preferred movement directions and their grid orientations. Among 251 conjunctive grid cells recorded from 15 rats, preferred directions were not significantly clustered around any multiple of the grid orientation (e.g., 0° + 60°k). This suggests that conjunctive cells are unlikely to explain the observed HM.

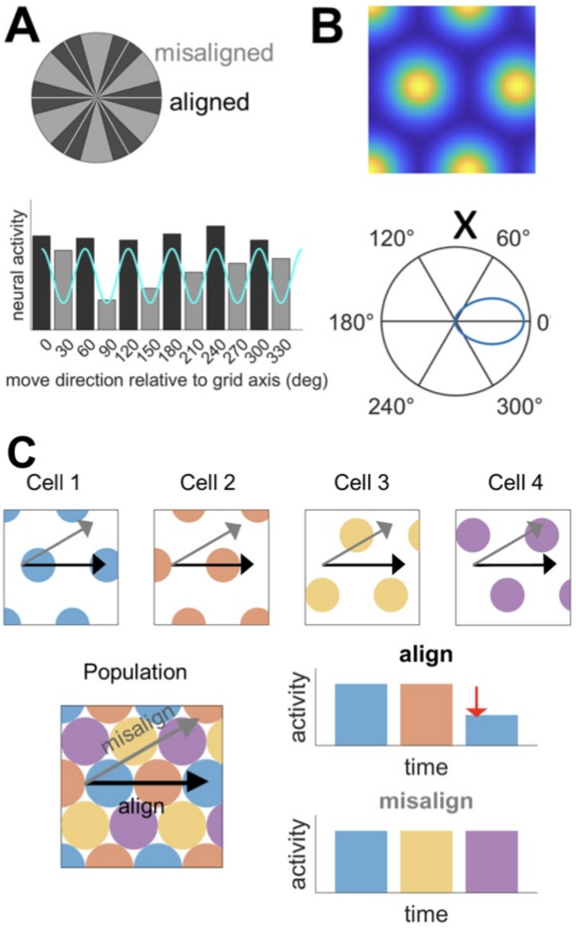

Figure 1: Hexadirectional modulation **(A)** could arise from conjunctive grid cells with head direction aligned to grid axes **(B)** or from firing rate adaptation **(C)**.

Hexadirectional modulation in grid cells

We first applied a simple linear regression to estimate hexadirectional modulation in the firing rate of grid cells, using the following model:

$$FR = 1 + \beta \cos[6(\theta_k - \phi)]$$

where θ_k is the instantaneous movement direction, ϕ is the grid orientation, and β quantifies the strength of HM. This analysis yielded negative β values, with high variability and not reaching significance, suggesting that HM is not reliably detected without accounting for behavioral confounds.

To address this, we used a Poisson multiplicative model that controls for variability in location and speed. Spike counts were binned by spatial location (x_i, y_j) and movement direction θ_k, and the expected spike count in each bin was modeled as:

$$\mathbb{E}(x_i, y_j, \theta_k) = f(x_i, y_j) \cdot [1 + \beta \cos(6(\theta_k - \phi))] \cdot \left[1 + \gamma \cdot v_{ijk}\right] \cdot t_{ijk}$$

Here, $f(x_i, y_j)$ is the mean firing rate at location (x_i, y_j), v_{ijk} is the normalized mean speed in each bin, γ is the speed modulation coefficient, and t_{ijk} is the time spent in the bin.

Using this model, we found a small but significantly negative β, indicating lower firing rates during grid-aligned movement (Fig. 2A). This result supports the firing rate adaptation hypothesis and highlights the importance of controlling for behavioral confounds in detecting HM.

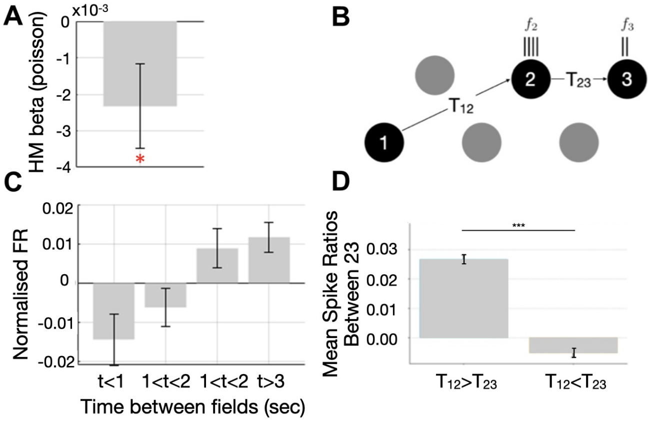

Figure 2: Evidence for firing rate adaptation and hexadirectional modulation. **(A)** Mean Poisson β across grid cells indicates significantly reduced firing during aligned movement (red asterisk, $p < 0.05$). **(B)** Schematic of triplet field visits used to compute adaptation across T_{12} and T_{23} intervals. **(C)** Normalized firing rate was lower when time since previous field visit was short. **(D)** Spike ratio, computed as the normalized difference in firing rates between the second and third fields $((f_i - f_j)/(f_i + f_j))$, was significantly higher when $T_{12} > T_{23}$, consistent with adaptation. *Error bars in A, C, and D indicate standard error of the mean (SEM).*

Reduced firing after short intervals

To further test firing rate adaptation, we delineated the grid fields and measured the time interval between successive visits (Fig. 2B). Firing rate adaptation predicts lower firing when the time since the previous field visit is shorter. We binned spike counts by location, speed, and inter-field interval, and estimated firing rates under Poisson assumptions using the following model:

$$\mathbb{E}(\text{loc}_i, \text{speed}_j, \text{time}_k) = f_{\text{loc}}(i) \cdot f_{\text{speed}}(j) \cdot f_{\text{time}}(k) \cdot t_{ijk}$$

Consistent with spike frequency adaptation timescales (Alonso & Klink, 1993; Giocomo & Hasselmo, 2008; Magistretti & Alonso, 1990), firing rates were lower for short intervals (< 2 s) than long intervals (> 2 s) (Fig. 2C).

We also performed a complementary triplet analysis on three consecutively visited fields. If firing rate adaptation is at play, the firing rate should decrease from the second to the third field when the interval between the second and third visits (t_{23}) is shorter than that between the first and second (t_{12}). In line with this, the ratio of firing between the second and third fields was significantly higher when $t_{23} < t_{12}$ (Fig. 2D).

Shorter intervals during aligned movement

Firing rate adaptation can give rise to hexadirectional modulation if grid-aligned movement leads to shorter time intervals between successive field visits. We found that aligned movement directions were associated with a significantly greater proportion of short inter-field intervals (Fig. 3A). However, the effect was small (<2%), likely due to highly tortuous trajectories, variable speeds, and imperfectly symmetrical grid fields in freely foraging animals (Fig. 3B). Thus, while alignment can predict reduced firing via adaptation, its influence is limited under naturalistic conditions.

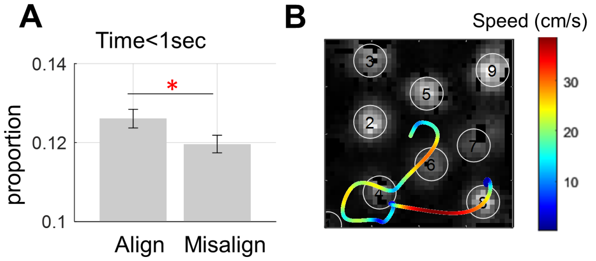

Figure 3: Shorter time between fields when running aligned. **(A)** Proportion of short inter-field intervals (<1 s) was higher during grid-aligned movement. **(B)** Variable speed during an example 20 s trajectory. Error bars are SEM.

Discussion

We present the first evidence of hexadirectional modulation in rodent grid cell firing after controlling for behavioral confounds. Our results support a mechanism in which grid-aligned movement increases the likelihood of short intervals between field visits, triggering firing rate adaptation and reducing firing. This sequence provides a cellular explanation for the six-fold modulation observed in human neuroimaging. Although the effect is modest in natural rodent foraging, it is likely to be stronger in human experiments featuring straight trajectories, uniformly distributed movement directions, and rather constant speeds. These findings inform further optimisation of experimental designs for non-invasive detection of grid cell activity.

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