Active pursuit gates egocentric coding in the retrosplenial cortex

Pearl Saldanha[1,*], Martin Bjerke[2], Benjamin A. Dunn[2,1,†], and Jonathan R. Whitlock[1,4,†,*]

[1]Kavli Institute for Systems Neuroscience and Centre for Neural Computation, Norwegian University of Science and Technology (NTNU), Trondheim 7089, Norway
[2]Department of Mathematical Sciences, Norwegian University of Science and Technology (NTNU), Trondheim 7491, Norway
[†]Joint Supervision
[4]Lead contact
[*]Correspondence: pearl.saldanha@ntnu.no, jonathan.whitlock@ntnu.no

SUMMARY

Spatial navigation is commonly studied in static environments, but adaptive behavior frequently hinges on tracking moving goals in real time. Active pursuit exemplifies this challenge: it is an inherently egocentric spatial behavior requiring continuous localization of a moving goal, yet the neural coding schemes supporting it remain poorly understood. We therefore performed Neuropixels recordings in the retrosplenial cortex (RSC), a hub for egocentric-to-allocentric reference frame transformations, during naturalistic bait-chasing. We identified head-centered target-encoding neurons that were functionally distinct from cells encoding environmental boundaries or static objects. Whereas egocentric boundary encoding was task-invariant, target-coding cells were specific to chasing and showed task-dependent retuning, with enhanced egocentric representation and reduced allocentric head-direction signaling during pursuit. These results demonstrate that classical ego- and allocentric tuning in the RSC coexists with a novel form of coding for moving goals, enabling population activity to adapt flexibly to the dynamic demands of pursuit.

KEYWORDS

egocentric boundary cells, egocentric target cells, spatial navigation, goal-directed behavior, active pursuit, ethological, Neuropixels, population coding, retrosplenial cortex

INTRODUCTION

The retrosplenial cortex (RSC) sits at a crossroads of spatial navigation systems in the brain, serving to translate between allocentric and egocentric reference frames [1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14]. This view has emerged from decades of work demonstrating that RSC neurons encode a rich repertoire of spatial and self-motion variables, including allocentric head direction [12, 15], spatial position [16, 17], rotational head velocity [18], visual motion and landmark information [19, 20, 21], as well as the position of environmental boundaries relative to the individual [7, 8, 22]. However, these insights derive largely from studies in static or minimally structured environments, typically during open field exploration or simple navigation tasks [23, 24, 7, 8, 25]. This ”static spatial” approach has been effective for characterizing how the RSC maps stable layouts, but does not address how such maps guide dynamic ethological behavior. Consequently, little is known of how the RSC supports more rapid forms of navigation that require online updating of spatial information, such as during pursuit and interception of a moving target [26, 27].

This gap is significant because real-world behavior extends far beyond exploration of open spaces. Animals must not only be able to self-localize in an environment, but also track moving goals around them, adjust their actions, and flexibly switch behavioral strategies in real time.

The RSC is well positioned to manage these demands, given its prominent coding of egocentric movement and boundaries [12, 28, 7, 22], as well as visual motion, allocentric space, landmarks and head direction [18, 1, 6]. Anatomically, the RSC receives convergent signals from visual, parietal and hippocampal areas while projecting to motor and premotor cortices, placing it to efficiently transform extrinsic signals from the world into body-centered action commands[29, 30, 10, 9, 2, 31]. Yet how this polyphony of ego- and allocentric signals is coordinated while an animal pursues a moving target is unknown, and it remains unclear whether egocentric coding of the target and environment occur in one overarching network or multiple modular systems.

We addressed these questions using a bait-chasing task in which rats pursue a dynamically moving food reward. In contrast with foraging in a static arena, the pursuit task requires continual updating of the target location while both it and the animal move through space. We performed Neuropixels recordings in the RSC during this naturalistic behavior, which revealed a previously undescribed sub-class of neurons encoding the moving target relative to the head: egocentric target cells (ETCs). These neurons were distinct from egocentric boundary cells (EBCs)– they did not emerge from the EBC population, nor did they encode static objects in the arena. At the population level, ETC activity occupied a task-specific state space during pursuit, while individual ETCs exhibited task-dependent 'remapping', shifting from predominantly allocentric head direction coding during foraging to encoding target location and running speed during chasing. Together, these findings suggest that the RSC employs modular egocentric representations that are re-weighted dynamically with allocentric signals to meet the changing demands of goal-directed behavior.

RESULTS

Egocentric target coding emerges in retrosplenial cortex during pursuit

To characterize spatial and egocentric tuning in the retrosplenial cortex (RSC), we recorded neural activity from four adult female Long Evans rats (3–4 months of age). Neuropixels 1.0 probes were chronically implanted in the dysgranular and granular RSC, yielding a total of $N = 459$ well-isolated single units across all animals and sessions (Figure S1). Animals were tracked using a marker-based 3D motion capture system (STAR Methods), and recorded in an open arena (140 $\times$ 140 $\times$ 50 cm) with a vinyl floor and white cue card on the north wall (Figure 1A). The behavioral tasks included a dynamic bait-chasing task in which animals tracked a dynamically moving food bait over several chasing trials interleaved by pauses to consume the food reward (Figure 1A; Video S1). The other behavioral conditions consisted of spontaneous foraging in the same arena either when it was empty or contained a static object (10 $\times$ 50 cm striped cylinder).

We first asked whether individual RSC neurons encoded the egocentric position of the bait in the chasing task, independent of allocentric location. To do this we constructed egocentric target rate maps (ETRs) by expressing the bait's position in a polar coordinate frame centered on the animal's head and aligned to the direction of the nose (Figure 1B). The egocentric angle (θ) spanned $0°$–$360°$ in $20°$ bins, and egocentric distance (d) was binned in 4.5 cm increments at distances up to 90 cm. For each neuron, the firing rate in each bin was computed as the total spike count divided by occupancy, yielding a head-centered representation of target-related spiking activity (Figure 1B, right). This revealed firing fields that stably encoded the distance and direction of the bait relative to the head (Figure 1C; Video S2).

Neurons were classified as egocentric target cells (ETCs) based on two metrics: the Skaggs mutual information (MI) index, which measures how much information a neuron's firing conveys about the target's egocentric position [32], and tuning stability, measured by the Pearson correlation between alternating 30 s (even-odd) blocks. Statistical significance for both metrics was assessed using 1000 circular spike-time shuffles, and only neurons exceeding the 99th percentile ($pseudo - p < 0.01$) of both null distributions were labeled as ETCs (Figure 1D). This conservative threshold identified 115 of 459 neurons (25.1%), as ETCs, confirming that the RSC contains stable, head-centered representations of moving targets comparable to those described in the posterior parietal cortex (PPC) [7, 33].

Having established ETCs at the single-cell level, we asked whether active pursuit of the target drove a task-specific reorganization of RSC population activity. We assessed this by embedding ETC population activity using Uniform Manifold Approximation and Projection (UMAP)[34] and compared the state spaces occupied during the chasing task vs. spontaneous open field foraging (Figure 1E). Each point in the embedding corresponded to a single time bin and was colored by behavioral task, evincing a clear separation of population dynamics across the two. We quantified the clustering using both the Silhouette score and Dunn index, with both measures confirming separation across contexts (Silhouette score = 0.26; Dunn index = 0.15; STAR Methods), and low (12.1%) overlap between time points acorss tasks. Moreover, linear discriminant analysis (LDA) achieved 91.3% accuracy in classifying behavioral context from ETC population activity (STAR Methods). Together, these results indicate that egocentric target coding is robust in a subset of RSC neurons, and that active pursuit reconfigures the population dynamics of those cells.

Egocentric boundary coding is unaffected by pursuit

We next explored the possibility that pursuit-related activity derived from a reassignment of the RSC's well-characterized egocentric boundary cells (EBCs) [7]. We reasoned that, if pre-existing boundary representations were reassigned to track salient targets during pursuit, ETCs should appear from from the existing EBC population rather than \textit{de novo}. We therefore examined whether egocentric boundary representations re-tuned or were preserved when animals transitioned from the open field task to active pursuit.

We first constructed egocentric boundary rate maps (EBRs) using open field recordings. The angle and distance to the nearest arena boundary were expressed in head-centered coordinates (Figure 2A), with boundary angles binned in 10° increments and distances of up to 60 cm in 5 cm steps. Boundary tuning strength was quantified using the mean resultant length (MRL; STAR Methods), similar to prior classification of EBCs [7]. As with ETCs, classification as an EBC required having MRL and stability measures both exceed the 99th percentile of the shuffled distribution using the same 1000 circular spike-time shuffles as described above (STAR Methods) (Figure 2B).

Using these criteria, 107 of 459 neurons (23.3%) were classified as EBCs, first in the open field then verified for stability in the chasing task. EBCs maintained largely similar tuning to boundary angles and distances across the tasks, though the firing fields of some cells could vary in shape or size (Figure 2C). Analysis of cross-correlograms (CCs) of the rate maps showed, however, that the peak shift of spatial CCs was comparable within- and across tasks (0.49 bin drift for even-odd segments in the open field vs, 0.45 bins across tasks), and the effect size of the difference was small (Cohen's d = 0.26) (Figure 2D).

To determine if switching tasks affected the temporal structure of the EBC network, we examined pairwise temporal relationships across 1,446 EBC–EBC cell pairs from open field and chasing sessions. We computed temporal cross-correlations of binned spike trains for cell pairs

during each task (Figure S2), but the change in peak correlation strength was near zero (median $\Delta = -0.004$; Wilcoxon signed-rank test, $p = 0.18$). Moreover, pairwise coupling strength was strongly preserved across conditions (Spearman $\rho = 0.44$, $p = 3.2 \times 10^{-10}$). Finally, we compared the lag of the temporal cross-correlation peak for EBC cell pairs within open field sessions (even-odd split), and between open field and chasing sessions, and found that the peak-lag varied less across tasks than within-task ($\sim$1.3 s for open field vs. chasing; $\sim$1.8 s for open field even-odd comparison). Thus, we found no evidence that active pursuit caused measurable spatial or temporal reorganization of EBC activity compared to the open field.

We further tested whether boundary and target encoding reflect separable explanatory dimensions in the spiking activity of individual neurons. We did so by quantifying the deviance explained (dDEV) by generalized linear models (GLMs) [35] fit separately to egocentric boundary position and egocentric target position (Figure 2E; STAR Methods). Rather than co-clustering, neurons tended to separate into groups either with stronger boundary-related or target-related explanatory power, supporting a functional dissociation between EBC and ETC tuning dimensions rather than a single, flexible boundary code. Finally, we assessed whether chasing behavior impacted the structure of EBC population activity using the same UMAP analysis performed on ETCs. In contrast to ETCs, time points from the open field and chasing tasks did not form separate clusters in the embedding space (Silhouette score = 0.055; Dunn index = 0.093), indicating stable EBC population dynamics (Figure 2F). This was accompanied by larger overlap across behavioral tasks (39.4% for EBCs vs. 12.1% for ETCs), and lower classification accuracy for behavioral context using LDA (66.2% for EBCs vs. 91.3% for ETCs).

Together, these analyses suggest that egocentric boundary representations in the RSC form a stable spatial scaffold that is maintained across behavioral contexts. The preservation of pairwise temporal correlations and the invariance of EBC population dynamics in state space further suggest that the pursuit-related signals of ETCs do not emerge from a remapping of the underlying boundary code.

Egocentric coding of static objects

Having established that ETCs were not a functional subset of EBCs, we next asked whether RSC contains neurons tuned to static objects within the arena, since it could be that ETCs are just one instance of a more generic code for nearby objects. We assessed this by including object sessions, in which a static object (10 $\times$ 50 cm striped cylinder) was placed in the southeast quadrant of the open field (Figure 3A). We tested for the expression of egocentric object cells (EOCs) using the same egocentric angle and distance framework, as well as the same MI and stability criteria (99th percentile threshold) as with ETCs. We performed this classification on recordings from the subset of animals used in object sessions ($N = 3$ rats, 402 neurons). This identified 44 single units (10.9%) as EOCs (Figure 3B) that stably encoded the position of the static object relative to the animal's head (Figure 3C). Notably, the spatial location of the object encoded by EOCs had no discernable relation to the preferred location of the target in the chasing task (Figure 3C, rightmost column), suggesting that EOCs primarily represented the static landmark rather than generalizing to dynamic targets. The tuning properties of EOCs and ETCs are compared further in the following section.

ETCs, EBCs and EOCs are three distinct egocentric populations

With three egocentric populations identified, we asked whether they followed any anatomical or topographical organization within the RSC, and if they were intermixed. All analyses included

the $N = 402$ neurons for which all three tuning classifications were applied. However, histological reconstruction revealed that EBCs, ETCs, and EOCs were thoroughly intermingled in the RSC, and that they spanned both dysgranular and granular subdivisions with no clear evidence of laminar segregation or topography (Figure 4A)[36]. This suggested that functional distinctions between egocentric populations were not likely attributable to gross regional differences in anatomical inputs.

The extensive intermingling of ETCs, EBCs and EOCs raised the possibility that many of them were the same cells that multiplexed multiple egocentric variables. However, the overlap between EBCs, ETCs, and EOCs was modest (Figure 4B). Of the 402 neurons (101 ETCs (25.1%), 94 EBCs (23.4%), and 44 EOCs (10.9%)), only a minority exhibited more than one type of tuning (4.5% classified as both EBC and ETC; 2.7% as both ETC and EOC; 1.5% as both EBC and EOC; 2.5% were all three), and the rest were assigned to a single egocentric category (Figure 4B). Hypergeometric tests (Figure 4B, right) indicated that the amount of overlap between EBCs and ETCs did not differ significantly from chance ($p = 0.146$), further indicating their mutual functional independence. The overlap between EOCs and ETCs and between EBCs and EOCs was modest but significantly greater chance (EBC–EOC: $p = 0.028$; ETC–EOC: $p < 0.001$), suggesting that representations of static objects share at least some neuronal resources with boundary- and target-related egocentric codes.

The clear segregation of EBCs and EBCs prompted us to test whether target and boundary information could be extracted from RSC population activity simultaneously. Using a Bayesian decoder, we simultaneously reconstructed the animal's egocentric bearing and distance to both the moving target and the arena boundaries from the same RSC ensemble during chasing (Video S3, Figure S3). For the bait, decoded distance had a median absolute error of 0.096 m (mean = 0.123 m) and decoded bearing a median error of 18° (mean = 29.6°), with both measures significantly exceeding chance levels established by circular spike-train shuffles (distance: $p = 0.00167$, $z = 3.14$; bearing: $p < 0.001$, $z = 5.63$). Wall decoding was similarly accurate, with median errors of 0.133 m for distance (mean = 0.18 m) and 12.6° for bearing (mean = 18.8°), both significantly above chance (both $p < 0.001$). That both boundary and target variables could be decoded concurrently from the same population during the same behavioral epochs demonstrates that RSC maintains parallel egocentric representations of static environmental boundaries and dynamic targets during active pursuit.

Finally, we asked if ETCs were not only a distinct population, but that their spatial tuning was exclusive to the pursuit target. We tested whether their spatial preferences generalized to other features by comparing the stability of target-centric rate maps (even vs. odd chasing segments) against rate maps for the same cells referenced to arena boundaries or the static object. This revealed a sharp divergence in coding: whereas tuning was stable for the moving target (mean $r = 0.46$), target-centric rate maps showed a near-zero spatial correlation with arena boundaries (mean $r = 0.06$; $D = 0.77, p < 0.001$) and the static object (mean $r = 0.14$; $D = 0.77, p < 0.001$) (Figure 4C). This demonstrated that ETC activity was anchored selectively to the moving goal rather than reflecting a generalized egocentric preference for environmental features. To determine if the target-centric code required active engagement in pursuit, we introduced the bait at an unattainable height ($> 50cm$) and moved it to mimic the dynamics of a chasing trial. Strikingly, this passive observation failed to evoke the characteristic target-induced firing fields seen during actual pursuit ($N = 44$ ETCs from one animal; $D = 0.55, p < 0.001$; Figure S4), providing direct evidence that ETCs are not merely driven by a moving object in space, but are gated by the animal's active engagement in pursuit.

RSC dynamically reweights behavioral variables during pursuit

Up to this point we established the single-cell tuning properties of ETCs and that their population activity reorganized during pursuit, but it was unclear what this reorganization reflected mechanistically. For example, we did not know whether the separation of ETC population states in UMAP were driven by a global change in excitability or if there was an associated change in the tuning properties of the neurons.

To address the question of state-dependent rate modulation, we classified each neuron as excited, suppressed or indifferent during chasing intervals (STAR Methods). Spike trains were binned at 50 ms resolution, firing rates were normalized by each neuron's mean firing rate across the full session, and we computed a Welch's t-statistic comparing normalized firing rates from chase intervals against 'baseline' rates from open field sessions. Statistical significance was assessed against a null distribution generated by randomly shuffling pursuit intervals throughout the session while preserving the number and duration of bouts (1000 shuffles). This uncovered subsets of chasing-excited and suppressed cells that rapidly and reliably swapped states at the start and stop of chasing in all four animals (Figure 5A). Across the total population ($N = 459$), 69.9% of neurons were chasing-excited, 11.1% were suppressed, and 19% as indifferent (Figure 5B), demonstrating that population state-shifts during pursuit were not due to global up- or down-modulation of spiking. We observed analogous proportions within the ETC population specifically (Figure S5).

The rapid-fire changes in firing activity spoke to strong modulatory effects of pursuit behavior, so we assessed whether chasing influenced *which behavioral variables* the cells encoded. Specifically, we asked whether the GLM-selected features that best explained ETC spiking activity changed between foraging and chasing tasks. We fitted GLMs [35] to predict spike counts from a comprehensive set of behavioral covariates (Figure 5C) that included allocentric head direction, body direction and their derivatives, running speed, egocentric bearing to target, egocentric distance to target, turning velocity, as well as postural features and their derivatives[37, 38]. Forward model selection was performed using ETC recordings from chasing bouts and the open field to identify the covariate set that best explained firing activity in each context. During open field exploration, ETC activity was most frequently explained by a mixture of allocentric head and body direction, as well as self-motion and distance from the walls (Figure 5C left). During chasing, ETC activity was distinguished by prevalent encoding of speed, egocentric bearing to the target, and self-motion variables (Figure 5C right), and a reduction in allocentric head and body direction. A tally of the net differences in features selected across tasks (Figure 5D) highlighted a clear shift away from allocentric head and body direction, and toward self-referential and motoric features, especially egocentric bearing to the target, running speed, egocentric bearing to the walls and postural derivatives.

This shift in tuning properties was associated with an increase in the complexity of tuning structure, with ETC firing reflecting a richer combination of behavioral covariates when animals pursued the moving target (Figure 5E; paired Wilcoxon signed-rank test, $p = 1.52 \times 10^{-5}$, $N = 101$). Importantly, the reweighting of tuning properties and increased model complexity persisted in speed-stratified controls, and was not recapitulated in high-speed epochs in the open field (Figure S6), speaking against modulation solely by locomotor speed.

Together, these results demonstrate that RSC does not merely amplify egocentric signals during pursuit via gain modulation. Instead, ETCs undergo a dynamic reallocation of representational bandwidth, shifting from allocentric landmarks to egocentric variables most critical for immediate action. This task-dependent reweighting provides a mechanistic basis for the population-level shifts in state space, and point to the RSC as an adaptive manager of representational resources.

DISCUSSION

The present study investigated how the retrosplenial cortex guides navigation when animals must simultaneously track a dynamic goal and the layout of the local environment. By recording neural populations during both open field exploration and naturalistic bait-chasing, we found that the RSC accomplishes this using a modular, parallel egocentric organization rather than a single overarching coordinate system. Whereas egocentric boundary cells stably encoded the geometry of the local environment across behaviors, egocentric target representations were recruited flexibly and re-tuned according to the demands of pursuit. This functional organization, where non-overlapping channels track boundaries and targets, points to the RSC as an active manager of representational resources rather than a passive coordinate translator. This enables the RSC to both maintain a stable scaffolding anchored to the environment and re-optimize dynamically for immediate action without mutual interference.

Modular coding of more than open space

The use of open, static recording environments has been critical to establishing the foundations allocentric spatial mapping in hippocampal–entorhinal circuitscitetaube1990head1,muller1987effects,fyhn and identifying a likely role for the RSC in translating between allocentric and egocentric reference frames. More recent work uncovered robust coding of egocentric boundaries, self-motion, head movement and visual flow in the RSC,[28, 7, 8, 18, 25], but again in the context of exploratory behavior or passive movement. The critical step in the present work was additionally engaging the animals in chasing a dynamic goal, and explicitly comparing neural codes during exploratory tasks versus active pursuit. This revealed that egocentric coding in the RSC is not monolithic but modular, comprised of specialized channels that represent diverse features in parallel.

We identified three functionally distinct populations– egocentric target cells (ETCs), egocentric boundary cells (EBCs), egocentric objects (EOCs)– that speak to a layered framework in the RSC. In it, EBCs provide a base layer anchoring the animal to the persistent spatial structure of the environment. Target representations occupy a higher, flexible layer on top of the environmental scaffolding, and are recruited dynamically to meet the demands of pursuit. Although these populations are anatomically intermingled, they differ in tuning properties, population dynamics and task dependence, suggesting that they are not implemented by a single, generic transformation mechanism. Instead, they speak to the RSC maintaining at an additional level of signaling on top of a fixed environmental base as behavioral demands change.

Boundaries as persistent infrastructure

A striking feature of our results was the near-total stability of egocentric boundary representations across behavioral contexts: EBCs maintained highly consistent tuning and pairwise temporal correlations during both open field exploration and active pursuit (Figure 2D)– despite the heightened locomotor and cognitive demands of chasing. In this respect, EBCs may serve a role in anchoring egocentric coordinates analogous to that of HD and grid cells, which map allocentric space universally across environments and behavioral states.[39, 40]

This stability also informs ongoing debates about allocentric–egocentric interactions in RSC. Whereas previous work emphasized tight coupling between HD signals and egocentric representations,[11, 15] our results show that egocentric boundary coding can remain stable even when allocentric variables are down-weighted during pursuit. This suggests that boundary coding may be computed locally within RSC or inherited from parietal or other inputs,[41] rather than

depending on allocentric maps.

Target coding as flexible resource allocation

In contrast to the stable scaffolding of EBCs, ETCs were context-dependent. During pursuit, the majority of ETCs were excited relative to baseline, and their population activity shifted into distinct state spaces (Figure 1E). More revealing than rate modulation was the systematic reweighting of behavioral covariates explaining ETC firing: whereas allocentric head direction dominated during exploration, speed and egocentric target bearing were prioritized during the chase (Figure 5E). This raises the intriguing possibility that a subset of neurons appearing as HD cells in the open field reference distal landmarks during foraging, but dynamically ”re-anchor” to track the moving bait during pursuit. In this view, ETCs are a dynamically re-tunable subclass of cells distinct from stable EBCs. While we cannot confirm this possibility with existing data, future experiments can test directly whether cells transition online from HD-to-ETCs as the bait is introduced.

Regardless of the underlying circuit mechanism, the allo-to-ego reweighting also indicates that ETCs are part of a state-dependent representational regime where neural resources are allocated to the most urgent variables. During pursuit, heading relative to the ”prey” becomes computationally more pressing than heading relative to the world, driving a transition from global spatial mapping to an action-centered view. We interpret this as a form of ’representational bandwidth allocation’ in the RSC. Because neural populations have finite capacity, encoding all spatial variables with equal fidelity at all times would be inefficient. By dynamically adjusting representational priority, the RSC flexibly supports shifting behavioral functions without requiring entirely separate populations for every task. This view is supported by the increased model complexity observed for ETCs during the chase (Figure 5F), reflecting the richer computational demands of target pursuit and interception.

Population dynamics suggest distinct operating regimes

The strongest evidence for a parallel architecture emerged at the population level, where ETC and EBC activity exhibited fundamentally different dynamics in neural state space. When embedded in UMAP space, it was clear that ETC activity occupied a distinct region of state space during the chasing task compared to the open field (Figure 1E), evincing unique population dynamics during pursuit. This separation was robust for ETCs (Silhouette score = 0.257; LDA accuracy = 91.3%), whereas EBC population dynamics remained largely invariant (Silhouette score = 0.055; LDA accuracy = 66.2%). speaking to fundamentally different operating regimes across ETC and EBC populations. This suggests that target tracking engages *specific subsets* of cells in the RSC in a distinct computational mode rather than a region-wide coordinate transformation. This mode—which reflects specific changes in activity patterns rather than global gain modulation—could arise from task-dependent gating of specific inputs or shifts in recurrent dynamics, providing a flexible framework for prioritizing goal-directed action over static environmental mapping.

A family of egocentric codes

The identification of egocentric object cells (EOCs) extends this modular architecture beyond pursuit, revealing a family of egocentric codes tuned to distinct elements the animals may encounter. While EOCs stably represented a static landmark, they failed to generalize to moving targets (Figures 3C, D), reinforcing the functional segregation between stable features and dynamic goals. Our overlap analysis (Figure 4B) showed that EOCs shared significant neural

resources with both the spatially stable EBCs ($p = 0.028$) and the dynamically-targeting ETCs ($p < 0.001$) which were themselves independent ($p = 0.146$). This could suggest that EOCs occupy a representational "middle ground" within the RSC. Whereas the EBC-EOC overlap could reflect the fact that boundaries and objects provide static landmarks encoded once and updated via self-motion, the stronger overlap between EOCs and ETCs may suggest that any discrete "thing" in the environment (whether a static cylinder or a moving bait) recruits a common neural resource for object-centered tracking. In this interpretation, EOCs are a common resource while boundary and target coding remain segregated. This allows the animal to track dynamically moving goals with high precision without interfering with stable environmental maps necessary for global navigation.

Implications for navigation and goal-directed behavior

Together, these results prompt a reconceptualization of RSC function. Rather than acting solely as a coordinate transformation hub, the RSC appears to manage multiple egocentric representations and shift between predominantly allo- and egocentric modes of coding. This permits the maintenance of stable environmental scaffolding while allocating additional resources for behaviorally urgent goals. Such a flexible, modular organization suggests the RSC functions as a dynamic integrator of stable contextual signaling and flexible tracking of immediate goals.

RESOURCE AVAILABILITY

Lead contact

Requests for further information and resources should be directed to and will be fulfilled by the lead contact, Jonathan R. Whitlock (jonathan.whitlock@ntnu.no).

Materials availability

This study did not generate new unique reagents.

Data and code availability

- The data collected in this study will be available from the corresponding author upon reasonable request.
- The custom written code developed for and used in this study will be made available on GitHub upon publication.
- Any additional information required to reanalyze the data reported in this paper is available from the lead contact upon request.

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AUTHOR CONTRIBUTIONS

Conceptualization, P.S., J.R.W. and B.A.D.; methodology, P.S., M.B. and B.A.D.; investigation, P.S.; formal analysis, P.S. and M.B.; writing—original draft, P.S. and M.B.; writing—review & editing, P.S. and J.R.W.; visualization, P.S. and J.R.W.; supervision, J.R.W. and B.A.D.; funding acquisition, J.R.W.

DECLARATION OF INTERESTS

The authors declare no competing interests.

MAIN FIGURE TITLES AND LEGENDS

Figure 1. Egocentric target coding emerges in retrosplenial cortex during pursuit

(A) Schematic of the electrophysiological approach and chasing task. Adult female rats (Long-Evans, 3–4 months old) were implanted chronically with Neuropixels 1.0 probes in the RSC and. The animals freely in a 1.5 m $\times$ 1.5 m arena and chased a food bait (denoted by red dot) attached to a rod the experimenter held and moved.
(B) Schematic for generating egocentric target rate maps. Left: the target position is expressed as the angle θ and distance d from the head of the animal. Right: schematic egocentric target rate map showing the color-coded firing rate of a cell coding for the target at that specific angle and distance (up to 90 cm, indicated by rings).
(C) Examples of three egocentric target cells (ETCs). Each row depicts (left to right): the path of the animal in the area with spike positions (colored by head direction) overlaid; egocentric target occupancy; rate map showing the preferred target location of the cell relative to the head (whole session average); rate maps with data from odd- or even-minutes only; cross-correlogram between odd- and even-minute maps showing the angle (x-axis) and distance (y-axis) of the shift of the correlation peak (red star).
(D) Selection criteria for ETCs. Left: histogram of pseudo-p values for within-session (even-odd minute) stability of the egocentric rate maps; orange bars indicate neurons exceeding the 99th percentile of the shuffled distribution (pseudo-$p < 0.01$). Middle: histogram of pseudo-p values for mutual information (MI); neurons above 99th percentile of the shuffled data again shown in orange. Right: joint distribution of stability and MI pseudo-p values; orange points indicate neurons classified as ETCs (pseudo-$p < 0.01$ for both criteria).
(E) UMAP embedding of ETC population activity. Each point represents a single time bin, colored by behavioral context (blue: open field; orange: chasing). The population activity occupied distinct regions of state space across contexts (Silhouette score = 0.257; Dunn index = 0.146; LDA accuracy = 91.3%; STAR Methods), indicating robust differences in population between the two session types.

Figure 2. Egocentric boundary cells remain stable during pursuit

(A) Schematic for generating egocentric boundary rate maps (EBRs). Left: recording arena in world-referenced (allocentric) coordinates with head direction indicated by color. Middle: same arena shown in head-referenced (egocentric) coordinates, illustrating animal-referenced boundary positions from different locations in the arena. Right: example egocentric boundary rate map (EBR) showing firing rate as a function of the angle and distance (20–60 cm rings) of the arena wall relative to the head.
(B) Selection criteria for EBCs. Upper left: histogram of pseudo-p values for within-session stability. Lower left: histogram of pseudo-p values for mean resultant length (MRL); BLUE bars indicate cells exceeding the 99th percentile of shuffled data for each measure. Right: joint distribution of MRL and stability pseudo-p values. BLUE points (lower left corner) indicate neurons classified as EBCs.
(C) Examples of egocentric boundary cells (EBCs). Each row for each neuron shows (left to right): the path of the animal with spikes overlaid; egocentric boundary rate maps (EBRs) for open field and chasing sessions; odd- and even-minute EBRs from each task; cross-correlograms (CCs) between odd- and even-minute EBRs. Cross correlogram peaks (red stars) shifted minimally both between- and within-task.

(D) The frequency of differences observed in CC peak shifts between even-odd segments in the open field versus the open field and chasing tasks (i.e. CC peak shifts within- vs. across tasks). The distribution of differences is centered near zero (net difference 0.04).
(E) Scatter plot of deviance explained (dDEV) for EBC versus ETC tuning. Orange points correspond to neurons with greater ETC than EBC tuning ($n = 74$), and blue points are neurons with greater EBC than ETC tuning ($n = 93$). The separation of categories along the diagonal demonstrates the predominant segregation between ETCs and EBCs as classified with a GLM (Methods).
(F) UMAP embedding of EBC population activity. Blue dots show open field time points; orange dots show chasing. In contrast to ETCs, which segregate in UMAP space, EBC population activity is intermingled across behavioral contexts (Silhouette score = 0.055; Dunn index = 0.093; LDA accuracy = 66.2%), indicating stable population dynamics.

Figure 3. Egocentric object cells encode the location of a static object relative to the head

(A) Schematic showing the open field with static object in it. The egocentric position of the object is computed as the angle θ and distance d relative to the animal's head direction.
(B) Classification statistics of egocentric object cells (EOCs). Left: histograms of pseudo-p values for within-session stability (top) and spatial mutual information (MI, bottom), with orange bars indicating neurons exceeding the 99th percentile of their respective shuffled distributions. Right: joint distribution of MI and stability pseudo-p values; orange points indicate neurons classified as EOCs (p ¡ 0.01 for both criteria).
(C) Example egocentric object cells (EOCs). Each row shows, from left to right: allocentric trajectory with spike positions colored by head direction and the object located at the open circle; egocentric object trajectory around the animal; egocentric coding of object location averaged for the whole session (rings indicate 30–90 cm distances); odd- and even-minute rate maps demonstrating stable tuning; cross-correlogram for odd- and even-minute rate maps (red stars denote CC peak); rate map showing the response of the same cell relative to the moving target during the chasing task (rightmost column). The firing patterns relative to the object do not carry over to the moving target during chasing.
(D) Left: UMAP embedding of EOC population activity showing that neural population states during the the object session (green dots) segregated largely from population activity during the chasing task (orange dots) (Silhouette score = 0.055; Dunn index = 0.093; LDA accuracy = 66.2%). Right: EBC population activity from the same object and chasing sessions was substantially less separated (Silhouette score = 0.055; Dunn index = 0.093; LDA accuracy = 66.2%), suggesting that EOCs distinguish the object and chasing conditions whereas EBCs do not.

Figure 4. Three distinct but overlapping egocentric populations in RSC

(A) Homogeneous distribution of egocentric cell types in the RSC. Left: schematic coronal section with reconstructed recording depths of each cell type, summed across animals, overlaid. The schematic does not illustrate exact medial-lateral probe placement, and functional cell types are separated in columns for illustration purposes only (EBCs are shown as blue circles, ETCs as orange squares, EOCs as green triangles. Recording sites spanned dysgranular RSC (dRSC) and granular RSC subdivisions (gRSCa, gRSCb, gRSCc), and grazed the corpus callosum (cc), leading to a drop in total cell yield from ca. 1900-2500 microns deep. Right: density distributions of each cell type as a function of distance from the cortical surface. All three populations were

distributed throughout RSC with no evidence of superficial or deep layer segregation. (See Figure S1 for histological micrographs and recording probe tracks for individual animals; cc, corpus callosum; SC, superior colliculus).

(B) Egocentric cell types consisted mainly of different groups of cells. Left: Venn diagram showing the proportions of neurons classified as EBCs (23.4%), ETCs (25.1%), and EOCs (10.9%) out of $N = 402$ total neurons from 3 rats. Percentages indicate both exclusive and overlapping classifications. Right: pairwise overlap frequency compared to overlap expected by chance. The observed EBC∩ETC overlap was not greater than chance levels (hypergeometric test, $p = 0.146$, n.s.). ETC∩EOC values were slightly but significantly larger than chance levels ($p < 0.001$), and EBC∩EOC overlap was marginally larger than chance levels ($p = 0.028$).

(C) ETC coding of target locations does not generalize to boundaries or objects. Left: cumulative distributions of spatial correlation coefficients for ETCs ($n = 101$) when rate maps for the target were measured against themselves (even- vs. odd-minutes during the chasing task, orange squares) vs. arena boundaries (blue circles). Correlation values for the target vs. boundaries was substantially lower than for the target for even vs. odd minutes (mean $r = 0.46$), indicating that ETCs did not also encode walls. Right: ETC tuning to the target in even- vs. odd-minutes (orange squares; mean $r = 0.14$) was also significantly better correlated that compared rate maps for the object (green triangles; mean $r = 0.14$), demonstrating that ETC coding of the target did not generalize to the stationary object.

Figure 5. Dynamic modulation of RSC neural activity and tuning during pursuit

(A) Chasing-excited and suppressed cells switched states sharply at the onset and offset of chasing vs. consumatory rest periods. Each graph shows the session-normalized mean firing rate for chasing-excited (red line) and chasing-inhibited (blue) neurons (Methods) in each of four rats, with chase intervals indicated in shading and rest intervals in white.

(B) Pie chart summarizing the fractions of chase response types across the total population of RSC cells. Nearly 70% of all RSC neurons were significantly up-modulated during chasing bouts, 11.% were supressed, and 19% were indifferent.

(C) Bar graph showing the frequency with which specific behavioral features were included in the best-fitting GLMs for the ETC cell population ($N = 101$). Left: in the open field, the most common features explaining ETC activity were allocentric head direction, change in head direction and change in body direction, followed by self-motion and egocentric bearing to the nearest wall. Right: same, but for chasing sessions, showing a marked increase in the prominence of speed, relative head angle, and self-motion covariates, and a decrease in allocentric head and body direction.

(D) Left: the change in covariates selected by the GLM between contexts (chasing minus open field). Green bars indicate covariates included more frequently during chasing; red bars indicate covariates less frequently included. The two most widely added features were relative angle of the bait and running speed, and the two most frequently lost features were allocentric head and body direction. Right: the distribution of best-model complexity k (number of covariates) for ETCs was significantly lower in the open field (blue) than in the chasing task (orange) (paired Wilcoxon signed-rank test: $p = 1.52 \times 10^{-5}$, $n = 101$).

STAR Methods

Experimental model and study participant details

Animals

Four adult Long Evans rats (female, 3-4 months of age) were used in this study (ID 28685-*Arwen*, 29865-*ToothMuch*, 29373-*PreciousGrape*, 30146-*MimosaPudica*). Animals were handled daily for ¿1 week prior to experiments. After handling, each animals was habituated to a black 140 $\times$ 140 $\times$ 50 cm recording arena for several days before surgery and recordings. The recording environment consisted of the recording box, a white cue card on the north wall, and recording hardware setup beyond the south wall. The recording environment remained consistent across all experiments in the study. The animals were screened for chasing behavior prior to experiments, so were already familiar with the task during recordings. The behavioral conditions consisted of open field foraging in the empty arena, in which the animal foraged for corn puff crumbs sprinkled on the arena floor by the experimenter. Chasing sessions were in the same area, and included a fishing rod with a food reward (a corn puff, either bare or smeared with Oreo cream or Nutella) suspended on the end of a string, and moved pseudorandomly by the experimenter to evoke chasing. A retroreflective spherical marker was attached 9 cm above the bait for tracking during chasing trials. The object session was the same as open field foraging. Recording sessions were approximately 20 minutes, and chasing sessions were repeated until achieving ¿20 minutes of active chasing behavior.

All experiments were performed in accordance with the Norwegian Animal Welfare Act, the European Convention for the Protection of Vertebrate Animals used for Experimental and Other Scientific Purposes, and the local veterinary authority at the Norwegian University of Science and Technology. All experiments were approved by the Norwegian Food Safety Authority (Mattilsynet; protocol ID 25094). The study contained no randomization to experimental treatments and no blinding. Sample size (number of animals) was set a priori to at least four, given the expected cell yield necessary to perform unbiased statistical analyses.

Method details

Surgery

Animals were anesthetized initially with 5% Isofluorane vapor mixed with oxygen and maintained at 1-3% throughout the surgical procedure. When anesthetized, they were placed in a stereotaxic frame and injected subcutaneously with Metacam (2.5mg/kg) and Temgesic (0.05mg/kg) as general analgesics. Body temperature was maintained at 37℃ with a heating pad. A local anesthetic (Marcain 0.5%) was injected under the scalp before the first surgical incision was made, followed by clearing the skull of skin and fascia. Following the clearing of tissue, adjustments were made to make sure the skull was leveled before a high-speed dental drill was used to open a craniotomy over the retrosplenial cortex (Kulzer GmbH, Germany). A single jeweler's screw was inserted over the cerebellum as a ground and reference screw for the probe implant, connected with a silver wire and sealed with SCP03B (silver conductive paint). Animals were subcutaneously administered fluids at the start, at the half-way point of surgery and immediately post-surgery.

A single-shanked silicon probe (Neuropixels version 1.0, IMEC, Belgium) was stereotaxically lowered in the right hemisphere of two animals and left hemisphere for other two animals at coordinates AP: -5.4-6.0, ML: 0.7-0.9, DV: 4.9-6.0. The craniotomy and unimplanted length of the probe were enclosed with a silicone elastomer (Kwik-Sil, World Precision Instruments Inc., USA). After allowing the silicone elastomer to harden, a thin layer of Super-Bond (Sun Medical Co., Ltd.,

Japan) was applied to it and the skull to increase bonding strength between the skull surface and dental acrylic. To minimize light induced electrical interference during the experiments the first layer of dental cement was dyed black and care was taken to completely cover any light-sensitive parts of the probe. Undyed cement was further applied to secure the probe. A custom 3D printed skull cap was used to encase the implant outside the brain, serving both as a protective housing and as a base for attaching the rigid body for tracking the head in 3D during recordings. The cap was secured to the skull with black-dyed cement. Once the cement had cured, sharp edges were removed with the dental drill at the end of surgery. Following surgery, the animals were placed in a heated (32℃) chamber to recover. Once awake and ambulating, they were returned to their home cage and allowed food and rest before data collection commenced.

Recording procedures

Electrophysiological recordings were performed using Neuropixels 1.0 acquisition hardware: A National Instruments PXIe-1071 chassis and PXI-6133 I/O module for recording analog and digital inputs. Implanted probes were operationally connected via a headstage circuit board and interface cable above the head. An elastic string was used to counterbalance excess cable, allowing animals to move freely through the entire recording area during recordings. Data acquisition was performed using SpikeGLX software (SpikeGLX, Janelia Research Campus), with the amplifier gain for AP channels set to 500x, LFP channels set to 250x, an external reference and AP filter cut at 300 Hz. Spike activity was recorded and collected from bank 0, i.e., the most distal 384 recording sites. Behavioral experiments and neural recordings were performed 532 a minimum of 2.5 hours after animals had fully recovered from surgery.

3D tracking and synchronization

Animals had circular marker cutouts of 3M retro-reflective tape (OptiTrack, catalog no. MSC 546 1040; Natural Point Inc., Corvallis, OR, USA) placed on their body at the neck, back and tailbase, which were shaved during surgery. To capture head rotations we used a rigid body with four retro-reflective spheres, size 7.9 mm, 9.5 mm and 12.7 mm (OptiTrack, catalog no. MKR079M3-10, MKR095M3-10 550 and MKR127M3-10, respectively) integrated with the skull cap. The markers were tracked retroreflectively using 8 near-infrared (NIR) recording cameras (7 NIR, and one black/white camera for referencing; Flex 13 cameras, Part #: FL13, OptiTrack, Oregon, USA) at 120 FPS in optical motion capture software (Motive version 2.2.0; OptiTrack).

Tracking and neural data streams were syncrhonized using three LEDs affixed to the sides of the recording arena, and captured by the motion capture system. The LEDs were controlled by an Arduino Microcontroller with C++ code to generate irregularly timed digital pulses (250 ms duration of each flash; random 250 ms $\leq$ IPI ≤ 1.5 sec) transmitted to acquisition systems throughout the recordings. Individual markers were labeled using built-in functions in Motive on raw .tak files. After recordings were completed, 3D data was reconstructed by creating a marker set for the three trunk markers, a rigid body construction for the capture of the animal's head and a rigid body was constructed to capture the synchronizing LEDs. Using Motive's built-in "autolabel" function, markers were identified and labeled, with remaining unlabeled markers manually labeled by the experimenter. Errors in marker assignment were also corrected manually. For frames where markers were occluded, tracked points were reconstructed using in-built functions in the Motive software. Different modes of reconstruction ("cubic") were used depending on the duration of occlusion, the behavior of the animal, marker identity and the potential for validating the reconstruction. After reconstruction and labeling of the markers, the positional information from each session was exported as a .csv-file. A custom-written Python script was used to process and align tracking information with the synchronization stream into a .pkl file. This file was

then loaded in a custom graphical user interface (GUI) to construct a head coordinate system and merge the tracking data with spiketimes into a final .mat file. These same procedures are described in further detail in ([38]). The minimum threshold for determining the animals' running speed was 5 cm/s.

Perfusion and probe placement verification

Upon completion of the recordings, each rat was euthanized with an overdose of Isoflurane. This was followed by intracardial perfusion using saline and 4% paraformaldehyde. The probe shank was kept inside the brain tissue and the entire skull was kept in 4% paraformaldehyde for 24 hours. The next day, the brain was then carefully extracted from the skull and transferred to a solution of 2% dimethyl sulfoxide (DMSO, VWR, USA) for cryoprotection for a period of one to two days before cryosectioning. Following cryoprotection, the brain was frozen and sectioned coronally into three series of 40 μm slices using a freezing sliding microtome (Microm HM-430, Thermo Scientific, Waltham, MA). The first series of sections was directly mounted onto Superfrost slides (Fisher Scientific, Göteborg, Sweden) and subsequently stained with Cresyl Violet. All brain sections were digitized using a digital scanner and appropriate illumination wavelengths with scanning software provided by Carl Zeiss AS (Oslo, Norway). The resulting images were then visualized using ZEN (blue edition) software. Post-hoc probe reconstruction was done using HERBS[36].

Chase interval annotation

Chase intervals were identified post-hoc during visual inspection of video recordings. An interval was classified as "chase" when the animal actively reoriented and initiated movement toward the bait, as indicated by locomotion direction, head orientation and sustained forward movement. After the animal caught the bait, the rod was removed from the arena; between-chase rest periods consisted of the animal consuming the food reward, as well as stationary grooming, rearing and undirected exploration. Two experimenters independently annotated and agreed on chase interval boundaries.

Egocentric rate map construction

Egocentric rate maps were constructed by computing the position of spatial features (arena boundaries, bait, or stationary objects) relative to the position and direction of the animal's head at each time bin. Spike data were binned at 8.33 ms ($\sim$120 Hz). For each time bin, egocentric angle θ was computed as the angular offset between the feature and the animal's head direction, and distance d was computed as the Euclidean distance from the animal's head to the feature. For egocentric boundary cells (EBCs), rays were cast at 1° intervals from $-180°$ to $179°$ relative to head direction, and distance to the nearest wall was computed along each ray. These distances were then averaged into 10° bins, yielding 36 angular bins.

For all egocentric cell types, rate maps were computed by accumulating occupancy and spike counts into a polar grid. Angular bins spanned $-180°$ to $180°$ in 10° increments (36 bins). Distance bins spanned 0–60 cm in 5 cm increments for EBCs (12 bins) and 0–90 cm in 4.5 cm increments for ETCs and EOCs (20 bins). Firing rate in each bin was computed as spike count divided by occupancy time. Rate maps were smoothed using a 2D Gaussian kernel (kernel size: 3 $\times$ 3 bins; standard deviation: 1.5 bins). Bins with fewer than 50 frames of occupancy ($\sim$0.4 s) were excluded from analysis.

Quantification and statistical analysis

Skaggs information index calculation

To test whether a particular neuron was tuned to a dynamic target or not, we used the Skaggs information index [32, 42]. The information index is an approximation of the mutual information metric and measures the amount of information the firing of one neuron carries relative to some observed variable. The mutual information between two variables X and Y, $\mathsf{I}(X;Y)$, is given as

$$\mathsf{I}(X;Y) = \sum_{x,y} P_{XY}(x,y) \log_2 \frac{P_{XY}(x,y)}{P_X(x)P_Y(y)}.$$

We let X denote the event that the animal is currently at a distance d from the target, with an angle θ between the target and the animal's current heading direction, and let Y be the event that a neuron fires exactly f spikes at the current time. Considering binned spike data, the approximation then allows the information index to be written as

$$\mathsf{SI} = \sum_{i,j} P_{ij} \frac{f_{i,j}}{\bar{f}} \log_2 \frac{f_{i,j}}{\bar{f}},$$

where $P_{i,j}$ denotes the probability of being in the i,j-th binned distance-angle bin, $f_{i,j}$ denotes the calculated firing rate in the i,j-th bin and $\bar{f}$ denotes the overall mean firing rate of the neuron.

Cell type classification using the Skaggs information index and stability

The Skaggs mutual information index was used as one of two tuning metrics for classifying egocentric target cells (ETCs) and egocentric object cells (EOCs). For ETCs, egocentric distance–angle coordinates were defined relative to the moving target during chasing bouts; for EOCs, coordinates were defined relative to the stationary object during object sessions.

Tuning stability was quantified as the Pearson correlation coefficient between egocentric rate maps generated from alternating 30-s blocks (even-odd split) of the relevant behavioral epochs, ensuring that the spatial structure of the head-centered firing field was preserved throughout the recording session.

The significance thresholds for both measures were established for each neuron by circularly shifting each cell's binary activity vector (1000 iterations) to generate a null distribution against which observed values were converted to pseudo-p values. Neurons were classified as ETCs or EOCs only if both the Skaggs mutual information index and the Pearson stability correlation exceeded the 99th percentile of their respective shuffled distributions ($pseudo - p < 0.01$). The shuffle and stability procedures were identical to those used for EBC classification, with the Skaggs mutual information index replacing mean resultant length (MRL) as the primary tuning metric for target- and object-related activity.

Egocentric boundary cell classification using mean resultant length

The mean resultant length (MRL) was used as our tuning metric for classifying neurons as egocentric boundary cells (EBCs). The MRL measures the concentration of firing around a preferred egocentric angle and is computed as:

$$\mathsf{MRL} = \frac{|\sum_{j,k} f_{j,k} e^{i\theta_k}|}{\sum_{j,k} f_{j,k}},$$

where $f_{j,k}$ denotes the firing rate in the j, k-th distance-angle bin, θ_k is the k-th egocentric angular bin in radians, and i is the imaginary unit.

Statistical significance of the MRL was assessed using a shuffle procedure, where the spike train was circularly shifted by a random offset (minimum 30 s) and the MRL was recomputed. This was repeated 1000 times to generate a null distribution, and a neuron passed the tuning criterion if its observed MRL exceeded the 99th percentile of the null distribution ($p < 0.01$).

As with ETCs and EOCs, temporal stability was assessed by splitting the session into alternating 30-second blocks (odd and even) and computing separate rate maps for each half. The Pearson correlation between the odd and even rate maps was computed, excluding bins with low occupancy, and a shuffled null distribution was generated by circularly shifting spike trains and recomputing the odd-even correlation (1000 shuffles). A neuron passed the stability criterion if its observed correlation exceeded the 99th percentile of the null distribution ($p < 0.01$). Neurons were classified as EBCs if they passed both the MRL and stability criteria (sequential testing following Alexander et al.[7]).

GLM covariate analysis

We fit generalized linear models (GLMs) with a Bernoulli family and logit link function to predict binarized spike counts (spike vs. no spike per time bin) from behavioral covariates (as done in Mimica et al.[38]). Covariates included allocentric head direction and body direction and their first temporal derivatives, running speed and its derivative, angular velocity of body and head, egocentric bearing to target (relative head angle), egocentric distance to target (relative distance), posture variables[37, 38] including head azimuth, head roll, head pitch, back pitch, neck elevation, and their temporal derivatives.

For each neuron, we used a forward model selection[43] procedure to identify the best-fitting combination of covariates. Starting from a null (intercept-only) model, covariates were iteratively added one at a time. At each iteration, all remaining covariates were tested as candidate additions. Each candidate model was evaluated using 10-fold cross-validation, in which each fold contained temporally distributed sub-blocks to reduce autocorrelation effects. Model parameters were estimated using L-BFGS optimization with L1 regularization ($\lambda = 0.0001$, $\alpha = 1$).

For each fold, we computed the log-likelihood ratio (LLR) between the candidate model and the null model, normalized by the number of spikes in the test set (LLR per spike). A candidate model was considered significantly better than the current best model if a one-sided Wilcoxon signed-rank test comparing cross-validated LLR-per-spike scores across folds yielded $p < 0.01$. Among all candidates meeting this significance threshold, the model with the highest mean LLR per spike was selected as the new best model. The forward selection procedure continued until no candidate model produced a significant improvement, at which point the current best model was returned.

This procedure was performed separately on the same cells for open field and chasing sessions to allow direct comparison of covariate importance across behavioral contexts. Model complexity k was defined as the number of covariates in the best-fitting model. Covariate usage frequencies and model complexities were compared between open field and chasing sessions using paired Wilcoxon signed-rank tests.

Deviance explained (dDEV)

To quantify the relative contribution of boundary- versus target-tuning for each neuron, we computed the deviance explained (dDEV) from GLMs. For each neuron, we fit separate GLMs predicting spike counts from (1) egocentric boundary position (angle and distance to nearest wall)

and (2) egocentric target position (angle and distance to bait). Deviance explained was computed as:

$$\text{dDEV} = \frac{D_{\text{null}} - D_{\text{model}}}{D_{\text{null}}} \times 100,$$

where D_{null} is the deviance of an intercept-only model and D_{model} is the deviance of the full model including spatial covariates.

Spatial cross-correlations between cell types

To quantify the similarity of the different types of egocentric tuning (relative to boundaries, chasing bait or object), we computed pairwise Pearson correlations between egocentric rate maps. For neurons classified as multiple cell types (e.g., both EBC and ETC), we correlated the EBC rate map (computed from open field data with respect to boundaries) with the ETC rate map (computed from chasing data with respect to the bait). Within-session stability was computed by generating correlations between rate maps for odd and even segments for a given session.

Correlations were computed only over bins with sufficient occupancy in both rate maps (minimum 50 overlapping bins). Rate maps were smoothed with a Gaussian filter ($\sigma = 1$ bin) before measuring correlations.

Temporal cross-correlations between cell pairs

To examine functional coupling between EBCs, we computed pairwise cross-correlations of binned spike trains. Spike trains were binned at 150 ms and detrended using a high-pass filter (window = 50 bins, polynomial order = 2) to remove slow drift. Cross-correlation coefficients were computed for lags spanning $\pm$60 bins ($\pm$9 s):

$$r(\tau) = \frac{1}{n - |\tau|} \sum_t \frac{(x_t - \bar{x})(y_{t+\tau} - \bar{y})}{s_x s_y},$$

where x and y are the normalized spike trains, $\bar{x}$ and $\bar{y}$ are their means, s_x and s_y are their standard deviations, and τ is the lag.

Cross-correlations were computed separately for open field and chasing sessions to assess whether functional coupling was preserved across behavioral states.

UMAP projections

UMAP (Uniform Manifold Approximation and Projection)[34] was employed to visualize population activity in a low-dimensional space. First, binned spike trains were smoothed using a Gaussian filter with standard deviation $\sigma = 3$ bins and converted to firing rates (Hz). The smoothed spike trains were then downsampled to every 12th bin. Before UMAP embedding, firing rates were rank-normalized within each neuron using a Gaussian inverse transform, and principal component analysis (PCA) was applied to reduce dimensionality to 6 components.

UMAP was then applied with parameters: n_neighbors = 100, min_dist = 0.8, metric = 'euclidean'. For comparing behavioral modulation in different tasks, UMAP was fit on concatenated data e.g. from both open field and chasing sessions, with time points colored by session type. Separate UMAP embeddings were computed for different cell populations (EBCs only, ETCs only) to assess how the different task conditions affected the state spaces occupied by each population.

UMAP quantification

To quantify the degree of separation between behavioral contexts in UMAP embeddings, we computed several complementary metrics. The Silhouette score measures how similar each point is to its own cluster compared to other clusters, ranging from -1 (poor clustering) to $+1$ (perfect clustering):

$$s(i) = \frac{b(i) - a(i)}{\max(a(i), b(i))},$$

where $a(i)$ is the mean distance from point i to other points in the same cluster and $b(i)$ is the mean distance to points in the nearest other cluster.

The Dunn index (DI) measures overall clustering quality; a higher Dunn index corresponds to tighter clustering in the data, with 0 being no clustering. For it, centroids were first calculated for every behavioral cluster. Distances between each point within a behavioral cluster and the cluster's centroid (intra-cluster distances) and the distances between centroids of different clusters (inter-cluster distances) were measured. The DI was then calculated as the ratio between the minimum inter-cluster distance and the maximum intra-cluster distance (as defined above). We computed a robust variant using the 5th percentile of inter-cluster distances to reduce sensitivity to outliers.

Context overlap was quantified as the percentage of time bins in the UMAP embedding that were closer to the centroid of the opposite behavioral context than to their own. Linear discriminant analysis (LDA) and support vector machine (SVM) classifiers were trained to predict behavioral context from UMAP coordinates using 5-fold stratified cross-validation.

Cell type overlap analysis

To quantify overlap between cell type classifications, we computed the percentage of neurons meeting criteria for multiple cell types (e.g., both EBC and ETC). Statistical significance of overlap was assessed using hypergeometric tests, which compare the observed overlap to the expected overlap under the null hypothesis of independent classification:

$$P(X \geq k) = \sum_{i=k}^{\min(K,n)} \frac{\binom{K}{i}\binom{N-K}{n-i}}{\binom{N}{n}},$$

where N is the total number of neurons, K is the number of neurons in class A, n is the number of neurons in class B, and k is the observed overlap.

FIGURES

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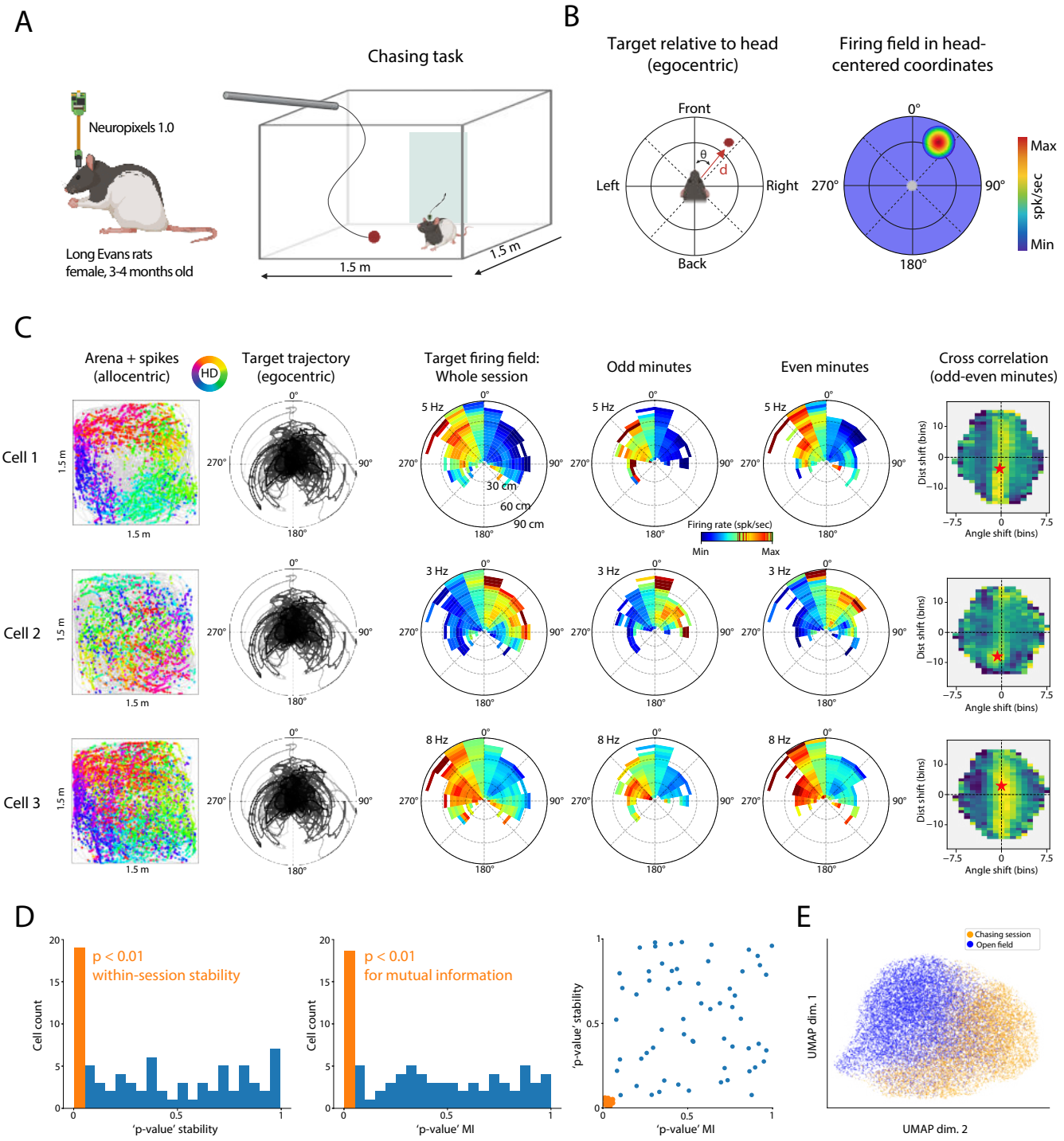

Figure 1: Egocentric target coding emerges in retrosplenial cortex during pursuit.

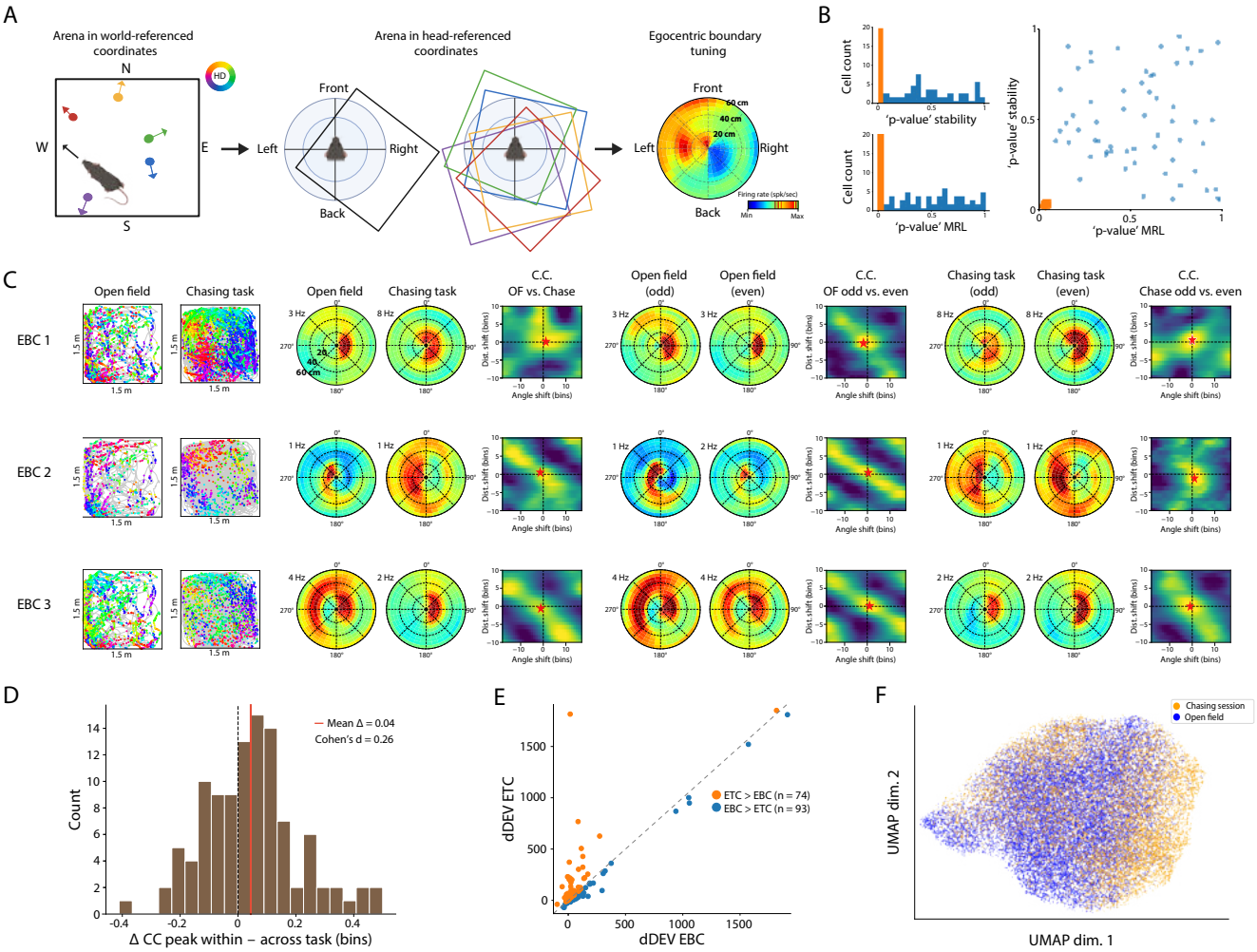

Figure 2: Egocentric boundary cells remain stable during pursuit.

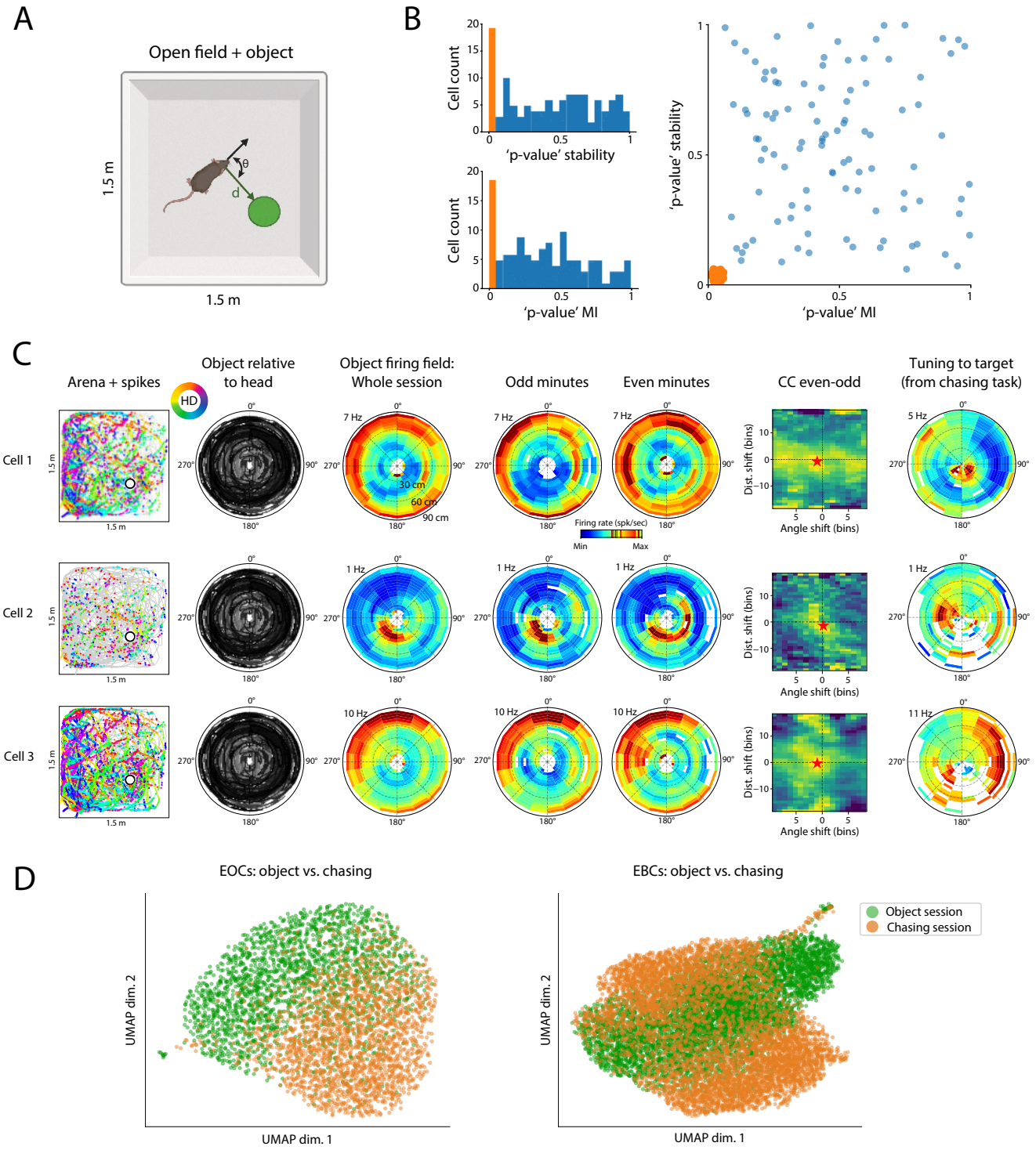

Figure 3: Egocentric object cells encode static targets

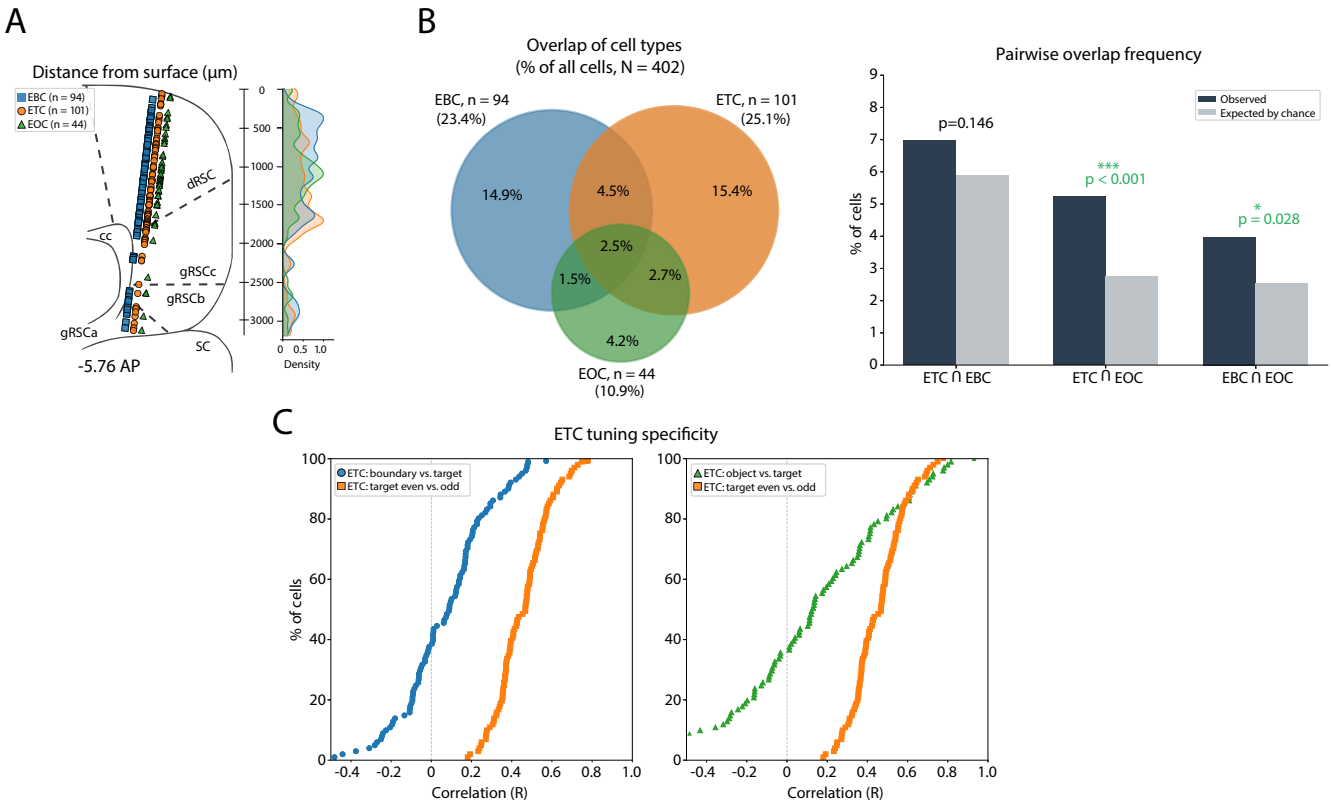

Figure 4: Three distinct but overlapping egocentric populations in RSC

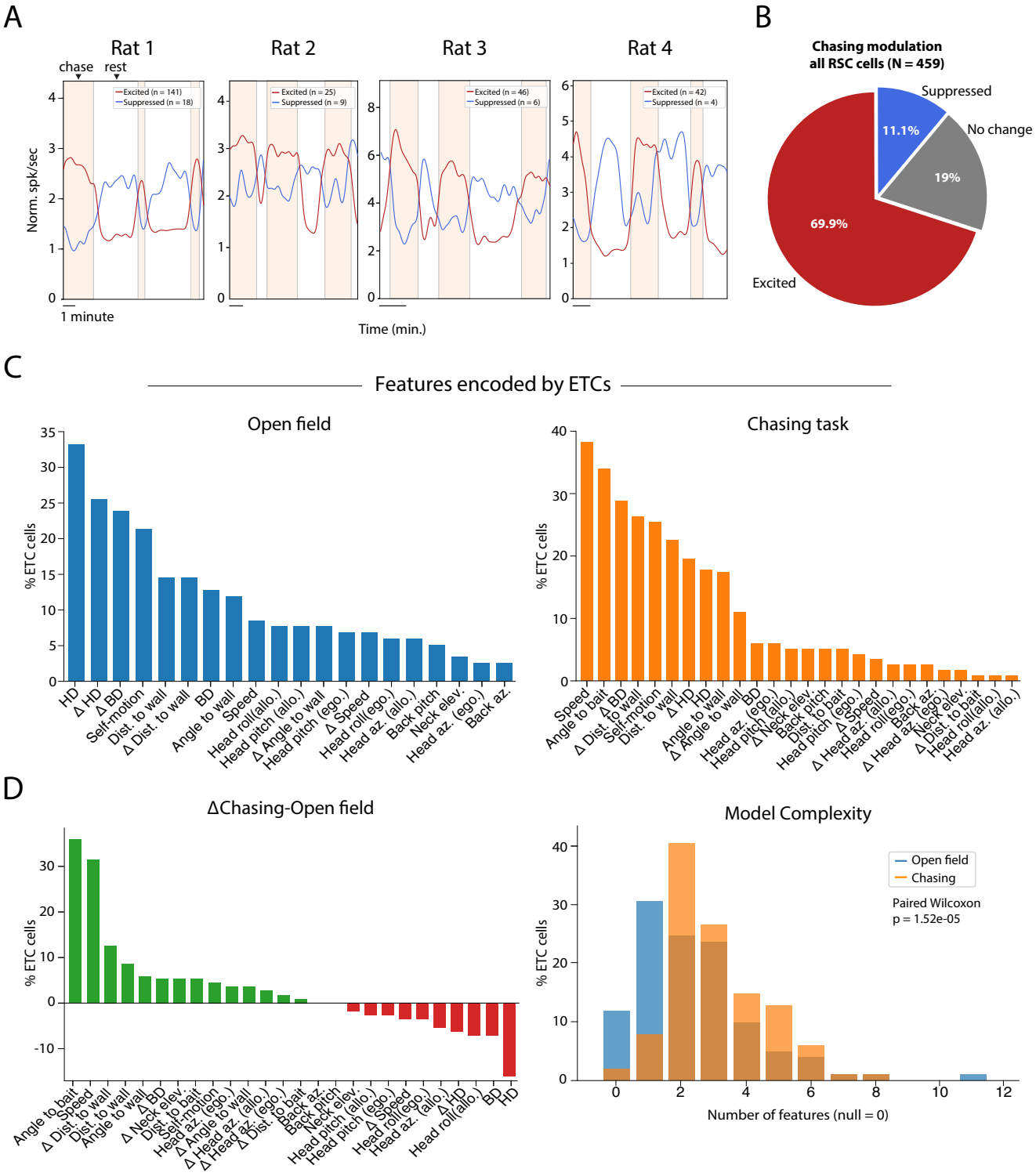

Figure 5: RSC dynamically reweights behavioral variables during pursuit

SUPPLEMENTAL VIDEO LEGENDS

Supplementary Video 1. Raw footage of bait-chasing behavior. Black-and-white video recording of a representative active chasing bout. The animal pursues a moving bait attached to a manually operated fishing rod within the open arena. The bait trajectory was controlled by the experimenter to elicit sustained pursuit behavior while maintaining the bait within the animal's visual field. Video is shown at real-time playback speed.

Supplementary Video 2. Egocentric visualization of target-tuned spiking activity. Animal-centered ("Mr. Stuck") representation of chasing behavior, in which the animal's position and heading are fixed at the center of the frame while the environment and bait position move relative to the animal. The bait location is indicated by a red dot. Audio represents spike events from a representative egocentric target cell (ETC), with each spike rendered as an audible click. Note that spiking activity increases when the bait is positioned to the right of the animal's heading, at approximately 50° eccentricity, demonstrating egocentric tuning to target direction.

Supplementary Video 3. Simultaneous Bayesian decoding of egocentric target and boundary directions. Real-time visualization of population-level decoding during a representative chasing bout. The animal's trajectory is shown in pink, with cardinal directions labeled. The red dot indicates the true bait position; the purple vector shows the decoded egocentric target direction. The blue vector indicates the true direction to the wall at any given time-point, while the green vector shows the decoded egocentric boundary direction. The black arrow denotes the animal's head direction. Inset text displays instantaneous decoding accuracy, including true and decoded bait distance and wall distance. Decoded directions were computed from simultaneous RSC population activity, demonstrating that egocentric representations of targets and boundaries can be read out in parallel.

Supplementary Figure Titles and Legends

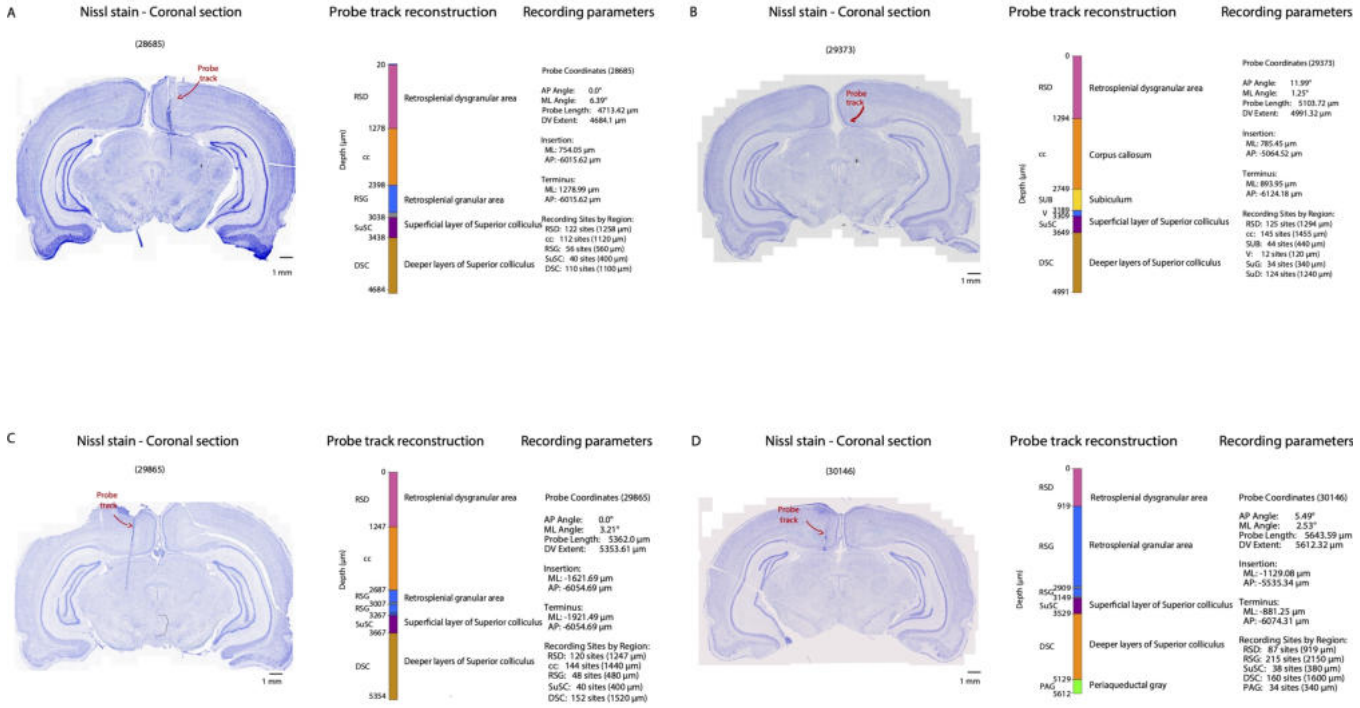

Figure S1: **Histological verification of Neuropixels probe placements, Related to Figure 1.** (A–D) Probe track reconstructions for each of the four animals used in this study (IDs: 28685, 29373, 29865, 30146). For each animal, the left panel shows a Nissl-stained coronal section with the probe track indicated (red arrow). Center panels show the reconstructed probe trajectory from HERBS software[36], with recording sites assigned to cytoarchitecturally defined regions along the dorsoventral axis. Abbreviations are: retrosplenial dysgranular cortex (RSD), corpus callosum (cc), retrosplenial granular cortex (RSG), superficial layers of superior colliculus (SuSC), and deeper layers of superior colliculus (DSC). Additional regions were identified in individual animals, including subiculum (SUB) and periaqueductal gray (PAG). Right panels list stereotaxic coordinates, probe insertion and terminus positions, probe angle, and the number of recording sites per region. Depth boundaries (in μm) are marked along each reconstruction. Scale bars: 1 mm.

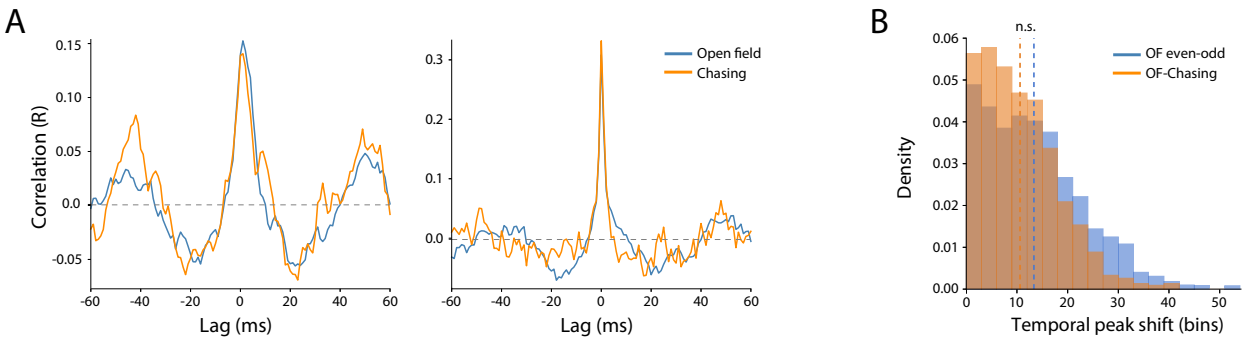

Figure S2: **Temporal coupling between EBC pairs is preserved across open field and chasing behavior, Related to Figure 2.** (A) Example temporal cross-correlation functions for representative EBC–EBC cell pairs during open field exploration (blue) and chasing behavior (orange). Cross-correlations were computed from binned spike trains, which showed similar peak structure across behavioral contexts. (B) Distribution of temporal peak shifts across EBC–EBC pairs. Peak-lag differences were compared between open field sessions (even–odd split; blue) and across behavioral contexts (open field vs. chasing; orange). Peak-lag variability was comparable or lower across tasks (~ 1.3 s for open field vs. chasing) than within the open field session (~ 1.8 s for the even–odd split), indicating that changing tasks did not affect temporal coordination. Across 1,446 EBC–EBC pairs, the change in peak correlation strength between conditions was centered near zero (median $\Delta = -0.004$; Wilcoxon signed-rank test, $p = 0.18$), and pairwise coupling strength was strongly preserved across behavioral contexts (Spearman $\rho = 0.44$, $p = 3.2 \times 10^{-10}$).

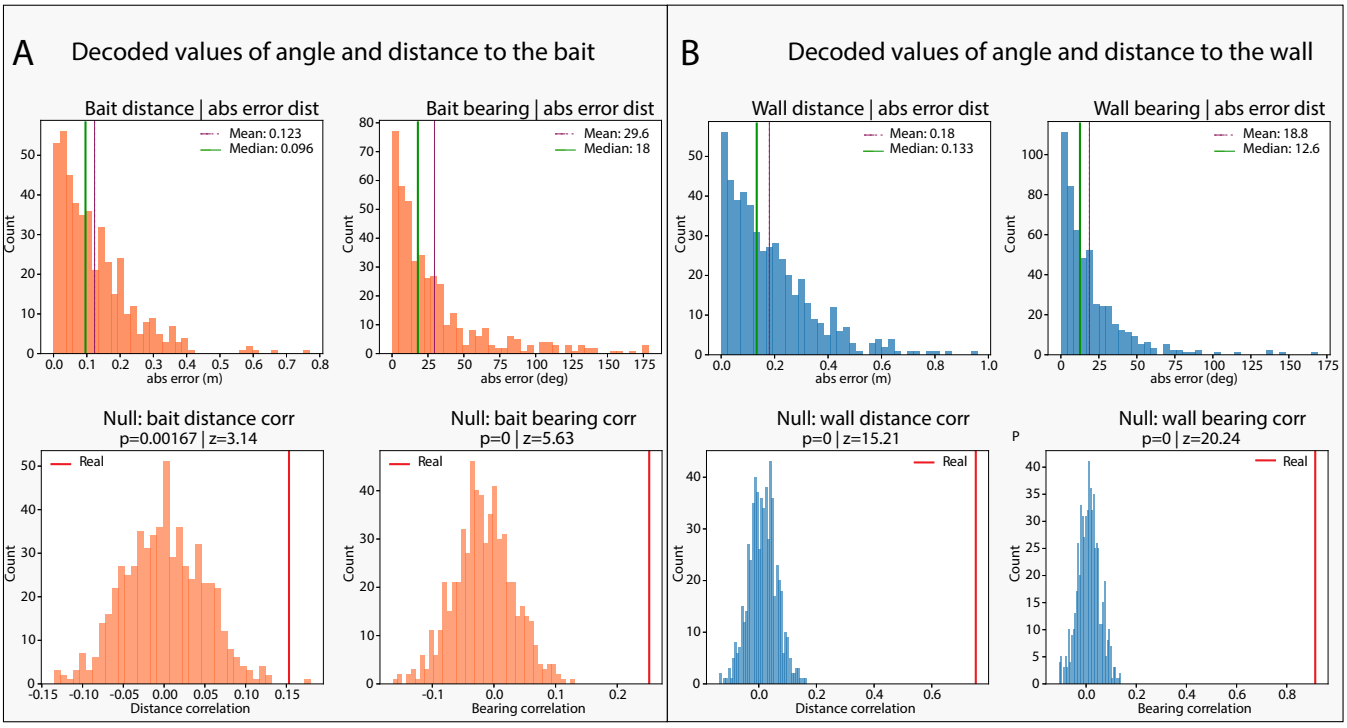

Figure S3: **Simultaneous decoding of egocentric distance and bearing to walls and bait during chasing, Related to Figure 4.** (A) Decoding performance for egocentric distance and bearing to the moving bait during chasing sessions. Histograms show absolute decoding error for bait distance (left) and bait bearing (right). Mean and median errors are indicated by dashed and solid lines, respectively (distance: mean = 0.123 m, median = 0.096 m; bearing: mean = 29.6°, median = 18°). Bottom panels show null distributions obtained by circularly shifting spike trains relative to behavior. Red vertical lines show actual decoding performance. Both bait distance and bearing correlations significantly exceeded chance levels (distance: $p = 0.00167$, $z = 3.14$; bearing: $p < 0.001$, $z = 5.63$). (B) Decoding performance for egocentric distance and bearing to the arena walls during chasing sessions. Histograms show absolute decoding error for wall distance (left) and wall bearing (right). Mean and median errors are indicated (distance: mean = 0.18 m, median = 0.133 m; bearing: mean = 18.8°, median = 12.6°). Bottom panels show null distributions for wall decoding. Real decoding correlations (red lines) were significantly greater than chance for both distance and bearing (both $p < 0.001$; $z = 15.21$ for distance and $z = 20.24$ for bearing). Together, this demonstrates that RSC population activity contains decodable information about both static environmental boundaries and dynamic targets in parallel during pursuit behavior.

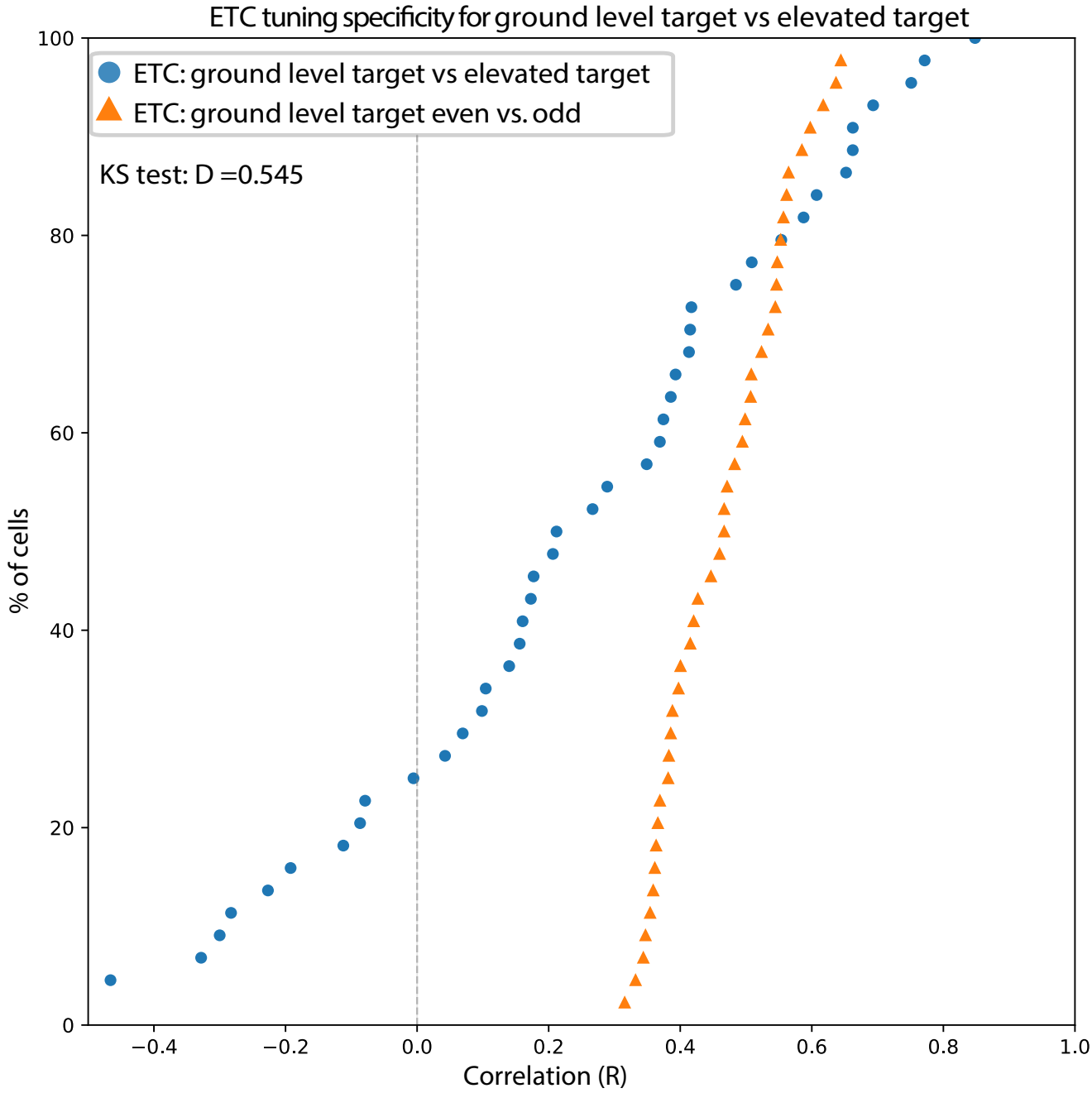

Figure S4: **ETC spatial tuning is not evoked by visual presentation of the bait alone.** Cumulative distributions of spatial firing-rate correlations for ETCs from one animal during active pursuit versus when the bait was above the arena. Correlations were computed between egocentric target-centered firing-rate maps in each condition. Orange triangles show within-session stability (computed by correlating even-odd segments) during the pursuit session; blue circles show correlations between maps obtained for ground-level pursuit and with the target elevated. Spatial correlations were substantially higher for the within-condition comparison than for the cross-target comparison (SHOW MEAN r-vals for each here), indicating that ETC tuning is specific to active engagement in pursuit of the ground-level target. The two distributions differed significantly (Kolmogorov–Smirnov test: $D = 0.545$) with p-value: 2.3423e-06 (p ¡ 0.001)

Supplementary videos

Supplementary Video 1. Raw footage of bait-chasing behavior. Black-and-white video recording of a representative active chasing bout. The animal pursues a moving bait attached to a manually operated fishing rod within the open arena. The bait trajectory was controlled by the experimenter to elicit sustained pursuit behavior while maintaining the bait within the animal's visual field. Video is shown at real-time playback speed. *Any human features visible in this video correspond to the main author of the study, who has provided consent for their inclusion.*

Supplementary Video 2. Egocentric visualization of target-tuned spiking activity. Animal-centered ("Mr. Stuck") representation of chasing behavior, in which the animal's position and heading are fixed at the center of the frame while the environment and bait position move relative to the animal. The bait location is indicated by a red dot. Audio represents spike events from a representative egocentric target cell (ETC), with each spike rendered as an audible click. Note that spiking activity increases when the bait is positioned to the right of the animal's heading, at approximately 50° eccentricity, demonstrating egocentric tuning to target direction.

Supplementary Video 3. Simultaneous Bayesian decoding of egocentric target and boundary directions. Real-time visualization of population-level decoding during a representative chasing bout. The animal's trajectory is shown in pink, with cardinal directions labeled. The red dot indicates the true bait position; the purple vector shows the decoded egocentric target direction. The blue vector indicates the true direction to the wall at any given time-point, while the green vector shows the decoded egocentric boundary direction. The black arrow denotes the animal's head direction. Inset text displays instantaneous decoding accuracy, including true and decoded bait distance and wall distance. Decoded directions were computed from simultaneous RSC population activity, demonstrating that egocentric representations of targets and boundaries can be read out in parallel.

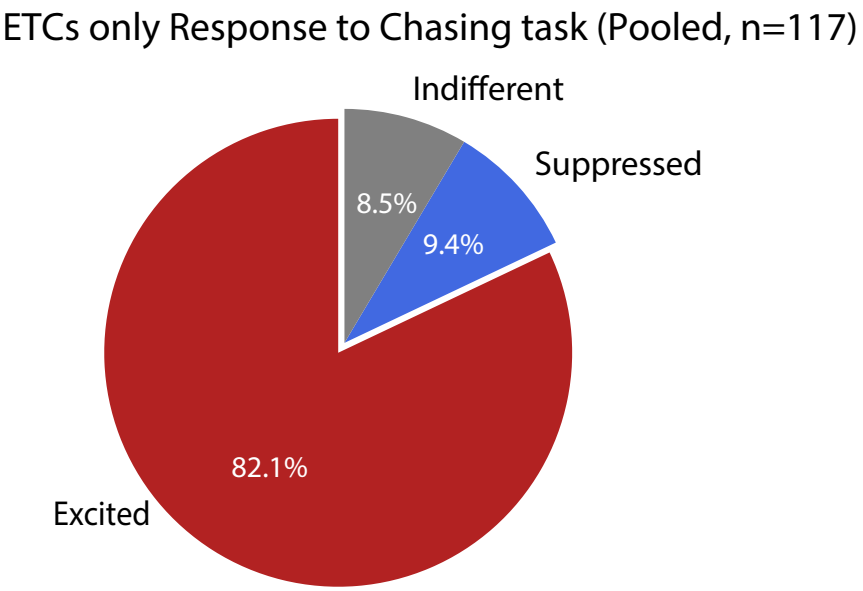

Figure S5: **Population response of ETCs during chasing.** Pie chart showing the distribution of excited, suppressed and unchanged ETCs during chasing behavior, pooled across sessions ($N = 117$ cells). Cells were classified based on changes in firing rate during chase intervals relative to non-chase periods. The majority of ETCs were significcantly up-modulated during chasing (82.1%), whereas smaller fractions were suppressed (9.4%) or showed no significant change (8.5%).

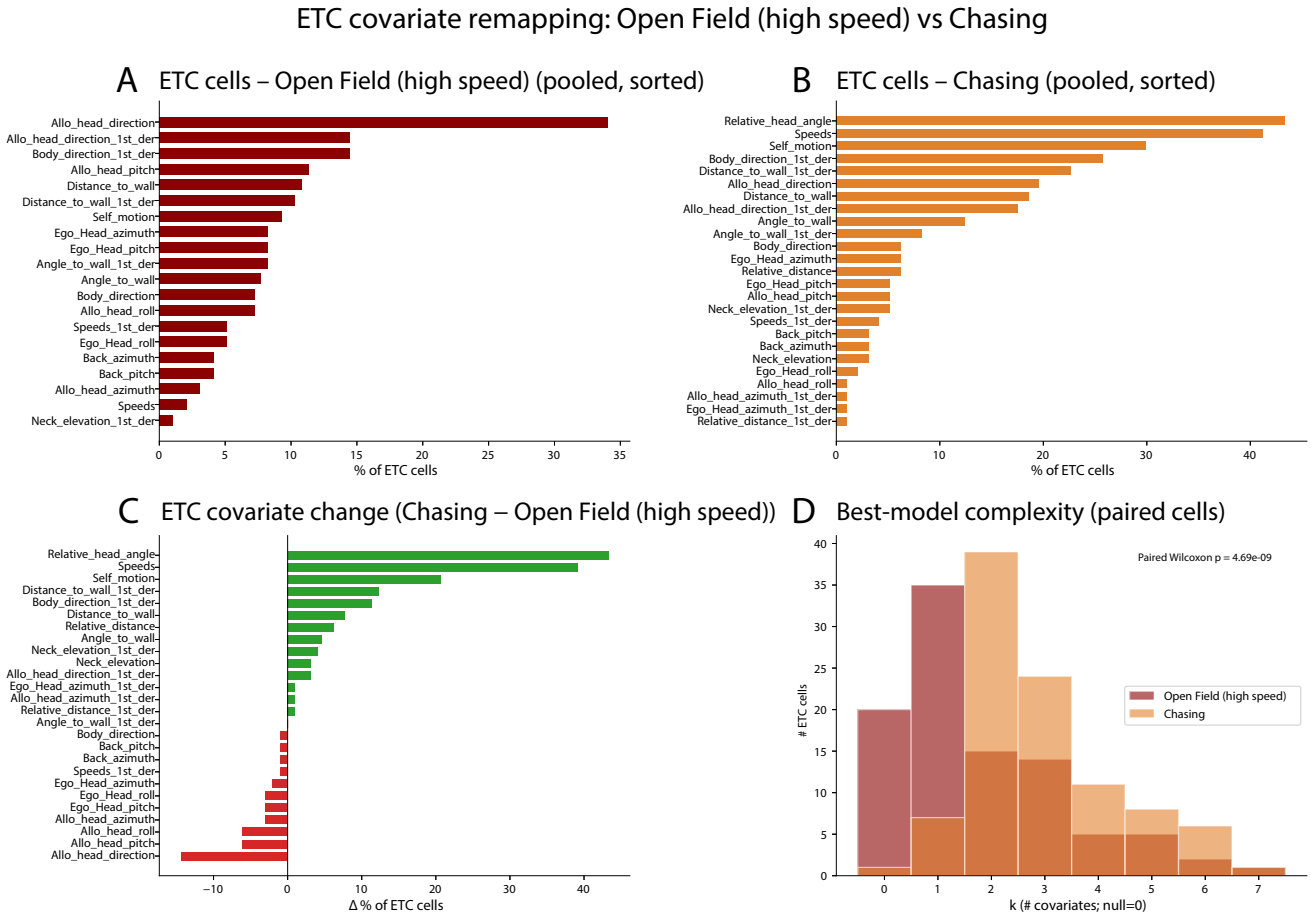

Figure S6: **ETC covariate changes persist between chasing and high-speed segments in the open field.** (A) Distribution of covariates included in the best-fit models for ETCs during high-speed epochs in the open field, pooled across cells and sorted by prevalence. "High speed" epochs are those in which locomotor speed was in the upper 50% of running speeds measured in the open field. Bars show the percentage of ETCs whose best-fitting model included a given behavioral covariate. Allocentric head direction and body direction derivatives were among the most common features in open field. (B) Distribution of best-model covariates for the same ETC population during chasing behavior. Relative head angle, speed, and self-motion variables became more prevalent along with target-related covariates. (C) The net change in covariate prevalence between conditions (chasing minus high-speed open field) showed that target-bearing and other egocentric variables (speed, self-motion in particular) showed the largest increases (net increases shown as green bars), whereas head direction decreased the most (net losses shown as red bars), followed by world-referenced postural variables. (D) Distribution of best-fit model complexity (number of covariates) for ETCs paired across conditions ($N = 79$). Model complexity increased significantly during chasing compared to high-speed epochs in the open field (paired Wilcoxon test, $p = 2.34 \times 10^{-8}$).

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