nature portfolio

Peer Review File

Neural Correlates of Trial Outcome Monitoring during Long-term Learning in Primate Posterior Parietal Cortex

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This file contains all reviewer reports in order by version, followed by all author rebuttals in order by version.

Version 0:

Reviewer comments:

Reviewer #1

(Remarks to the Author)
Liu et al. investigate neuronal activity in the posterior parietal cortex (PPC) of Macaque monkeys during a learning task using two-photon calcium imaging longitudinally. While the authors demonstrate the feasibility of two-photon microscopy in the primate posterior parietal cortex (PPC) across days, it does not present a sufficiently significant scientific advancement and remains too preliminary, requiring additional data and analysis. The technology itself—although very crucial for studying individual neuronal activity—has already been established by the same group in the prefrontal cortex. Consequently, this study provides little advancement in imaging methods and in elucidating the role of the PPC beyond what has previously been demonstrated.

Major comments
The potential significance of this study in elucidating the specific role of the PPC in learning is unclear. Prior studies have addressed the PPC function in learning and memory, and the authors do not convincingly demonstrate how their data substantially deepen or challenge existing knowledge. The listed studies—Morcos & Harvey (2016, Nature Neuroscience), Pho et al. (2018, Nature Communications), and Hanks et al. (2015, Nature), and more studies (Buneo and Andersen, 2006 Neuropsychologia; Erlich et al., 2016 eLIFE; Goard et al., 2015 eLIFE; Harvey et al., 2012 Nature; Raposo et al., 2014 Nature Neuroscience; Shadlen and Newsome, 2001 Journal of Neurophysiology; Zhang et al., 2022 Nature Communications)—have demonstrated the role of the posterior parietal cortex (PPC) in outcome history-dependent decision-making across various tasks. In addition, numerous studies support the role of the PPC in learning and memory, including its function in evidence accumulation. However, the authors only focus on a narrow niche of sensorimotor association within the broader domain of associative learning. It is uncertain whether this study on sensorimotor association provides new insights or meaningful scientific advancements beyond the existing body of knowledge. In this sense, the authors primarily emphasize the proof-of-concept aspect in long-term imaging rather than providing robust new insights into neuronal processing during learning tasks.

Another potential significance of this study lies in tracing the dynamic changes in single-neuron activity to reveal how learning progresses over days. However, the study does not make clear which representational components undergo change across days. In their task, termed the Image-Saccade Association Learning (ISAL) task, the authors propose that remapping occurs during learning. Yet, it remains unclear what this remapping represents, as novel objects were introduced from the beginning of learning. If PPC neurons merely encode object identity, remapping could occur independently of sensorimotor association, making it difficult to determine whether neural representation changes are linked to learning. The task structure, therefore, complicates the interpretation of how neuronal activity reflects the process of sensorimotor association formation. To make clear what this remapping represents, another task, such as the reversal version of Image-Saccade Association learning task, is necessary. A simple change in neuronal activity during the learning period does not necessarily constitute an important finding, especially since previous single-neuron studies in macaque monkeys have already demonstrated similar representational changes (McMahon et al., 2014 Journal of Neurophysiology; Yasuda et al., 2012 Journal of Neuroscience)

The introduction requires revision, as the discussion of technology and scientific concepts is intermingled, making it difficult to grasp the main focus of the study. From a scientific perspective, associative learning encompasses various forms, yet the importance of specifically studying sensorimotor association remains unclear. The authors argue that this function has been ‘traditionally less appreciated,’ but it is necessary to clarify why this aspect warrants investigation. Given the substantial

body of research on both the PPC and sensorimotor association learning, the introduction should reference relevant studies to justify the need for examining the PPC's role in this specific learning process. Strengthening the conceptual framework and clearly distinguishing the technological contribution from the scientific inquiry would improve the clarity and impact of the introduction.

The authors posed two questions as follows: 'How does the neuronal representation of trial outcomes change throughout the learning process to facilitate learning? And what governs these changes in neuronal representation of trial outcomes?'

However, the questions in the introduction are not sufficiently addressed by the results, as they do not clarify which neuronal representations facilitate learning or what governs these changes. To address these questions, a behavioural causality study would be necessary—such as investigating how inhibiting neural representation changes affects learning performance. In other words, the scope of the questions exceeds the range of the conclusions that can be drawn from the results, highlighting a gap between the study's initial inquiries and its empirical findings.

Further analyses are necessary to characterize the function of the primate PPC. The authors have primarily focused on neuronal activity following the target-off period, yet their task includes cue-on, delay, and target-on periods, which could provide valuable insights into the PPC's role in associative memory, working memory, and action value. To draw a robust conclusion about whether the PPC is selective for history-based choice, both single-neuron and population-level analyses are needed. Expanding the analyses to these task phases would provide a more comprehensive understanding of PPC function in learning and decision-making.

Lastly, what do the authors mean by 'monitoring'? The study does not clearly define this term, making it difficult to interpret its relevance to their findings. To draw a meaningful conclusion, the authors must first provide a clear definition of monitoring and then evaluate whether the neuronal representation in the PPC aligns with this definition. At this stage, the observed neuronal activity in the PPC appears to be a simple outcome representation that persists into the next trial, rather than an explicit monitoring mechanism.

Minor comments
Did you confirm whether AAV expression is selectively restricted to layers 2 and 3? The explanation on page 3 regarding AAV expression is unclear and may confuse readers. If not, please revise the sentence.

The figures require revision to improve clarity: For example, FOV figures lack scale bars. Figures such as 4E and 4J are missing X and Y axis labels. Additionally, the ordering of panels within each figure is difficult to follow. Overall, the figures should be revised to include all necessary information for proper interpretation.

The behavioural paradigm in Figure 1 should specify the inter-trial interval (ITI) to provide a complete description of the task structure.

Line 98: multiples days → multiple days
Line 128: during inter-trial interval → during the inter-trial interval
Line 244: notable after learned → notable after learning
Line 746: difference in monkeys' eye positions → differences in monkeys' eye position
p.3 Figure S2 (A): significant different → significant difference
There are additional grammatical errors in the manuscript that need to be addressed. The manuscript needs to be revised by a native English speaker.

Line 133: What does 0.57 ± 0.08 represent in relation to fixation breaks? Please clarify.
• Line 134: Figure S4 should be incorporated into Figure 2 in the main text.
• Line 174: What does WN type n refer to? Should this be EN type n instead?
• Line 250: I cannot find the data for outcome encoding correlation in the referenced figure. Please indicate where this data is presented.
• Line 568: Is 400 mm the correct value? Please verify.
Additionally, how was N.-N. calculated in Figure 5A?

(Remarks on code availability)

Reviewer #2

(Remarks to the Author)
The study "Neural correlates of trial outcome monitoring during long-term learning in primate posterior parietal cortex" by Liu, Jiang, she, Fang, Tang and Zhou shows a unique and important data set tracking the responses in parietal area 7a during associative learning. The study shows robust representation of decision outcome in a subset of these neurons. This is the case both at the end of the trial during reward as well as - for the outcome of the previous trial - during the current trial. Their data also indicate changes in the representation during learning and postulates a role for network connectivity in constraining this. However, - as currently presented - the direct link between these changes in neuronal representations and learning could be shown more clearly.

The major strength of the study lies in the technically challenging tracking of individual neurons in parietal cortex during several sequences of associative learning for a image-saccade pair. The data showing robust representations of behavioural outcome in area 7a are compelling.

1) Some critical aspects of the presentation make it difficult for the reader as well as the reviewer and additional information/clarifications are needed with regard to the following aspects:

- in the abstract/title: the monkey species and the area recorded from (7a) must be named. Actually the species seems to be missing from the entire paper. It should also be mentioned at the beginning of the methods.

-introduction: PPC in the primate is large with divers areas and response properties. I would have expected a paragraph discussion the role and response properties previously found in area 7a, so the reader has a chance to situate this study and its importance better. In this context, a brief sketch of the response properties in 7a in relations to what is known about sensorimotor processing and trial outcome would be essential. The only area referred to in the introduction seems to be lateral intraparietal area (line 60, ref 37), which is wrongly referred to as "lateral parietal cortex", which needs correcting. As it is, it is not clear why 7a is explored at all, other than being on the surface of the (macaque??) brain and therefore more accessible.

- Figures, Results: as the two monkeys differed in their learning , all results should also be reported for each monkey separately and in all figures the examples should be clearly associated with the animal they come from - as as is custom in the field.

- Results: a critical piece of evidence relies on assigning neurons to a specific type of representation (e.g. lines 101-109). An Anova on FEV seems to be used. In the results, the exact statistical criterion and what the correction for multiple comparisons was should be included.

- Results, Figure 2H, line 115-116: text and figure appear to contradict each other: Figure shows about 63% of neurons below one 1s, text states it the other way round.

- Results, all Figures, timing labels "Time to target off": these are generally positive numbers, so I presume it is time "after" or "from" target off that is meant. This is highly confusing and needs to be corrected. Please also check the "time to cue on" and revise, so the language is unambiguous and consistent.

- Results, Figures 2A, B, J, K: the meaning of the dashed lines in relation to the task is unclear. Please label appropriately, perhaps with arrows and names on top.

- Results 2C, 2L are not very clear. What colour is which type? It seems the yellow cells might only be present in smaller subparts of the FOV, which could be considered as clustering?

-Results, lines 133 in brackets: what is being measured (trials?) in what units (%?).

-Results Figure 3D, what is suddenly "WN"?

-Results Figure 4D and 4I: the abbreviations the legends N., D1 .... are not explained.

-Results Figures 4B,C,G,H: what is the x-axis?

2) Interpretation of the results.

The section "relevance of outcome encoding in 7a for associative learning shows that it is not reward and punishment alone. The link between outcome encoding and weight of previous choice is does not directly show a contribution to learning. the same applies for the saccade bias. Also, Figure 3H seems to rely heavily on data points that show neither a sizeable weight of the previous answer nor sizeable encoding strength.

While the data presented are suggestive of a link between behavioural outcome representation and associative learning, this is at present rather descriptive rather than compelling. What is needed is a quantitative, statistical link between changes in representation and changes in behavioural performance either day-to-day or trial-by-trial.

Since outcome encoding of the previous choice is ongoing when the animal makes a decision, so interacts directly with incoming activity (rather than what is measured at the end of the trial). Any correlation with actually learning of the association might be clearer for this epoch.

At present l201-203 only the first part is clearly supported by the data, "this encoding is closed correlated with AL" should be removed until some robust direct link can be made statistically to learning outcome. The same applies for the first sentence of the next paragraph.

l235-252: the lack of a link between outcome encoding and saccade direction encoding is also problematic in this context.

From Figure 5 and results text, it is not clearly presented whether or how the reorganisation trial outcome representations is linked to learning. This part of the results is crucial for understanding and impact of these results. At the moment, this could do with a clearer structure reporting the key results.

3) Results, Figure 2I and 2R: The latency data (negative numbers) seems to suggest that trial outcome is encoded BEFORE the reward is given. So the animal had no feedback yet. This should change your interpretation of what is encoded or at least taken into account?

4) Relationship of outcome representation to neuronal tuning for saccade localisation and relationship of previous trial to current trial outcome representations. If the outcome representations contribute to learning the appropriate association. The tuning for stimuli and saccade of neurons should matter. Doe the authors find any evidence whether different subgroups of neurons related to their tuning show different outcome representations and that the link between the two properties affect learning success?

5) Network connectivity lines 296ff and Figure 6: Potentially an interesting results. But between what neurons (tuning, entry criterion) was the noise correlation measured?

Which time window did you use? As you have effects of previous trial outcome represented in some of the time windows, you would expect some correlation due to this signal. How did you take this into account and investigate/eliminate this effect on correlations?

Figure 6F,G,J,K need clearers description in the figure legends. What data am I looking at and what is the critical piece of data shown.

Minor:

Abbreviations: the many abbreviations make the text not very readable, e.g. AL, lSAL could be written out as they are not standardly abbreviated terms.

Introduction: l51: sentence is unclear and grammatically incorrect.

Figure 1D. Legends states data are from each monkey. What part is from what monkey?

Figure: the labelling by letter seems haphazard in most figures and does not follow a clear pattern. Some panels are not really visible, like e.g. the bottom of 1G

Figurelegends: it would be helpful if the main outcome for each panel is summarised as well as only describing what is plotted.

P-values for stats are sometimes "p" (Methods) and sometimes "P" (Results). Very confusing as you also look at proportions. I suggest to standardise to "p =" and always write directly after naming the test.

Labelled neurons in Figure 4A and 4F are not easily discernible.

(Remarks on code availability)

Reviewer #3

(Remarks to the Author)
This study investigates the changes in the activity of cells in the primate area 7a, measured with two-photon calcium imaging, during an associative learning task that demands new visual-motor associations over multiple days. The results showed a strong neural representation of the trial outcome, which was closely correlated with monkeys' learning. This neural outcome signal was reorganized as monkeys transitioned from exploitation to exploration and showed a gradual evolution while the animals learned new visual-motor associations. In addition, the encoding of trail outcome was not closely related to the cell encoding of the output saccadic movements. Finally, the modulation in outcome encoding was constrained

by the local network connectivity.
This is a very interesting and well performed study, providing novel information about how the PPC incorporates local outcome signals to promote visual-motor associations along multiple days. An important strength of this manuscript is the use of calcium imaging with two-photon that allows the measurement of the activity of the same population of neurons during periods of learning that last days. Another strength in the study is the use of a simple task where two images are associated with two saccade directions over a period of days, which permits to measure the reconfiguration of the cell and network responses during the process. Nevertheless, a have some concerns. First, it is difficult to know whether the data of the figures are from the monkey with stable saccadic behaviors or some of them are from the monkey with high frequency of fixation breaks. A direct comparison of the functional properties of 7a neurons between monkeys is in place. The heterogeneity in the outcome signal modulation across FOVs is large, that should be clear in the paper and something needs to be say about what is the roles of such heterogeneity in AL. In addition, it is important to know where the FOV where within area 7a, which is a large structure and limits with 7ab and 7m that have different connectivity and functional properties. The present results describe how outcome encoding changes during long-term AL, not how the outcome signals strengths the visuomotor associations during learning. The same question is valid for the observation that neurons coactivated more closely were more likely to develop strong connections over time, leading to similar outcome encoding, but the question remains of how this encoding signal directly affects the association between image-saccade direction across days. Finally, for Figure 6 I highly recommend computing the mutual information of the neural activity during the task period between cells to generate robust graphical models of functional connectivity across cell types and learning epochs.

Minor comments
Please describe what the PsyTrack parameters are and why this model is better than others to describe AL.
The change of outcome signaling in the population showed in Figure 4 is astonishing. Please add the color code for outcome encoding of Figure 2C in Panels 4A and F.
Is the outcome signal in PPC used locally for AL or is also used in other areas?
How the complex remapping of outcome encoding linked to new associations is used to generate the AL. How the previous and actual trial outcome signal affects the image-saccade association, when the mapping of outcome encoding remapping is to complex.
What is the outcome signal doing to generate the association between the image and the saccade direction? The outcome-history encoding of the majority 7a neurons predominantly emerged and localized within the fixation period of the ISAL task, so these signals have an effect on how the cue images are progressed or kept in memory? Or is a signal that changes the reaction time of the saccades or the saccade direction?
What is the functional meaning of having outcome signals that are different between the actual and the previous trial?
What is the effect of the speed of learning (e.i. 3 vs 7 days) on the magnitude of the encoding of outcome by neurons and on dynamics across days.
What is the proportion of cells that encode the stimulus onset, the image identity, the saccade movement preparation and execution and the outcome. How many neurons encoded more than one parameter, and how the outcome signal changes during AL modified the encoding of the image and the saccade direction in the same cells or in other populations.
How many neurons encode both the previous and the actual outcome as a function of learning.
Please provide Figures of the FOV with pronounced outcome selectivity for the second monkey as in Figure S1 and S9.

Typos.
There are many typos in the manuscript such as:
from two monkeys. (see Methods).
Figure.2C

(Remarks on code availability)

Version 1:

Reviewer comments:

Reviewer #1

(Remarks to the Author)
I have carefully reviewed the authors' revised manuscript and their detailed responses to the reviewers' comments, and I greatly appreciate the substantial effort the authors have invested in addressing the concerns raised in the initial review. The revised manuscript now presents a much clearer and stronger narrative. The authors have provided additional analyses linking outcome encoding in PPC area 7a to behavioral strategies, performance accuracy, and outcome history. They have also added new figures, including revised Fig. 3H, Fig. 4A–F, and Fig. S13–S15, which convincingly demonstrate the robustness of their findings. In addition, they clarified the conceptual framework in the Introduction and Discussion by distinguishing methodological contributions from the scientific insights, explicitly acknowledged the correlational nature of their findings while highlighting the importance of future causal approaches, and improved the clarity of their description of the experimental design and the interpretation of neural remapping across learning days.

I find that the emphasis on outcome monitoring is well presented. I also find it very important that the authors highlighted that these effects were observed in the primate PPC, which significantly strengthens the impact of the study. While it is somewhat disappointing that the authors were not able to perform reversing of the learned associations, the revised

manuscript clearly focuses on how the PPC monitors outcomes during sensorimotor associative learning. The inclusion of multiple independent lines of evidence linking neural activity to learning and behavior is a substantial improvement, and this revision convincingly underscores the significance of the study.

I also found the authors' interpretation that "We propose that neural activity in area 7a may not primarily reflect the formation of sensorimotor associations, based on several observations" to be particularly interesting. This provides a new and valuable perspective on PPC function, and the deeper analyses presented in the revision convincingly strengthen the manuscript. Taken together, these points show that the authors have not only addressed the previous concerns but have also added depth and clarity to their central argument.

I commend the authors for their thorough and thoughtful revisions. I am generally satisfied with their responses, and I support acceptance of this manuscript for publication. As a final recommendation, I strongly encourage the authors to seek a round of native English proofreading to ensure clarity and polish throughout the manuscript.

(Remarks on code availability)

Reviewer #2

(Remarks to the Author)
The authors' extensive revisions have addressed my main concerns and I have no further comments.

In summary, the study "Neural correlates of trial outcome monitoring during long-term learning in primate posterior parietal cortex" by Liu, Jiang, Shi, Fang, Tang and Zhou shows a unique and important data set tracking the responses in parietal area 7a during associative learning. Their manuscript makes an important contribution to our understanding of the cortical encoding of association learning.

(Remarks on code availability)

Reviewer #3

(Remarks to the Author)
The authors answered properly all the reviewers´ comments. The new version of the paper is quite better, making a strong and congruent scientific story. I recommend the authors to make the black start of significant effect larger and clearer. Also please consider referring to the following papers on spatial cognition signals in area 7a:

Crowe, D. A., Chafee, M. V., Averbeck, B. B., & Georgopoulos, A. P. (2004). Neural activity in primate parietal area 7a related to spatial analysis of visual mazes. Cerebral Cortex, 14(1), 23-34.

Crowe, D. A., Goodwin, S. J., Blackman, R. K., Sakellaridi, S., Sponheim, S. R., MacDonald III, A. W., & Chafee, M. V. (2013). Prefrontal neurons transmit signals to parietal neurons that reflect executive control of cognition. Nature neuroscience, 16(10), 1484-1491.

Merchant, H., Battaglia-Mayer, A., & Georgopoulos, A. P. (2004). Neural responses during interception of real and apparent circularly moving stimuli in motor cortex and area 7a. Cerebral Cortex, 14(3), 314-331.

(Remarks on code availability)

Authors' Response to Reviewers

We sincerely thank all reviewers for their thoughtful and detailed feedback on the earlier version of our manuscript. The comments and questions were very helpful, and we have addressed each of them in the responses below and incorporated the corresponding revisions into the manuscript. As a result, the manuscript has been substantially improved. The revised version includes updates to all sections of the text, revised Figures 1–6 (including a new Figure 4), and 5 new supplementary figures. In brief, we have added multiple lines of evidence showing that outcome encoding in area 7a was closely linked to the monkeys' learning behavior during associative learning (AL). Notably, 7a neurons primarily encoded salient outcomes and may directly contribute to exploration behavior during AL. In the response below, reviewers' comments are shown in black, with our point-by-point replies in blue.

REVIEWER COMMENTS

Reviewer #1 (Remarks to the Author):

Liu et al. investigate neuronal activity in the posterior parietal cortex (PPC) of Macaque monkeys during a learning task using two-photon calcium imaging longitudinally. While the authors demonstrate the feasibility of two-photon microscopy in the primate posterior parietal cortex (PPC) across days, it does not present a sufficiently significant scientific advancement and remains too preliminary, requiring additional data and analysis. The technology itself—although very crucial for studying individual neuronal activity—has already been established by the same group in the prefrontal cortex. Consequently, this study provides little advancement in imaging methods and in elucidating the role of the PPC beyond what has previously been demonstrated.

Response: We acknowledge that our original manuscript did not sufficiently highlight the novelty of our findings or fully clarify the interpretation of the results. In the revised version, we have added new analyses and substantially refined our introduction and discussion to address these points. Please refer to our detailed point-by-point responses below.

Additionally, while our study does not focus on two-photon calcium imaging in behaving monkeys as a technical advancement (as this method has been well-established in our lab for years), it does represent an important methodological improvement relative to our prior PFC study. For the first time in primates, we tracked a large population of neurons across long-term learning, enabling us to observe how neuronal encoding evolves over learning days.

Major comments

The potential significance of this study in elucidating the specific role of the PPC in learning is unclear. Prior studies have addressed the PPC function in learning and memory, and the authors do not convincingly demonstrate how their data substantially deepen or challenge existing knowledge. The listed studies—Morcos & Harvey (2016, Nature Neuroscience),

Pho et al. (2018, Nature Communications), and Hanks et al. (2015, Nature), and more studies (Buneo and Andersen, 2006 Neuropsychologia; Erlich et al., 2016 eLIFE; Goard et al., 2015 eLIFE; Harvey et al., 2012 Nature; Raposo et al., 2014 Nature Neuroscience; Shadlen and Newsome, 2001 Journal of Neurophysiology; Zhang et al., 2022 Nature Communications)—have demonstrated the role of the posterior parietal cortex (PPC) in outcome history-dependent decision-making across various tasks. In addition, numerous studies support the role of the PPC in learning and memory, including its function in evidence accumulation. However, the authors only focus on a narrow niche of sensorimotor association within the broader domain of associative learning. It is uncertain whether this study on sensorimotor association provides new insights or meaningful scientific advancements beyond the existing body of knowledge. In this sense, the authors primarily emphasize the proof-of-concept aspect in long-term imaging rather than providing robust new insights into neuronal processing during learning tasks.

Response: We agree with the reviewer that the original version of the manuscript did not sufficiently clarify the primate posterior parietal cortex (PPC)'s specific role in associative learning (AL). As noted, the PPC's involvement in decision-making (e.g., evidence accumulation in perceptual/reward-based tasks) and spatial memory is well-documented in primates and rodents. In contrast, our work focuses on the PPC's contribution to sensorimotor AL—a process critical for adaptive actions, decision-making, and motor control, enabling organisms to respond effectively to changing environments.

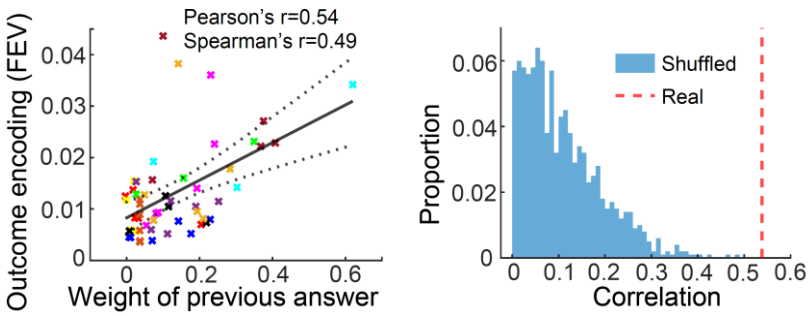
Studying sensorimotor AL offers critical insights into how the brain links sensory inputs to motor outputs based on experience to guide behavior. A key component of this process is behavioral outcome monitoring, which enables the brain to evaluate the consequences of actions, strengthen relevant associations, and adapt behavior accordingly. Therefore, understanding how the brain represents behavioral outcomes—and how these representations support learning—is essential for uncovering the neural mechanisms underlying AL. While regions such as the prefrontal cortex (PFC), cingulate cortex, and subcortical structures have been shown to represent trial outcomes during AL, the neural mechanisms underlying outcome monitoring remains poorly understood. Although previous studies in rodents have implicated the PPC in processing stimulus history and in biasing action selection based on that history, it remains an open question whether and how the primate PPC similarly monitors outcomes to support learning. In this study, we addressed this gap by proposing three key criteria that a brain region should meet to be considered central to outcome monitoring during AL. While previous studies only partially met one or two criteria, our findings demonstrate that area 7a of the primate PPC satisfies all three criteria, suggesting that this region plays an active role in monitoring trial outcomes and contributes functionally to sensorimotor AL.

To date, only one prior nonhuman primate study has reported outcome encoding (rewarded/unrewarded) in the PPC (LIP, a subregion of PPC) during a matching pennies game task, where monkeys made free choices against a computer opponent based on reward history. In contrast, our study focused on a different and less-characterized subregion of the PPC—area 7a. We found that outcome encoding in area 7a is not merely a response to reward delivery, but reflects outcome monitoring during AL, whereas the

previous LIP study did not control for potential sensory feedback confounds in outcome encoding.

Critically, **we found that outcome encoding in area 7a was closely linked to the monkeys' learning behavior in several key ways**: (1) Encoding strength predicted trial-by-trial learning strategies, such as win-stay/lose-shift patterns, across learning days (revised Figure 4A-4B); (2) Stronger encoding was associated with better learning performance, showing enhanced representation for associations with higher performance accuracy (revised Figure 4C-4D and Figure S14); (3) Outcome representations were dramatically influenced by the outcome history, as it was significantly stronger after outcome switches (correct→error or error→correct transitions) than after outcome repetitions (correct→correct or error→error) (new results, new Figure 3H and new Figure S13), suggesting area 7a primarily represents salient outcome; (4) The strength of outcome encoding significantly predicted the monkeys' learning strategy in subsequent trials, with higher outcome selectivity linked to exploratory behavior(switching saccade choice direction for the same association), suggesting that outcome encoding in area 7a might directly contribute to exploration behavior during learning (new results, new Figure 4E-4F and new Figure S15); (5) Encoding strength markedly decreased when the monkeys were not engaged in learning (Figure 3A-3D); and (6) Outcome representation was dynamically linked to learning content, with population-level encoding patterns remapping when the monkeys shifted from exploiting familiar associations to exploring novel ones (Figure 6). Together, these findings reveal potential mechanisms for outcome monitoring in primate area 7a and substantially extend current understanding of decision-making and learning processes in the PPC.

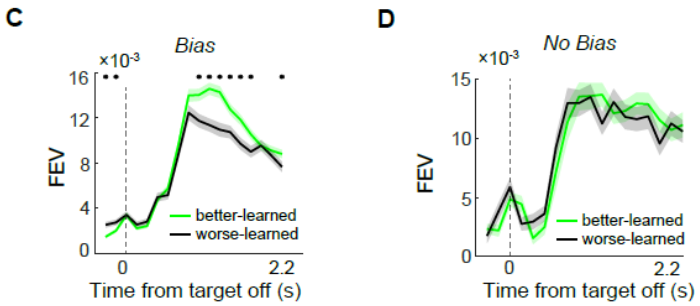
In the revised manuscript, we expanded our analyses to clarify how outcome encoding in area 7a contributes to sensorimotor AL through outcome monitoring. **First**, we examined whether and how this encoding relates to the monkeys' learning strategies. For each learning day, we fitted monkeys' behavior to a 'Psytrack' model to generate a weight of previous answer, quantifying the extent to which the monkeys' saccade choice was followed an outcome-guided strategy (win–stay or lose–shift) or an outcome-ignored strategy (win–shift or lose–stay). We found a positive correlation between the averaged strength of outcome encoding and the extent to which the monkeys relied on win–stay/lose–shift strategies across different learning days (revised Figure 4A-4B, shown in bellow). This result indicates that outcome encoding in area 7a is closely linked to the expression of adaptive choice strategies during learning. We have revised the interpretation of this result to clarify this point, shown as follows:



We have revised the corresponding results section as follows:

"Next, we investigated the relationship between outcome encoding in area 7a and the monkeys' learning behavior during AL. If outcome encoding in 7a supports outcome monitoring and contributes to sensorimotor AL, we would expect that the strength of population-level outcome encoding correlates with the monkeys' choice strategies and behavioral performance over the course of learning. To test this, we first assessed whether outcome encoding strength was associated with the extent to which monkeys relied on an outcome-guided strategy (win–stay or lose–shift) versus an outcome-ignored strategy (win–shift or lose–stay) during learning. We modeled the monkeys' saccade choices for each image-saccade association pair using the PsyTrack model, which estimates a trial-by-trial weight for outcome-guided strategies. Across different learning days, we found a significant positive correlation between the outcome-guided strategy weight and the strength of outcome encoding in area 7a (Figure 4A-4B; Pearson's r = 0.54; Spearman's r = 0.49). This result indicates that outcome encoding in 7a tracked the monkeys' use of outcome-driven strategies, supporting its role in trial-by-trial outcome monitoring during AL."

Second, we investigated whether outcome encoding in area 7a was associated with the monkeys' learning performance (revised Figure 4C-4D and Figure S14). For each learning day, we categorized the two tested associations as either better-learned or worse-learned based on the monkeys' behavioral accuracy. We found that outcome encoding was significantly stronger for the better-learned association compared to the worse-learned one. In contrast, when the monkeys' performance was similar across the two associations, outcome encoding strength did not differ significantly. These findings suggest that stronger outcome encoding in area 7a may contribute to more effective AL. We have revised the interpretation of this result and added it into the main figures, shown as follows:

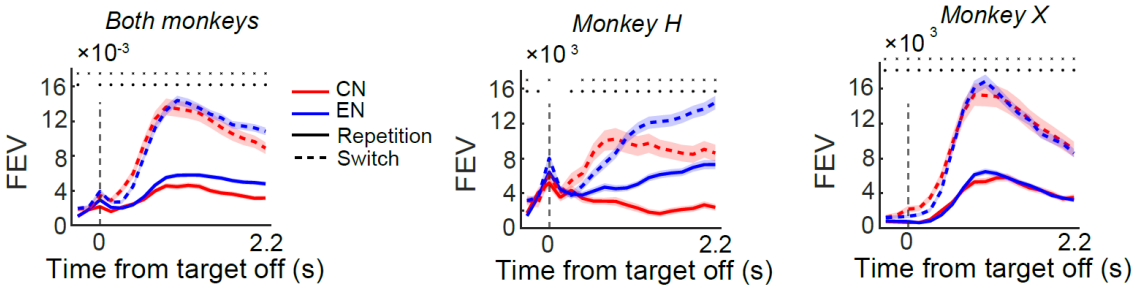


We have revised the corresponding results section as follows:

"Second, we examined whether outcome encoding in area 7a was associated with the monkeys' learning performance. For each learning day, we categorized the two tested associations as either better-learned (higher accuracy) or worse-learned (lower accuracy), based on the monkeys' performance accuracy. In sessions with a notable performance difference between the two associations, 7a neurons exhibited significantly stronger outcome encoding for the better-learned association compared to the worse-learned one (Figure 4C; P = 1.56e-7, t(17) = -8.5, paired t-test), regardless of whether the better-learned association was linked to contralateral or ipsilateral saccades (Figure S14). This difference in outcome selectivity is unlikely to be attributable to saccade direction encoding alone, as

saccade choices were made at least one second prior to target offset. In contrast, in sessions where behavioral performance did not differ significantly between the two associations, outcome encoding remained comparable across conditions (Figure 4D; P = 0.68, t(8) = 0.42, paired t-test). Together, these results indicate that outcome encoding in area 7a is closely linked to both the monkeys' behavioral strategies and their learning performance, reinforcing its proposed role in outcome monitoring during AL."

Third, we have included new analysis to investigate whether outcome encoding in area 7a was modulated by outcome history, specifically the outcome of the preceding trial. We quantified the encoding strength separately for trials following outcome switches (correct→error or error→correct transitions) versus outcome repetitions (correct→correct or error→error). Notably, outcome encoding was much stronger following outcome switches compared to outcome repetitions. This effect was consistent across both error-preferring and correct-preferring neurons. These findings suggest that outcome encoding strength in area 7a reflects the monkeys' trial-by-trial behavioral performance during learning, and that area 7a is particularly sensitive to outcome changes. This observation aligns with the well-established role of the primate PPC in salience representation. Additionally, our results are consistent with prediction error dynamics, as prediction errors are expected to decrease after the repetition of the same outcome. Taken together, these findings suggest that area 7a plays a key role in dynamic outcome monitoring, potentially facilitating adaptive learning in changing environments. Detailed results are shown in bellow:

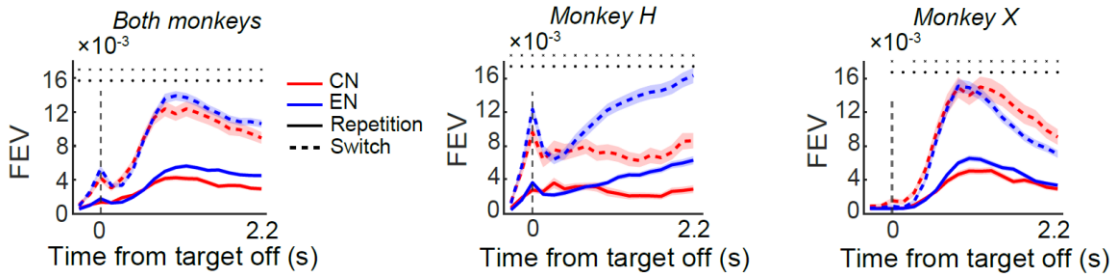


We have added one paragraph in the revised Results section to describe this result as follows:

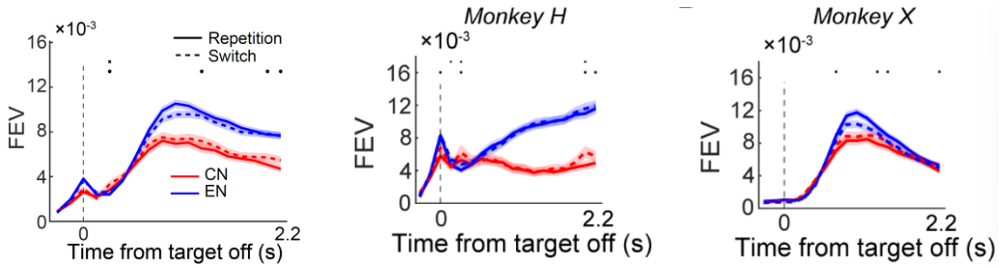
"Third, we investigated whether outcome encoding in area 7a was modulated by outcome history, specifically the outcome of the preceding trial. We quantified the encoding strength separately for trials following outcome switches (correct→error or error→correct transitions) versus outcome repetitions (correct→correct or error→error). Notably, outcome encoding was much stronger following outcome switches compared to outcome repetitions (Figure 3H, P = 6.7e-14, t(116) = -8.5, paired t-test). This effect was consistent across both error-preferring and correct-preferring neurons for both monkeys (Figure S13). Together, these results demonstrate that outcome encoding in area 7a extends beyond simple reward reception. It reflects the evaluation of both positive and negative choice consequences, and dynamically incorporates recent behavioral history. These findings align with the established role of the primate posterior parietal cortex (PPC) in salience

representation, suggesting that area 7a preferentially encodes behaviorally salient outcomes during AL."

Fourth, we conducted new analyses to test whether outcome selectivity in area 7a directly influenced the monkeys' learning behavior. Trials were categorized into two conditions: exploration, in which monkeys switched saccade direction relative to the previous trial with the same image-saccade association, and exploitation, in which they repeated the same saccade direction for the same association. For each condition, we measured outcome selectivity in the preceding trial. Outcome selectivity was significantly stronger prior to exploratory trials compared to exploitative ones (new Figure 4E), suggesting that enhanced outcome encoding in area 7a predicted subsequent exploratory behavior. The phenomenon was found for both correct-preferring and error-preferring neurons across two monkeys (Figure S15). In contrast, area 7a neurons did not show increased outcome selectivity prior to trials in which the monkeys switched saccade direction independently of the image-saccade association (new Figure 4F and Figure S15). These findings suggest that outcome representations in area 7a may contribute specifically to strategy shifts during sensorimotor association learning, rather than reflecting general switching behavior. The detailed results were shown as follows:



These figures plot the strength of outcome encoding (quantified by FEV) for trials prior to the exploration and exploitation trials respectively.



These figures plot the strength of outcome encoding (quantified by FEV) for trials prior to saccade direction switching and repeating trials independently of the image-saccade association. The left panel shows the averaged results from two monkeys.

We have added one paragraph in the revised Results section to describe this result as follows:

'Furthermore, we investigated whether outcome selectivity in area 7a directly influenced the monkeys' learning of associations. Trials were categorized into two conditions: exploration, where monkeys switched saccade direction relative to the previous

trial for the same image–saccade association, and exploitation, where they repeated the same saccade direction. For each condition, we assessed outcome selectivity in the preceding trial. Outcome selectivity was much stronger prior to exploratory trials than exploitative ones (Figure 4E, P = 6.94e-13 , t(115) = -8.1, paired t-test), suggesting that enhanced outcome encoding in area 7a predicted subsequent exploratory behavior. This effect was observed in both correct-preferring and error-preferring neurons across both monkeys (Figure S15A to S15B). In contrast, 7a neurons did not exhibit elevated outcome selectivity prior to trials where monkeys switched saccade direction independently of the image–saccade association (Figure 4F and Figure S15C to S15D). These findings suggest that outcome representations in area 7a may contribute specifically to strategy shifts during learning sensorimotor associations, rather than reflecting general motor variability. ”

Additionally, we substantially revised the Discussion sections to better clarify the advancements our study makes beyond the existing body of knowledge, as detailed below:

“The outcome signal observed in area 7a exhibited several notable features that support its important role in outcome monitoring during sensorimotor AL. First, the outcome encoding in 7a dramatically diminished in the simple saccade task that involved no learning. Second, the strength of outcome encoding significantly decreased when the time-out punishment was removed for the incorrect trials. These results suggest that the outcome representation in 7a transcends simple physical reinforcement feedback, implying involvement of cognitive processing. Third, the strength of outcome encoding in area 7a correlated with both the extent to which the monkeys' saccade choices followed a win-stay, lose-shift strategy and their performance accuracy during AL. Fourth, the outcome encoding in 7a was contingent on the specific learning content and gradually evolved during AL. This strong reliance of outcome representation on monkeys' learning behavior implies a functional contribution of the outcome signal in 7a to AL. Fifth, outcome representations in area 7a were particularly sensitive to outcome switches. This aligns with the established role of the primate PPC in salience representation. Finally, stronger outcome encoding in area 7a predicted exploratory choices in the following trial but did not predict improved learning performance. This indicates that while outcome signals in 7a may not directly facilitate the formation of sensorimotor associations, they may instead promote exploratory behavior during the learning process. Together, these results suggest that the primate PPC satisfy the three criteria for outcome monitoring. It contributes to sensorimotor AL by monitoring behavioral outcomes to facilitate exploratory behavior, extending its recognized role in reward-based decision-making.

Previous studies have identified outcome representation in brain areas outside PPC, emphasizing the significance of the frontal-striatum network in evaluating reward reinforcement. Outcome signal has been proposed to serves as a teaching signal, reinforcing correct associations while weakening incorrect ones. However, the observed outcome representation in PPC differs markedly from these earlier findings, instead highlighting a predominant sensitivity to salient, negative outcomes that may drive exploratory choices during AL. This suggests a potential complementary role of PPC in monitoring trial outcomes during sensorimotor AL, relative to the role of the frontal-striatum network. While the frontal-striatum circuitry primarily evaluates reward reinforcement and

supports the acquisition and exploitation of learned associations during AL, PPC may instead play a predominant role in detecting salient 'error signals' and facilitating exploratory behavior throughout the learning process. The outcome signal in area 7a may be primarily relayed to brain regions such as the frontal pole, which are known to drive exploratory behavior during learning. This is also in line with opponent-process theories, which propose that emotional experiences and motivated behavior are regulated by pairs of opposing processes. '.

Another potential significance of this study lies in tracing the dynamic changes in single-neuron activity to reveal how learning progresses over days. However, the study does not make clear which representational components undergo change across days. In their task, termed the Image-Saccade Association Learning (ISAL) task, the authors propose that remapping occurs during learning. Yet, it remains unclear what this remapping represents, as novel objects were introduced from the beginning of learning. If PPC neurons merely encode object identity, remapping could occur independently of sensorimotor association, making it difficult to determine whether neural representation changes are linked to learning. The task structure, therefore, complicates the interpretation of how neuronal activity reflects the process of sensorimotor association formation. To make clear what this remapping represents, another task, such as the reversal version of Image-Saccade Association learning task, is necessary. A simple change in neuronal activity during the learning period does not necessarily constitute an important finding, especially since previous single-neuron studies in macaque monkeys have already demonstrated similar representational changes (McMahon et al., 2014 Journal of Neurophysiology; Yasuda et al., 2012 Journal of Neuroscience)

Response: We agree with the reviewer that a simple change in neuronal activity during learning does not, in itself, constitute a major finding. We also acknowledge that our original description of tracking dynamic changes in single-neuron activity across learning days was unclear. Our study represents, to our knowledge, one of the first in primates to systematically examine how neural encoding across a large neuronal population evolves over multiple learning days—extending beyond prior studies that tracked only a small number of neurons longitudinally (as the suggested references).

We acknowledge that our original explanation of the experimental design spanning the learning course could have been clearer, which may have led to reviewer misunderstanding. For each neural population (FOV), recordings began on the familiar day (when monkeys performed well-learned associations) and continued until they had successfully acquired a novel pair of associations. Thus, the learning phase (novel day) spanned from the second recording day through the final day. This design allowed us to analyze how neural encoding changed both during the transition from familiar to novel associations, and throughout the process of acquiring the new associations. We have revised the results section to clarify this point in more details, shown as follows:

'The longitudinal recordings for each FOV spanned from the initial day when monkeys performed the ISAL task with familiar associations to the day when monkeys mastered a

new pair of associations."

During data analysis, we primarily focused on the changing dynamics of outcome representation across days. As shown in revised Figure 5 and 6 (original Figure 4 and 5), we found that the outcome encoding remapped when monkeys transitioned from exploiting familiar associations to learning novel ones, and then gradually evolved during the process of learning new associations over multiple days. In contrast, the saccade direction encoding (including both pre- and post-saccadic intervals) did not exhibit such remapping phenomenon, and evolved significantly slower across days. These results suggest that outcome encoding in area 7a is linked to the content of learning, rather than reflecting highly generalized reinforcement signal.

Furthermore, our revised analysis (summarized in the previous response) showed that the strength of outcome encoding was correlated with the monkeys' choice strategy across learning days (revised Figure 4A-4B), indicating that the dynamics of outcome encoding closely tracked learning behavior.

Importantly, we believe that the remapping of outcome encoding in area 7a is not simply a reflection of visual stimulus encoding, for two main reasons. First, the 7a neurons we recorded did not appear to be involved in cue identity encoding, as fewer than 4% showed significant selectivity for cue identity during the cue presentation period. This contrasts with prior electrophysiological studies in area 7a that reported robust encoding of task-relevant visual stimuli. Second, outcome encoding in our data emerged more than two seconds after the visual stimuli had disappeared. Together, these suggest that the outcome encoding we observed is not driven by residual visual information. Instead, we propose that the learning content–dependent outcome signals in area 7a may reflect the formation of a content-specific "engram" representing accumulated learning experiences.

Finally, we agree with the reviewer that additional experiments—such as reversing the learned associations—could provide deeper insights into the functional significance of neural remapping of outcome representations during learning. Unfortunately, we were unable to conduct such follow-up experiments, as the monkeys used in this study have since been retired. Nonetheless, we recognize the importance of this question and have highlighted it as a direction for future research in the revised Discussion section, as follows:

"Further studies—such as reversing the sensorimotor associations and causally manipulating activity dynamics—are needed to clarify the potential role of encoding remapping during AL."

The introduction requires revision, as the discussion of technology and scientific concepts is intermingled, making it difficult to grasp the main focus of the study. From a scientific perspective, associative learning encompasses various forms, yet the importance of specifically studying sensorimotor association remains unclear. The authors argue that this function has been 'traditionally less appreciated,' but it is necessary to clarify why this aspect warrants investigation. Given the substantial body of research on both the PPC and sensorimotor association learning, the introduction should reference relevant studies to

justify the need for examining the PPC's role in this specific learning process. Strengthening the conceptual framework and clearly distinguishing the technological contribution from the scientific inquiry would improve the clarity and impact of the introduction.

Response: We thank the reviewer for raising this important point. We acknowledge that the original introduction did not clearly articulate the conceptual framework or adequately distinguish the study's technological contributions from its scientific questions. In response to the reviewer's suggestion, we have substantially revised the Introduction section to better clarify these aspects, shown as in below:

"The ability to learn appropriate motor responses to different environmental stimuli is essential for an animal's survival in complex environment. During AL, the effectiveness of establishing new sensorimotor associations relies on the brain's ability to monitor the positive and negative outcomes of our action choices during individual learning trials. This outcome monitoring allows brain to evaluate the action consequences, reinforce relevant associations, and guide adaptive changes. Theoretically, brain regions central to outcome monitoring during AL should fulfill several criteria. First, they should exhibit significant encoding of trial outcomes (correct vs. incorrect) following the completion of each trial, with outcome signals that go beyond mere reward reception to reflect higher-level cognitive processing. Second, the strength or pattern of outcome encoding should correlate with the animal's learning performance or strategy and adapt dynamically to the learning context. Third, such outcome representations should contribute causally or functionally to animal's subsequent behavior during the learning process.

The prefrontal cortex (PFC), cingulate cortex, and other subcortical areas have been implicated in representing trial outcomes during sensorimotor AL, as their neural activity reflects the correctness of animals' action choice during short-term AL. These outcome representations were not merely reflective of reward reception; some were correlated with the animals' behavior performance during leaning. However, how the brain monitors behavioral outcomes remains elusive, as the previous studies only partially met one or two criteria. Furthermore, acquiring complex sensorimotor skills often requires gradual learning over periods spanning days or longer, but previous primate studies have primarily focused on the changes of neuronal activity during relative short period of minutes to hours during the learning process. Since no study has tracked the evolution of neuronal encoding of the same population of neurons in the primate brain over different learning days, our understanding of the mechanisms underlying long-term AL is highly limited. Key questions regarding outcome monitoring remain untested, particularly during long-term AL: How does the neuronal representation of trial outcomes correlate with the animals' behavior to potentially influence learning? How does the outcome representation change throughout the learning process and what factors govern these changes?

Previous studies have highlighted the fronto-striatal circuit as the central hub for sensorimotor AL, with less emphasis placed on the roles of other brain areas. However, changes in neuronal activity were observed in other brain areas both during and after sensorimotor AL, suggesting the engagement of a potentially broader brain network. The

PPC, a crucial association cortex positioned at the intermediate stage of sensorimotor transformation, has been implicated in various cognitive functions, but was usually not implicated in AL. The PPC has been shown to play important roles in representing stimulus-stimulus association and sensorimotor association after long-term learning, implying potential involvement in sensorimotor AL. Specifically, neuronal activity in the lateral parietal cortex has been reported to reflect outcome information (rewarded vs. non-rewarded) during a decision task. Nonetheless, the exact role of PPC in long-term AL remains largely unexplored. ”

The authors posed two questions as follows: ‘How does the neuronal representation of trial outcomes change throughout the learning process to facilitate learning? And what governs these changes in neuronal representation of trial outcomes?’

However, the questions in the introduction are not sufficiently addressed by the results, as they do not clarify which neuronal representations facilitate learning or what governs these changes. To address these questions, a behavioural causality study would be necessary—such as investigating how inhibiting neural representation changes affects learning performance. In other words, the scope of the questions exceeds the range of the conclusions that can be drawn from the results, highlighting a gap between the study’s initial inquiries and its empirical findings.

Response: We thank the reviewer for raising this important issue. We agree that our study does not establish a causal link between changes in outcome representation and the monkeys’ learning behavior. Although we have considered causally manipulating neural activity in area 7a to directly test its role in sensorimotor AL, we found that demonstrating such a causal relationship in the primate brain remains highly challenging with currently available techniques, for two main reasons.

First, the primate PPC is implicated in a broad range of cognitive functions. To establish a causal role for outcome encoding in area 7a, any manipulation would need to target outcome-related signals specifically during the inter-trial interval. This would require both high temporal and spatial precision, which is difficult to achieve in nonhuman primates. Techniques such as reversible inactivation lack the necessary temporal resolution, while microstimulation faces limitations due to the spatial overlap of different neuron types in area 7a—such as outcome-selective neurons (both correct- and error-preferring) and saccade-selective neurons—making selective targeting of outcome encoding unfeasible. Second, it is technically difficult to selectively suppress or modify the evolution of neural representations related to learning without also disrupting ongoing task-relevant activity. Current tools do not allow for precise interference with the trajectory of representational changes over time without broadly affecting neural processing.

However, in the revised manuscript, we present six lines of evidence demonstrating that outcome encoding in area 7a is closely associated with the monkeys’ learning behavior during AL: 1) The strength of outcome encoding predicted the monkeys’ trial-by-trial learning strategy (win-stay/lose-shift) across different learning days (revised Figure 4A-4B);

2) Encoding strength correlated with learning performance, showing stronger outcome representations for associations with higher behavioral accuracy (revised Figure 4C-4D and Figure S14); 3) 7a neurons primarily represented the salient outcome, as it exhibited significantly stronger outcome selectivity after outcome switches than after outcome repetitions (new Figure 3H and Figure S13); 4) The strength of outcome encoding significantly predicted the monkeys' learning strategy in subsequent trials, with higher outcome selectivity linked to exploratory behavior (new Figure 4E-4F and Figure S 15). This suggests that the outcome encoding in area 7a may directly influence the monkeys' subsequent learning strategy; 5) Encoding strength markedly decreased when the monkeys were not engaged in learning; 6) Outcome representations were linked to learning content, with population-level encoding undergoing significant remapping as monkeys transitioned from familiar to novel associations. Together, these findings suggest that outcome encoding in primate area 7a plays a meaningful role in supporting AL

Based on the findings list above, we have changed our questions in the revised manuscript as: *"How does the neuronal representation of trial outcomes correlate with the animals' behavior to potentially influence learning? How does the outcome representation change throughout the learning process and what govern these changes? ".*

Furthermore, we have addressed the second question in original Figure 6 (revised Figure 7). We demonstrated that the functional connectivity between neurons (measure by noise correlation) within local network significantly predicted the changes in outcome encoding of these neurons across learning days. Such prediction power was significantly greater than the functional similarity (measured by signal correlation) between neurons. These results suggest that functional connectivity between neurons was a notable factor which constrained the evolution of outcome encoding of individual 7a neurons across learning days.

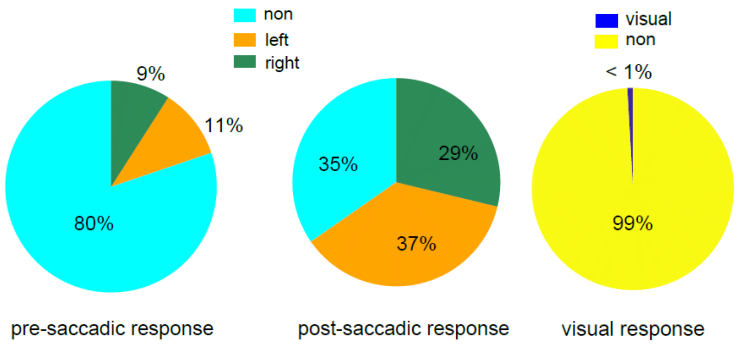
Additionally, we recognize the importance of casual experiment to test the roles of PPC played during AL and have highlighted it as a direction for future research in the revised Discussion section, as follows:

'Nonetheless, the role of the primate PPC in outcome monitoring and its potential contribution to exploration-driven learning processes warranting further investigation with causal manipulations.".

Further analyses are necessary to characterize the function of the primate PPC. The authors have primarily focused on neuronal activity following the target-off period, yet their task includes cue-on, delay, and target-on periods, which could provide valuable insights into the PPC's role in associative memory, working memory, and action value. To draw a robust conclusion about whether the PPC is selective for history-based choice, both single-neuron and population-level analyses are needed. Expanding the analyses to these task phases would provide a more comprehensive understanding of PPC function in learning and decision-making.

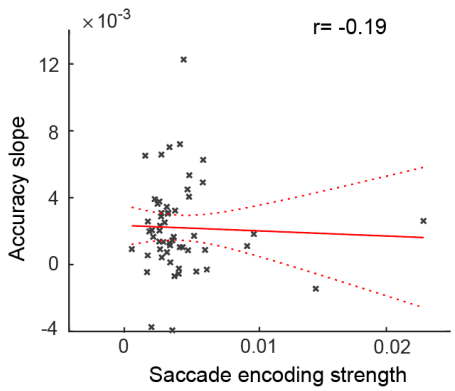
Response: We agree with the reviewer that additional analyses of neuronal activity during the cue-on, delay, and target-on periods could provide a more robust understanding of the

PPC's role in history-based decision-making. In the original manuscript, we reported that fewer than 1% of area 7a neurons exhibited significant visual responses during the cue-on period (original Figure S8). Approximately 20% of 7a neurons showed significant encoding of saccade direction—which reflects image-saccade associations—during the delay period, while around 65% exhibited significant saccade direction encoding during the target-on period. These results were included as revised Figure S9, and shown in below. We have also demonstrated that the saccade direction encoding (combining both pre- and post-saccadic intervals) evolved significantly slower than that of outcome encoding across days (revised Figure 6C).



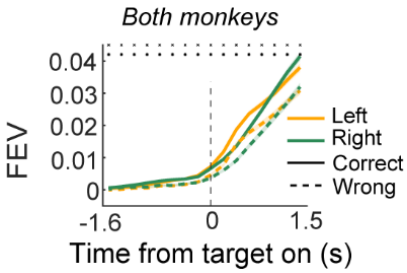
The proportion of neurons exhibited different selectivity in the ISAL task.

Following the reviewer's suggestion, we have included additional analyses of saccade direction encoding during the delay period in the revised manuscript, as this activity could potentially reflect the formation of image-saccade associations. Specifically, we examined how saccade direction encoding during the delay period evolved across different learning days. We did not find systematic relationship between the averaged strength of saccade direction encoding during the delay interval and the monkeys' performance accuracy during learning. This result was included in new Figure S10 in the revised manuscript. This indicates that the saccade direction encoding in area 7a might not directly contribute to acquiring the image-saccade associations. The detailed result is shown as follows:



Furthermore, we have added new analysis to investigate whether the saccade direction encoding was modulated by the trial outcome in the preceding trials. Specifically, we found that saccade direction encoding of 7a neurons—which likely reflects image-saccade associations—was very modestly, though significantly, stronger when the

monkey's saccade choice was correct in the preceding trial compared to when it was incorrect. This effect was observed in both left-preferring and right-preferring neurons. These findings suggest that outcome signals may modulate saccade direction encoding in the subsequent trial in area 7a. These results have been included as new Figure S16, and shown in bellow:



However, the current study specifically focused on the role of the area 7a of primate PPC in AL, particularly in outcome monitoring, for two reasons. First, prior studies—including our own causal works—have demonstrated that the primate PPC is critical for perceptual and categorical decision-making (Zhou & Freedman, 2019; Zhou et. al., 2023), yet its role in AL remains unclear. Second, as listed in bellow, we believe area 7a may not be central to acquiring or storing sensorimotor associations.

We propose that neural activity in area 7a may not primarily reflect the formation of sensorimotor associations, based on several observations. First, very few 7a neurons (< 4%) exhibited significant visual responses encoding cue identity or specific cue–saccade associations. Second, only a small subset of neurons (approximately 20%, much less than the outcome selective neurons) showed significant pre-saccadic activity during the delay period indicative of association encoding. And the averaged encoding strength of saccade direction was very weak during the delay period. Third, as detailed in the above response, this pre-saccadic encoding did not display systematic changes across learning days that correlated with the monkeys' learning behavior (new Figure S10). These findings suggest that area 7a may not play a central role in forming or encoding sensorimotor associations directly. Accordingly, the current study focuses on elucidating the role of area 7a in monitoring trial outcomes during AL.

To clarify these points, we have expanded the discussion on the role of area 7a in AL in the revised manuscript, as shown below:

'The outcome signal observed in area 7a exhibited several notable features that support its important role in outcome monitoring during sensorimotor AL. First, the outcome encoding in 7a dramatically diminished in the simple saccade task that involved no learning. Second, the strength of outcome encoding significantly decreased when the time-out punishment was removed for the incorrect trials. These results suggest that the outcome representation in 7a transcends simple physical reinforcement feedback, implying involvement of cognitive processing. Third, the strength of outcome encoding in area 7a correlated with both the extent to which the monkeys' saccade choices followed a win-stay, lose-shift strategy and their performance accuracy during AL. Fourth, the outcome encoding in 7a was contingent on the specific learning content and gradually evolved

during AL. This strong reliance of outcome representation on monkeys' learning behavior implies a functional contribution of the outcome signal in 7a to AL. Fifth, outcome representations in area 7a were particularly sensitive to outcome switches. This aligns with the established role of the primate PPC in salience representation. Finally, stronger outcome encoding in area 7a predicted exploratory choices in the following trial but did not predict improved learning performance. This indicates that while outcome signals in 7a may not directly facilitate the formation of sensorimotor associations, they may instead promote exploratory behavior during the learning process. Together, these results suggest that the primate PPC satisfy the three criteria for outcome monitoring. It contributes to sensorimotor AL by monitoring behavioral outcomes to facilitate exploratory behavior, extending its recognized role in reward-based decision-making.

Previous studies have identified outcome representation in brain areas outside PPC, emphasizing the significance of the frontal-striatum network in evaluating reward reinforcement. Outcome signal has been proposed to serves as a teaching signal, reinforcing correct associations while weakening incorrect ones. However, the observed outcome representation in PPC differs markedly from these earlier findings, instead highlighting a predominant sensitivity to salient, negative outcomes that may drive exploratory choices during AL. This suggests a potential complementary role of PPC in monitoring trial outcomes during sensorimotor AL, relative to the role of the frontal-striatum network. While the frontal-striatum circuitry primarily evaluates reward reinforcement and supports the acquisition and exploitation of learned associations during AL, PPC may instead play a predominant role in detecting salient 'error signals' and facilitating exploratory behavior throughout the learning process. The outcome signal in area 7a may be primarily relayed to brain regions such as the frontal pole, which are known to drive exploratory behavior during learning. This is also in line with opponent-process theories, which propose that emotional experiences and motivated behavior are regulated by pairs of opposing processes.

The critical roles of the fronto-striatal network in AL have been demonstrated in previous studies using causal approaches, such lesion studies. However, the causal involvement of the primate PPC in long-term AL remains to be tested in future studies. Our findings suggest that area 7a may not play a central role in forming or encoding sensorimotor associations during AL. First, only a small fraction of 7a neurons showed significant encoding of the visual cue during either the cue presentation (<4%) or delay (20%) periods of the ISA task. Second, encoding of saccade direction prior to the monkeys' choices was notably weak. Third, this pre-saccadic encoding did not show systematic changes across learning days that aligned with the monkeys' learning performance. The relative lack of attention to the primate PPC in earlier AL studies aligns with our observation that area 7a may not be directly involved in acquiring new associations. Instead, it may contribute to outcome monitoring—specifically by encoding salient outcome signals that promote exploratory behavior during AL. Nonetheless, the role of the primate PPC in outcome monitoring and its potential contribution to exploration-driven learning processes warranting further investigation with causal manipulations. Previous rodent studies have reported that PPC was causally involved in processing sensory-stimulus history and

history-based action selection bias to guide decision behavior. Moreover, PPC was also shown to represent discrepancies between the current model and observed state transitions in reinforcement learning, as well as reward-related belief updating during flexible decision-making. Collectively, these results indicate that PPC likely plays an important role in updating the estimation of the current state by incorporating past experience, to guide decision-making and learning.'.

Therefore, the revised manuscript remains focusing on investigating the PPC's potential role in monitoring trial outcomes during sensorimotor AL. Outcome signals can serve as teaching signals that guide learning, so we centered our analyses on the post-target period, when monkeys could evaluate trial outcomes. We found that area 7a exhibited robust encoding of trial outcomes, which went beyond mere reward reception and was closely linked to the monkeys' learning behavior. Notably, 7a neurons primarily represented the salient outcome during leaning, and the strength of this encoding potentially drive the monkeys' learning strategy shifts. These findings highlight a key role for primate PPC in monitoring outcomes during sensorimotor AL. In contrast, because the monkeys could not determine the trial outcome during the cue-on, delay, and target-on periods, we did not prioritize those epochs in our analysis.

Lastly, what do the authors mean by 'monitoring'? The study does not clearly define this term, making it difficult to interpret its relevance to their findings. To draw a meaningful conclusion, the authors must first provide a clear definition of monitoring and then evaluate whether the neuronal representation in the PPC aligns with this definition. At this stage, the observed neuronal activity in the PPC appears to be a simple outcome representation that persists into the next trial, rather than an explicit monitoring mechanism.

Response: We thank the reviewer for this valuable suggestion, which motivated us to reorganize both the introduction and results sections. In theory, outcome signals can function as teaching signals to drive AL. In the revised manuscript, we propose that neurons involved in outcome monitoring during AL should meet three criteria: First, they should exhibit significant encoding of trial outcomes (correct vs. incorrect) following the completion of each trial, with outcome signals that go beyond mere reward reception to reflect higher-level cognitive processing. Second, the strength or pattern of outcome encoding should correlate with the animal's learning performance or strategy and adapt dynamically to the learning context. Third, such outcome representations should contribute causally or functionally to animal's subsequent behavior during the learning process.

In the revised introduction, we clearly articulate these criteria for identifying outcome-monitoring neurons. We also revised the results section to demonstrate that outcome encoding in area 7a meets these criteria. The updated introduction is provided below:

'Theoretically, brain regions central to outcome monitoring during AL should fulfill several criteria. First, they should exhibit significant encoding of trial outcomes (correct vs. incorrect) following the completion of each trial, with outcome signals that go beyond mere reward reception to reflect higher-level cognitive processing. Second, the strength or pattern of outcome encoding should correlate with the animal's learning performance or

strategy and adapt dynamically to the learning context. Third, such outcome representations should contribute causally or functionally to animal's subsequent behavior during the learning process.'

Minor comments

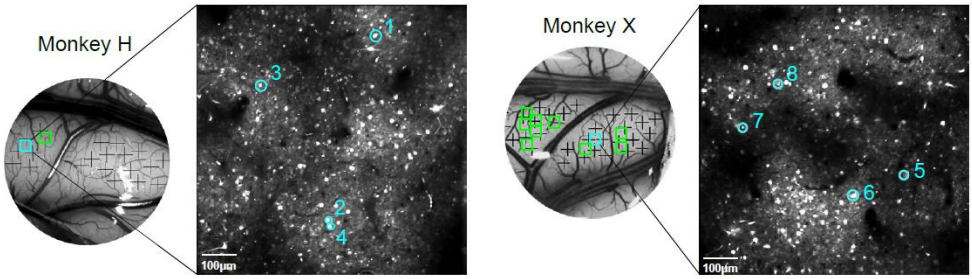
Did you confirm whether AAV expression is selectively restricted to layers 2 and 3? The explanation on page 3 regarding AAV expression is unclear and may confuse readers. If not, please revise the sentence.

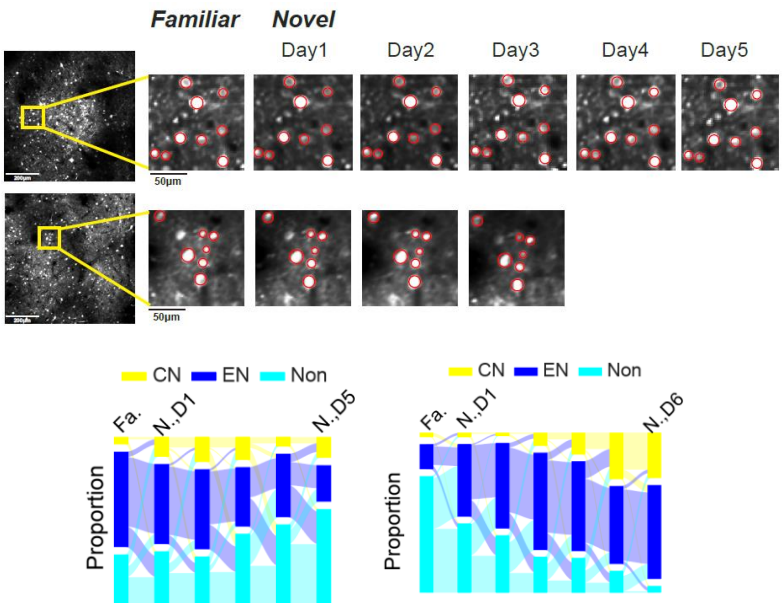
Response: We realize that our original statement may have been unclear and potentially confuse readers. The details of our protocol for virus expression in the macaque brain has been demonstrated previously (Li, M., Liu, F., Jiang, H., Lee, T. S., & Tang, S. (2017).). The virus was injected a depth about 400 μm in the cortex. We are not sure that the AAV virus expression was only restricted to layers 2-3. Because of the technical limitation of two-photon calcium imaging in monkeys, we could only neuronal activity in layers 2-3 with good signal to noise ratio. We have revised this sentence to make it clear in the revised results section, shown as follows:

'To monitor the neuronal activity during sensorimotor AL, we injected AAV to induce GCaMP6f expression in neurons at a depth of ~400 μm within area 7a, allowing us to simultaneously record the activity of hundreds of neurons over multiples days (Figure 1B, 1G and 1H). The recordings spanned depths of 200–300 μm beneath the arachnoid membrane, primarily corresponding to layers 2–3.'

The figures require revision to improve clarity: For example, FOV figures lack scale bars. Figures such as 4E and 4J are missing X and Y axis labels. Additionally, the ordering of panels within each figure is difficult to follow. Overall, the figures should be revised to include all necessary information for proper interpretation.

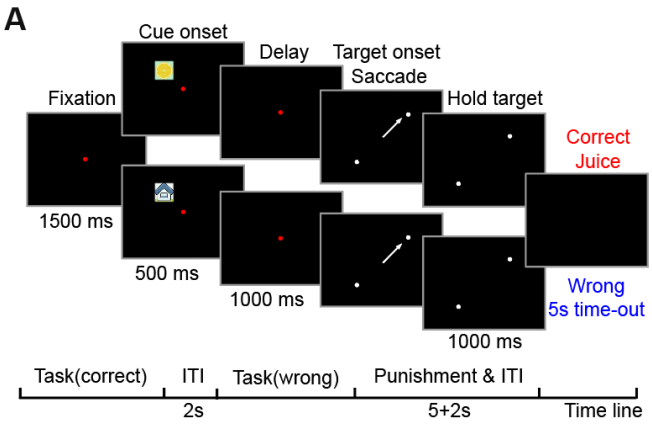
Response: Thank you for the helpful suggestion. In the revised manuscript, we have added more details to the figures to improve clarity and readability. For example, we included scale bars in the FOV images and added X and Y axis labels in the Sankey diagrams (e.g., Figures 4E and 4J). In addition, we reorganized the layout of most main figure panels to enhance visual coherence. Some of the revised figures are shown below:





The behavioural paradigm in Figure 1 should specify the inter-trial interval (ITI) to provide a complete description of the task structure.

Response: Thank for the suggestion. We have revised the Figure 1A and added a time line, shown as follows:



Line 98: multiples days → multiple days
Line 128: during inter-trial interval → during the inter-trial interval
Line 244: notable after learned → notable after learning
Line 746: difference in monkeys' eye positions → differences in monkeys' eye position
p.3 Figure S2 (A): significant different → significant difference
There are additional grammatical errors in the manuscript that need to be addressed. The manuscript needs to be revised by a native English speaker.

Response: Thanks for the corrections. We have corrected them all in the revised manuscript.

Line 133: What does 0.57 ± 0.08 represent in relation to fixation breaks? Please clarify.

Response: We realized that our description in the original manuscript might be unclear. 0.57 ± 0.08 represents fix-break ration of this monkey during performing the task. We have revised this sentence to make it clear as follows:

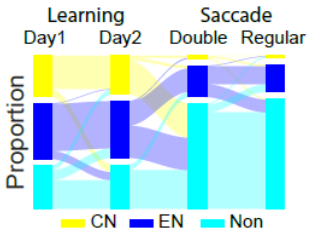
'For this analysis, we focused solely on data from one monkey due to the high frequency of fixation breaks (fix-break ratio = 0.57±0.08) during performing the task exhibited by the other monkey.'

• Line 134: Figure S4 should be incorporated into Figure 2 in the main text.

Response: We thank the reviewer for the valuable suggestion and agree that the results in the original Figure S4 are important and align with those in Figure 2. However, Figure 2 is already quite large, and adding Figure S4 would make it difficult to organize all the panels effectively. Therefore, we have decided to keep Figure S4 as a supplementary figure. That said, if the reviewer feels it is necessary, we are happy to incorporate Figure S4 into the revised Figure 2.

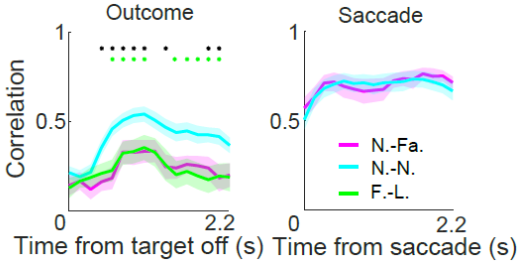
• Line 174: What does WN type n refer to? Should this be EN type n instead?

Response: Thanks for pointing out this typo. We have corrected it in the revised Figure3D, shown as follows:



• Line 250: I cannot find the data for outcome encoding correlation in the referenced figure. Please indicate where this data is presented.

Response: We thank the reviewer to point this out. We have added this results in the revised Figure 6 A, shown as follows:



• Line 568: Is 400 mm the correct value? Please verify.

Response: Thanks for pointing out this typo. This should be 400 μm. We have corrected it in the revised manuscript.

Additionally, how was N.-N. calculated in Figure 5A?

Response: We realized that this part might not be very clear. In the original Methods section, we described the detailed method for plotting this figure:

"In order to quantify the stability of the neural encoding across each learning course, we calculated the correlation of outcome/saccade encoding between two consecutive days within the learning course for each FOV. This process involved two steps. 1) For each individual neuron within the FOV, we quantified the strength of the outcome/saccade encoding using FEV analysis and determined their preferences for each time point. Consequently, for each time point, we constructed an encoding vector that represented the neural encoding within each FOV. This vector incorporated both the strength and preference of neural encodings for all identified neurons within this FOV. 2) At each time point, we calculated the Pearson correlation between the encoding vectors for two consecutive days. This resulted in a correlation value that indicated the similarity of the neural encodings between those two days for each of the given tine point. We repeated the procedures for all time points included in the analysis."

To improve clarity, we have added a paragraph in the revised Methods section to further detail the analysis:

'In Figure 6A, neural encoding correlations were computed separately for two conditions: (1) between the familiar day and the first learning day (familiar-to-novel) and (2) between successive learning days (novel-to-novel). For the novel-to-novel condition, we first calculated pairwise correlations between each pair of successive learning days (while monkeys were acquiring the same novel associations) and then averaged these correlation values.'

Reviewer #2 (Remarks to the Author):

The study "Neural correlates of trial outcome monitoring during long-term learning in primate posterior parietal cortex" by Liu, Jiang, she, Fang, Tang and Zhou shows a unique and important data set tracking the responses in parietal area 7a during associative learning. The study shows robust representation of decision outcome in a subset of these neurons. This is the case both at the end of the trial during reward as well as - for the outcome of the previous trial - during the current trial. Their data also indicate changes in the representation during learning and postulates a role for network connectivity in constraining this. However, - as currently presented - the direct link between these changes in neuronal representations and learning could be shown more clearly.

The major strength of the study lies in the technically challenging tracking of individual

neurons in parietal cortex during several sequences of associative learning for a image-saccade pair. The data showing robust representations of behavioural outcome in area 7a are compelling.

Response: We thank the reviewer for the positive comments on our work. We have addressed all the comments as described below, which were very helpful in improving the manuscript.

1) Some critical aspects of the presentation make it difficult for the reader as well as the reviewer and additional information/clarifications are needed with regard to the following aspects:

- in the abstract/title: the monkey species and the area recorded from (7a) must be named. Actually the species seems to be missing from the entire paper. It should also be mentioned at the beginning of the methods.

Response: Thanks for this suggestion! We used Rhesus Macaque in the current study. We have added the monkey species in the Abstract, Results, and Methods sections of the revised manuscript. We have also added area 7a in the revised abstract, shown as follows:

"Using two-photon calcium imaging in behaving macaque monkeys, we tracked the neuronal activity in area 7a of the posterior parietal cortex (PPC), an important associative cortex that has not been typically implicated in associative learning (AL), while the monkeys learned new visual-motor associations over multiple days."

-introduction: PPC in the primate is large with divers areas and response properties. I would have expected a paragraph discussion the role and response properties previously found in area 7a, so the reader has a chance to situate this study and its importance better. In this context, a brief sketch of the response properties in 7a in relations to what is known about sensorimotor processing and trial outcome would be essential. The only area referred to in the introduction seems to be lateral intraparietal area (line 60, ref 37), which is wrongly referred to as "lateral parietal cortex", which needs correcting. As it is, it is not clear why 7a is explored at all, other than being on the surface of the (macaque??) brain and therefore more accessible.

Response: This is a good suggestion! We have added a paragraph in the Introduction section to summarize the known role of area 7a in sensorimotor processing and its previously reported response properties, as shown below:

"Area 7a, a subregion of the PPC, occupies the highest level of the visual processing hierarchy in the parietal lobe. Traditionally, it has been implicated in diverse cognitive functions, including spatial attention, multisensory integration, motor planning, coordinate transformations, and other higher-order visuospatial processes. However, its core computational roles remain debated. Notably, Area 7a exhibited direct connections with regions involved in learning and memory, such as PFC and medial temporal lobe, as well as with structures engaged in sensorimotor transformation, such as oculomotor areas, suggesting a potential role in sensorimotor AL. Despite this, direct empirical evidence for

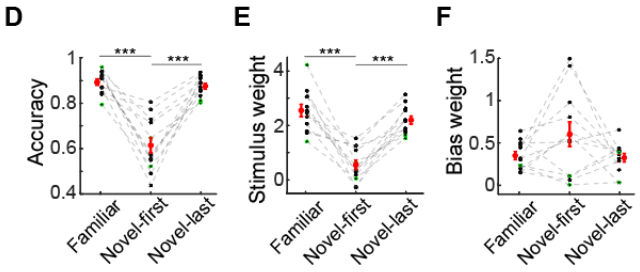
its involvement in AL—and the mechanisms through which it may contribute—remains unexplored"

Furthermore, LIP (lateral intraparietal area) is a subregion of the PPC in primates and has been studied more extensively than area 7a.

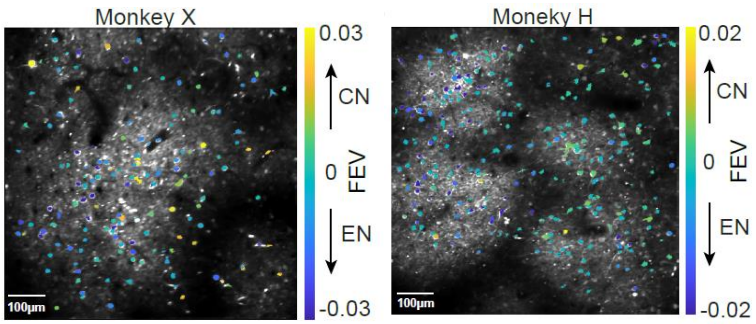
- Figures, Results: as the two monkeys differed in their learning, all results should also be reported for each monkey separately and in all figures the examples should be clearly associated with the animal they come from - as is custom in the field.

Response: Thank you for the suggestion! In the revised manuscript, we have added supplementary figures presenting the results for each monkey separately for most analyses, except for Figures 5, 6 and 7 which investigate the evolution of outcome encoding across learning course. This exception is due to the fact that we only recorded a complete learning course from two FOVs in one monkey, and the data from this animal alone is insufficient for meaningful statistical analysis. Comparisons of neuronal encoding between the two monkeys are provided in the revised Figures S1, S2, S6, S11, S13, and S15.

Furthermore, we have revised several figures and indicated the data from the two monkeys in the figure legends throughout the manuscript. For example, in the revised Figure 1D–F, we used different colors to distinguish between the monkeys (green for Monkey H and black for Monkey X), as shown below:



Additionally, we have included the monkey's name in the example FOVs, such as the revised Figure 2C and 2L.



- Results: a critical piece of evidence relies on assigning neurons to a specific type of representation (e.g. lines 101-109). An Anova on FEV seems to be used. In the results, the exact statistical criterion and what the correction for multiple comparisons was should be

included.

Response: Thanks for pointing this out. We realized that we missed the important method information for identifying outcome-selective neurons in the original results section. We classified neurons into different types of outcome-selective neurons based on the following method as stated in the original methods section:

"To identify neurons with significant encoding of trial outcome, we applied a Wilcoxon ranksum test to assess activity between the correct and error trials in the task period following target offset (1-16 frames (0-2.2s) after target offset). We further divided this period into four smaller windows, each comprising 4 frames. If statistically significant differential activity between correct and error trials was observed within at least one of these windows ($P < 0.01$, Bonferroni corrected), we classified the neuron as outcome-selective. If the neurons exhibited significant greater activity on correct trials, we classified them as correct neurons, and classified them as error neurons vice verses.'

In the revised manuscript, we have included a new sentence to clarify the statistical method used for identifying the outcome selective neurons in the results section, which is shown as follows:

"We found that some 7a neurons significantly encoded trial outcome after the saccade target disappeared (ranksum test, $P < 0.01$, Bonferroni corrected, see Methods)."

- Results, Figure 2H, line 115-116: text and figure appear to contradict each other: Figure shows about 63% of neurons below one 1s, text states it the other way round.

Response: We realized our original figure was unclear and caused confusion to the reviewer. As described in our previous response, we examined the outcome selectivity in a time period consist of 16 recoding time frames, and separated the time periods into four small time windows within each consist of four time frames. Since the temporal resolution of our two-photon calcium imaging recoding was 7.4 HZ, neuron exhibited significant outcome selectivity within two time windows indicating that its outcome encoding sustained more than 1s. We have changed the corresponding X tick in Figure 2H and added one sentence in the revised figure legend to clarify this point, which is shown as follows:

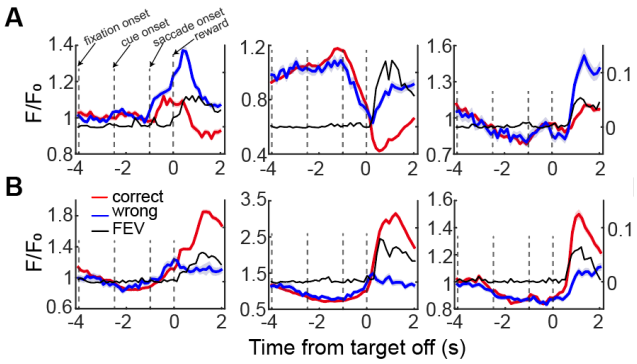
'(H) Duration of outcome encoding for all outcome-selective neurons. Each time window spans 0.54s. 63% neurons exhibited significant outcome encoding two or more time windows"

- Results, all Figures, timing labels "Time to target off": these are generally positive numbers, so I presume it is time "after" or "from" target off that is meant. This is highly confusing and needs to be corrected. Please also check the "time to cue on" and revise, so the language is unambiguous and consistent.

Response: Thank you for the suggestion! We have updated the revised figures to use 'Time from' accordingly.

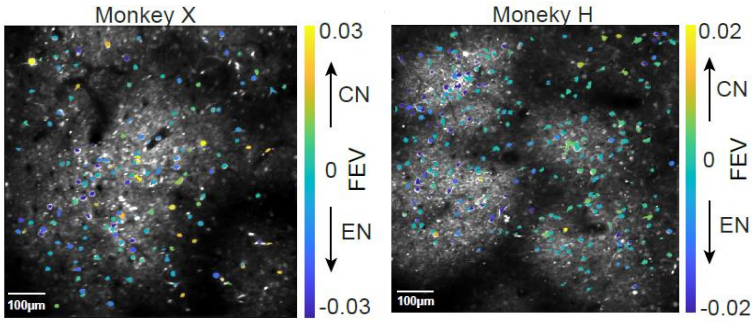
- Results, Figures 2A, B, J, K: the meaning of the dashed lines in relation to the task is unclear. Please label appropriately, perhaps with arrows and names on top.

Response: Thanks for this suggestion! We have revised the figures following the reviewer's suggestion and added arrows and names on the top of these figures, shown as follows:

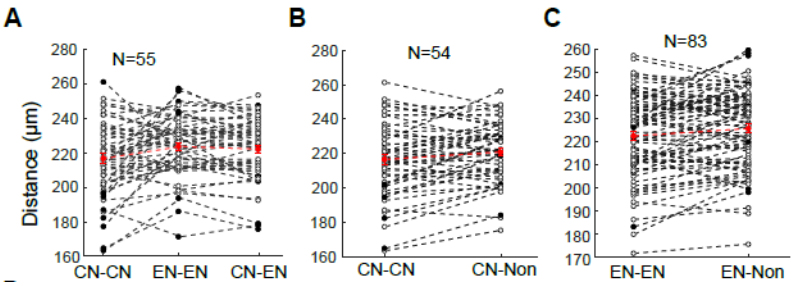


- Results 2C, 2L are not very clear. What colour is which type? It seems the yellow cells might only be present in smaller subparts of the FOV, which could be considered as clustering

The color bar (ranging from blue to yellow) represents the average strength (quantified by FEV values) and the preference of neuronal encoding for trial outcomes in the current trial (Figure 2C) or the previous trial (Figure 2L). Yellow indicates neurons that prefer correct outcomes, while blue indicates neurons that prefer error outcomes. We have included this information in the revised figures (shown in below).



These yellow-labeled neurons are not clustered within a small subregion but are instead distributed across both FOVs. Furthermore, we calculated Euclidean Distance to quantify the spatial clustering of different neuron types, and the analysis did not reveal significant clustering of outcome-encoding neurons within most FOVs. The results are shown below:



In the above figures, we quantified the mean distances between neurons within each type (correct-preferring neurons [CN], error-preferring neurons [EN], and non-selective neurons) as well as between different types. Data from Figure 2C are highlighted in red. Although there was a trend suggesting that within-type distances were smaller than between-type distances, this difference did not reach statistical significance.

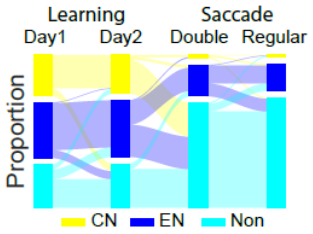
-Results, lines 133 in brackets: what is being measured (trials?) in what units (%?).

Response: We realize that the original statement may have been unclear. The value "0.57 ± 0.08" refers to the proportion of fixation-break trials during task performance. We have revised the sentence accordingly, as follows:

"For this analysis, we focused solely on data from one monkey due to the high frequency of fixation breaks (fix-break ratio = 0.57±0.08) during performing the task exhibited by the other monkey. This resulted in an excessive number of trials with no choice interspersed between completed trials.".

-Results Figure 3D, what is suddenly "WN"?

Response: Thanks for pointing this type out. We have corrected it in the revised Figure3D shown in bellow:



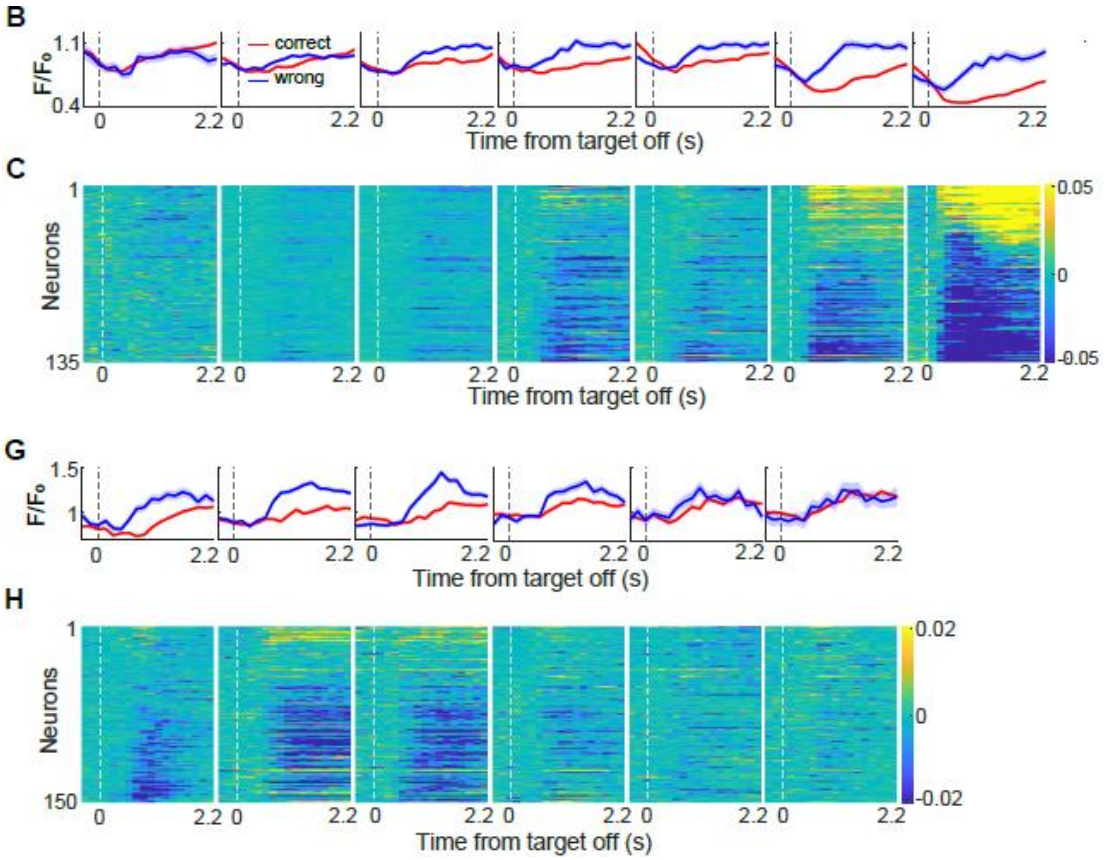
-Results Figure 4D and 4I: the abbreviations the legends N., D1 .... are not explained.

Response: In the revised figure legend, we have added detailed annotation for these abbreviations, shown as follows:

"Fa.: familiar day; N., D1: novel stimuli, day 1; N., D2: novel stimuli, day2; and so on".

-Results Figures 4B,C,G,H: what is the x-axis?

Response: In Figures 4B,C,G,H (revised Figure 5), the x-axis denotes the time period after target off. We have added the labels of the x-axis in these figures in the revised manuscript, shown as follows:



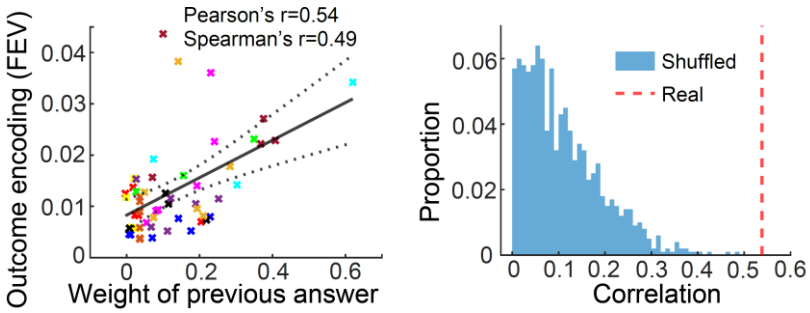
2) Interpretation of the results.

The section "relevance of outcome encoding in 7a for associative learning shows that it is not reward and punishment alone. The link between outcome encoding and weight of previous choice is does not directly show a contribution to learning. the same applies for the saccade bias. Also, Figure 3H seems to rely heavily on data points that show neither a sizeable weight of the previous answer nor sizeable encoding strength.

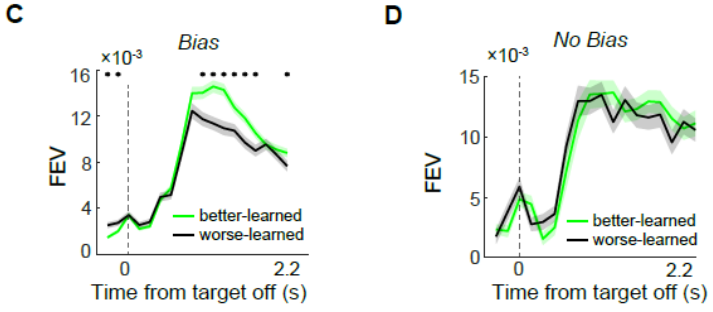
Response: We acknowledge that the original manuscript did not fully clarify the interpretation of our results. We also agree with the reviewer that the link between outcome encoding, the weight of the previous choice, and the relationship between outcome encoding and choice bias did not directly demonstrate the contribution of outcome signals to learning, but rather reflected a correlation between outcome encoding and the monkeys' learning behavior. In the revised manuscript, we have reinterpreted these results and added further analysis to demonstrate that outcome encoding in area 7a is closely linked to the monkeys' learning strategy and is correlated with their performance during learning.

First, we have revised the interpretation of the results regarding the positive correlation between the strength of outcome encoding and the weight of previous choices. The "weight of previous choice" refers to how much the monkeys' saccade choices rely on

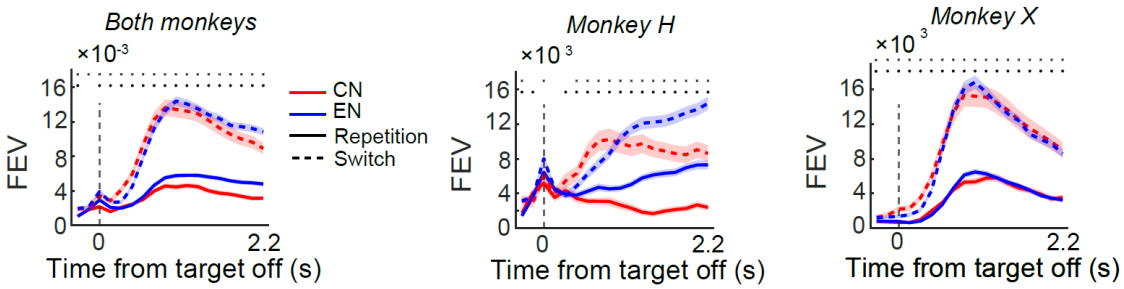
the win-stay and lose-shift strategy, a common strategy used by animals to learn novel associations. Outcome information is expected to support the application of this strategy. We found a positive correlation between the daily strength of outcome encoding and how much the monkeys' saccade choices relied on this win-stay/lose-shift strategy across different learning days. This indicate that the fluctuation of the outcome encoding strength across learning days correlated with monkeys' daily learning strategy. Regarding Figure 3H, we have added two additional analyses to confirm the findings. On one hand, we found a significant rank correlation (r = 0.49) between the strength of outcome encoding and the weight of the previous choice on the monkeys' decisions, in addition to significant Pearson correlation reported in the original Figure 3H (now revised Figure 4A). Second, we demonstrated that this correlation value was substantially higher than the correlation values from a shuffle analysis. These results indicate that the observed correlation is not just driven by data points with very week encoding strength. Therefore, we are confident that the positive correlation in Figure 3H reflects a genuine relationship. The revised results are shown below:



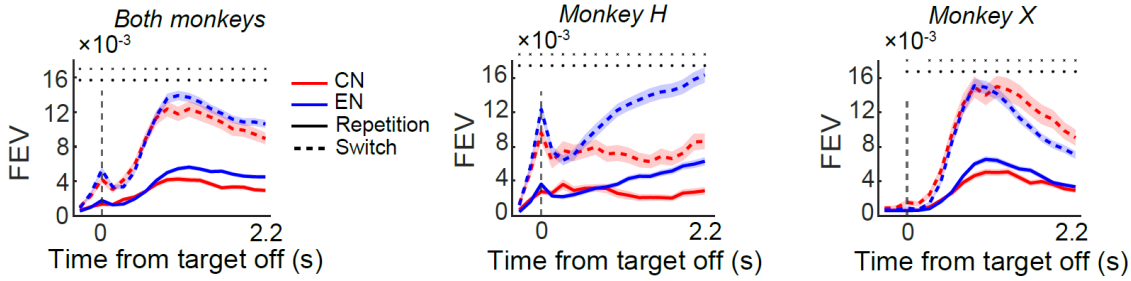
Second, we revised the analysis to explore the relationship between the strength of outcome encoding and the monkeys' choice bias. In fact, the monkeys' performance accuracy was generally higher in the biased condition than in the opposite condition. Therefore, we separated the two image-saccade association conditions into "better-learned" and "worse-learned" conditions based on the monkeys' average performance accuracy for each learning day. We found that outcome encoding was stronger for the better-learned association than for the worse-learned association, particularly when there was a notable difference in performance accuracy between the two associations. However, when the performance accuracies were similar between the two associations, the strength of outcome encoding was comparable. These results are presented in the revised Figure 4C-4D, as shown below:



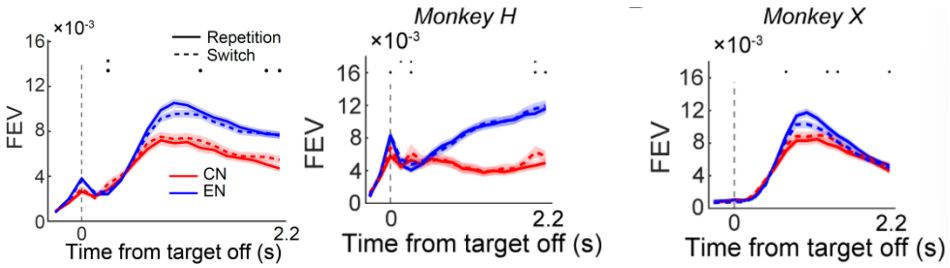
Third, we added an analysis to show that outcome encoding dramatically influenced by the outcome history (outcome in the preceding trial). We separately quantified encoding strength for trials following outcome switches (correct→error or error→correct transitions) versus outcome repetitions (correct→correct or error→error). Notably, outcome encoding was much stronger after outcome switches than after outcome repetitions (new Figure 3H, shown in below). This effect was observed in both correct-preferring and error-preferring neurons across both monkeys (Figure S13). This indicates that outcome encoding strength in area 7a tracked the monkeys' trial-by-trial behavioral performance during learning, and may support associative learning through heightened sensitivity to salient outcomes.



Fourth, we conducted new analyses to test whether outcome selectivity in area 7a directly influenced the monkeys' learning behavior in the subsequent trial. We categorized trials into two conditions: exploration (monkeys switched saccade direction relative to the preceding trial with the same image-saccade association) and exploitation (monkeys repeated the same saccade direction for the same association). For each condition, we quantified outcome selectivity in the preceding trial. We found significantly higher outcome selectivity prior to the exploration condition compared to the exploitation condition, suggesting that stronger outcome encoding in area 7a predicted exploratory behavior in the subsequent trial (new Figure 4E). The phenomenon was consistent for both correct-preferring and error-preferring neurons across two monkeys (new Figure S15). In contrast, area 7a neurons did not show increased outcome selectivity prior to trials in which the monkeys switched saccade direction independently of the image-saccade association (new Figure 4F and Figure S15). These results imply that area 7a's outcome representations may drive learning strategy shifts during learning sensorimotor associations, rather than reflecting general motor variability. The detailed results were shown as follows:



These figures plot the strength of outcome encoding (quantified by FEV) for trials prior to the exploration and exploitation trials respectively.



These figures plot the strength of outcome encoding (quantified by FEV) for trials prior to saccade direction switching and repeating trials independently of the image-saccade association. The left panel shows the averaged results from two monkeys.

In summary, we believe we have provided strong evidence demonstrating that outcome encoding in area 7a is closely correlated with the monkeys' learning behavior, and potentially contrite to their learning strategy. We also acknowledge that a causal experiment would be more suitable for establishing a direct link between outcome encoding and associative learning. The necessity of such a causal experiment for future studies has been discussed in the revised discussion section, shown in below:

'Nonetheless, the role of the primate PPC in outcome monitoring and its potential contribution to exploration-driven learning processes warranting further investigation with causal manipulations.'.

While the data presented are suggestive of a link between behavioural outcome representation and associative learning, this is at present rather descriptive rather than compelling. What is needed is a quantitative, statistical link between changes in representation and changes in behavioural performance either day-to-day or trial-by-trial.

Response: We agree with the reviewer that a quantitative, statistical link between changes in neural representation and behavioral performance provides strong evidence that outcome signals may contribute to associative learning. In response to this suggestion, we have conducted four additional analyses in the revised manuscript.

First, we found that the averaged outcome encoding was stronger for the better-learned association compared to the worse-learned association for each learning day (revised Figure 4C-4D). This result was detailed in a previous response.

Second, as described earlier, we demonstrated that outcome encoding dramatically influenced by the outcome history, as it was significantly stronger after outcome switches than after outcome repetitions (new Figure 3H and Figure S13). This finding further suggests that outcome encoding is closely correlated with the monkeys' trial-by-trial behavior during learning, potentially facilitating adaptive learning through emphasizing the salient outcome signal.

Third, as described in the earlier response, we conducted new analyses to show that outcome selectivity in area 7a may directly influence the monkeys' learning behavior in the subsequent trials, as the stronger outcome selectivity lead to exploration behavior during

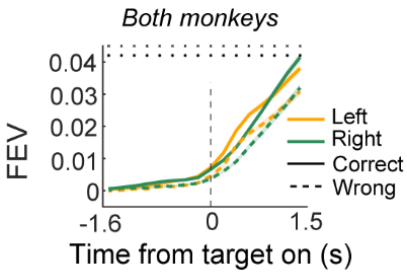
learning (new Figure 4E-4F and Figure S15).

In summary, we have revised our original analysis and added more data to strengthen the link between outcome encoding in area 7a and the monkeys' learning behavior. As a result, we believe the connection between outcome representation in area 7a and AL is now more compelling than in the original version.

Since outcome encoding of the previous choice is ongoing when the animal makes a decision, so interacts directly with incoming activity (rather than what is measured at the end of the trial). Any correlation with actually learning of the association might be clearer for this epoch.

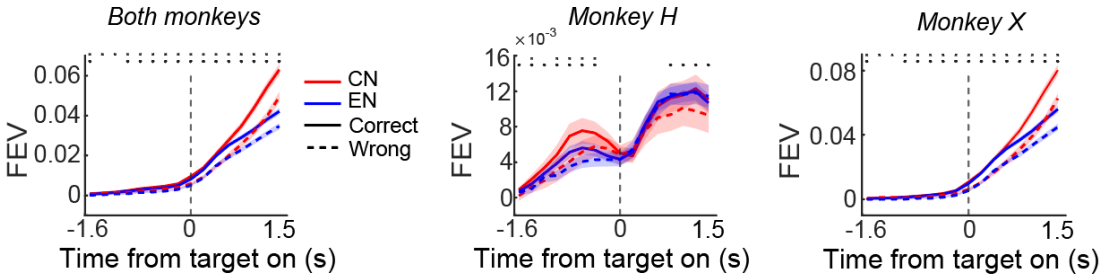
Response: We thank the reviewer for this inspiring suggestion. In the revised manuscript, we have added analysis examining whether outcome history influenced the neural encoding of trial encoding in the current trial. As detailed in our previous responses, we found that the outcome encoding in area 7a was significantly stronger after outcome switches than after outcome repetitions (new Figure 3H and Figure S13).

Furthermore, we examined whether the encoding of outcome history influences the encoding of image-saccade associations during the cue presentation and delay periods of the learning task—prior to the monkeys' saccade choice. We performed this analysis using two complementary approaches. First, we examined whether outcome history influenced the encoding of image-saccade associations in neurons that significantly represented saccade direction /these associations. Our analysis revealed that saccade direction encoding—which likely reflects image-saccade associations—was very modestly, but significantly, stronger when the monkey's saccade choice was correct in the preceding trial compared to when it was incorrect. This effect was observed in both left-preferring and right-preferring neurons. These findings suggest that outcome signals may modulate saccade direction encoding in the subsequent trial, potentially contributing to the learning of new associations. These findings have been incorporated into the revised manuscript as Figure S16 (shown below):



Second, we focused specifically on neurons that showed significant encoding of outcome history. These neurons were categorized into correct-preferring neurons (those showing greater activity following correct trials) and error-preferring neurons (those showing greater activity following error trials). Both types of neurons exhibited a trend of greater saccade direction encoding when the preceding trial was correct compared to when it was incorrect (shown in below). Since this result is similar as the one described above,

we did not include it into the revised manuscript.



In summary, we found that outcome history affected both outcome encoding and saccade direction encoding in area 7a neurons during subsequent trials. However, these two types of encoding were modulated by outcome history in distinct ways. Notably, the influence of outcome history was substantially stronger on outcome encoding than on saccade direction encoding in the following trial. Both sets of results have been included in the revised manuscript.

At present l201-203 only the first part is clearly supported by the data, "this encoding is closed correlated with AL" should be removed until some robust direct link can be made statistically to learning outcome. The same applies for the first sentence of the next paragraph.

Response: We recognize that the original statement may have been overly conclusive. In the revised manuscript, we have added further analyses and carefully revised the interpretation of some original results to more accurately demonstrate the relationship between outcome encoding and the monkeys' learning behavior.

As detailed in our previous responses, the revised manuscript presents six lines of evidence showing that outcome encoding in area 7a is closely associated with learning behavior during AL: 1) The strength of outcome encoding predicted the monkeys' trial-by-trial learning strategy (win-stay/lose-shift) across different learning days (revised Figure 4A-4B); 2) Encoding strength correlated with learning performance, showing stronger outcome representations for associations with higher behavioral accuracy (revised Figure 4C-4D); 3) Outcome encoding was significantly stronger following outcome switches compared to following outcome repetitions (new Figure 3H and Figure S13); 4) The strength of outcome encoding significantly predicted the monkeys' learning strategy in subsequent trials, with higher outcome selectivity linked to exploratory behavior (new Figure 4E-4F and Figure S15); 5) Encoding strength markedly decreased when the monkeys were not engaged in learning (Figure 3A-3D); 6) Outcome representations were linked to learning content, with population-level encoding undergoing significant remapping as monkeys transitioned from familiar to novel associations (Figure 6). Together, these findings strengthen our confidence in concluding that outcome encoding in primate area 7a is closely correlated with the monkeys' behavior during AL.

Accordingly, we have substantially revised this part in the revised result section, and concluded this part as follows:

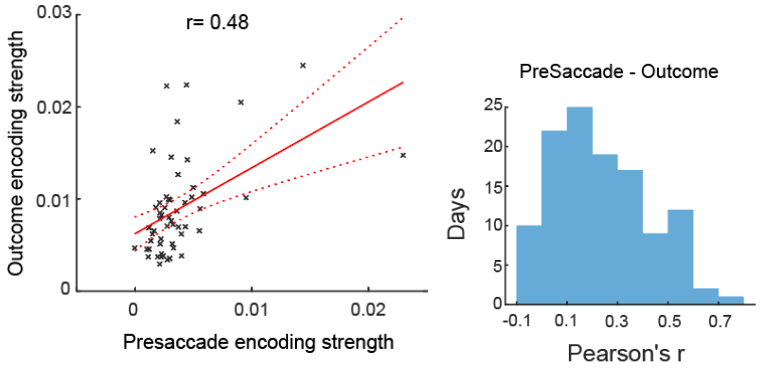
"Together, these findings indicate that outcome encoding in area 7a fulfills all three proposed criteria for outcome monitoring, suggesting that this region plays an active role in monitoring trial outcomes and contributes functionally to sensorimotor AL.".

l235-252: the lack of a link between outcome encoding and saccade direction encoding is also problematic in this context. From Figure 5 and results text, it is not clearly presented whether or how the reorganisation trial outcome representations is linked to learning. This part of the results is crucial for understanding and impact of these results. At the moment, this could do with a clearer structure reporting the key results.

Response: Thank you for the suggestion. We realized that the original manuscript did not systematically address the relationship between outcome encoding and saccade direction encoding. To fill this gap, we performed two analyses on saccade direction encoding during the delay interval (potentially encode the image-saccade associations) in the revised manuscript.

First, we tested whether the strength of saccade direction encoding differed between trials with distinct outcome histories. This analysis revealed that saccade direction encoding was slightly stronger when the monkey's saccade choice was correct in the preceding trial compared to when it was incorrect, suggesting that outcome signals may modulate saccade direction encoding in the subsequent trial, potentially influencing the learning of new associations. We have included this results as new Figure S16. Please refer to our previous response.

Second, we examined the relationship between outcome encoding and saccade direction encoding. At the population level, we found that the average strength of outcome encoding was positively correlated with the average strength of saccade direction encoding (during the delay period) across learning days (R = 0.48). A similar positive correlation between outcome encoding and saccade direction encoding was observed in the majority of recording sessions. These results are now presented in the new Figure S10A-S10B, shown below:



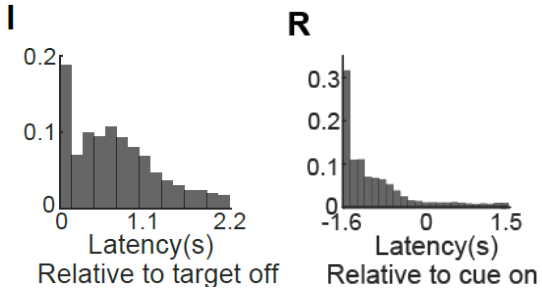
Notably, although the above results demonstrate correlations between the outcome signal and the saccade direction encoding, our results do not support a critical role for primate area 7a in representing or acquiring sensorimotor associations. First, only a very

small proportion of 7a neurons (<4%) exhibited significant visual responses encoding cue identity or specific cue–saccade associations. Second, only a limited subset of neurons (approximately 20%)—substantially fewer than outcome-selective neurons—showed significant pre-saccadic activity during the delay period that could reflect association encoding, and even this pre-saccadic activity was markedly weaker than outcome encoding. Third, the strength of this pre-saccadic activity did not systematically change across learning days in a manner that correlated with behavioral performance (new Figure S10). These findings suggest that area 7a is unlikely to play a central role in directly forming or encoding sensorimotor associations. Rather, the outcome-related signals observed in area 7a may be relayed to other brain regions more directly involved in acquiring and storing these associations. We have added on paragraph in the revised discussion section to discuss this point:

"Our findings suggest that area 7a may not play a central role in forming or encoding sensorimotor associations during AL. First, only a small fraction of 7a neurons showed significant encoding of the visual cue during either the cue presentation (<4%) or delay (20%) periods of the ISA task. Second, encoding of saccade direction prior to the monkeys' choices was notably weak. Third, this pre-saccadic encoding did not show systematic changes across learning days that aligned with the monkeys' learning performance. The relative lack of attention to the primate PPC in earlier AL studies aligns with our observation that area 7a may not be directly involved in acquiring new associations. Instead, it may contribute to outcome monitoring—specifically by encoding salient outcome signals that promote exploratory behavior during AL.".

3) Results, Figure 2I and 2R: The latency data (negative numbers) seems to suggest that trial outcome is encoded BEFORE the reward is given. So the animal had no feedback yet. This should change your interpretation of what is encoded or at least taken into account?

Response: We realized that the original Figure 2I and 2R may have been unclear and potentially caused confusion for the reviewer. Specifically, Figure 2I illustrates the latency of outcome encoding in the current trial relative to target offset, with no negative values on the x-axis. In contrast, Figure 2R depicts the latency of outcome history encoding from the previous trial relative to visual cue onset. In the revised manuscript, we have updated these figures and added clear labeling on the x-axis to indicate the corresponding time windows in each panel, as shown below:

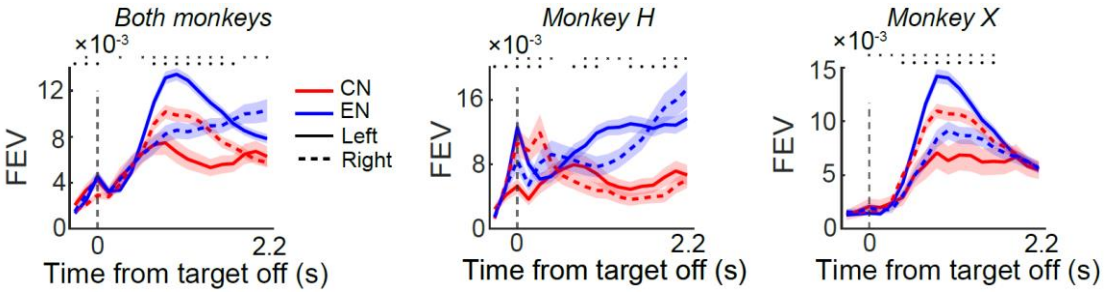


4) Relationship of outcome representation to neuronal tuning for saccade localization and

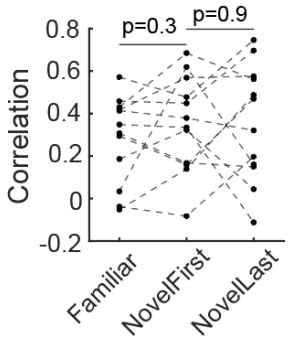
relationship of previous trial to current trial outcome representations. If the outcome representations contribute to learning the appropriate association. The tuning for stimuli and saccade of neurons should matter. Doe the authors find any evidence whether different subgroups of neurons related to their tuning show different outcome representations and that the link between the two properties affect learning success?

Response: This is a valuable suggestion. In the revised manuscript, we have added analysis examining whether outcome history influenced the neural encoding of outcome encoding in the current trial. As detailed in our previous responses, we found that the outcome encoding in area 7a was significantly stronger after outcome switches than after outcome repetitions (new Figure 3H and Figure S13).

Furthermore, we have included two new analysis to address the link between the saccade direction encoding and outcome encoding in area 7a. First, we tested whether there were differences in outcome encoding between neurons that preferred to encode different saccade directions. Specifically, we separated neurons that exhibited significant saccade direction encoding into left-preferring neurons and right-preferring neurons, and then tested whether there were differences in the strength and preference in their outcome encoding. Since there are very few 7a neurons significantly encoded the cue-identify during the cue-presentation period, we focused this analysis during the delay and target-on periods. This revealed different patterns of outcome encoding for left-preferring and right-preferring neurons: the left-preferring neurons exhibited significantly greater outcome encoding in both monkeys when they are ENs than when they are CNs; while the right-preferring neurons exhibited opposite results between two monkeys. We believe that these results do not indicate a direct contribution of outcome encoding to the association formation. The detailed results are shown in below:



Second, we examined whether there was relationship between the strength of outcome encoding and the strength of saccade direction encoding for each 7a neurons. We found significant positive correlations between the strength of outcome encoding and the strength of saccade direction encoding for the majority of the recording sessions. However, such correlations did not exhibit systematic change as the learning progressed, shown as follows:



The figure above shows the correlation between the strength of outcome encoding and saccade encoding across different learning epochs, with each dashed line representing data from a single FOV.

Notably, as described earlier, we conducted new analyses demonstrating that outcome selectivity in area 7a may drive the monkeys' learning behavior in subsequent trials. Specifically, stronger outcome selectivity led to increased exploration behavior (i.e., switching choice direction for the same association), as shown in new Figure 4E-4F and Figure S15. This effect was observed in both correct-preferring and error-preferring neurons across monkeys. These results suggest that outcome signals in area 7a may not directly contribute to forming sensorimotor associations during learning but instead likely facilitate exploratory behavior.

5) Network connectivity lines 296ff and Figure 6: Potentially an interesting results. But between what neurons (tuning, entry criterion) was the noise correlation measured?

Response: We realized that the description in the original manuscript might not be clear. In the original Figure 6A-6C, we calculated the noise correlation between each pair of recorded neurons for each FOV. We have added this information in the revised manuscript as follows:

"We calculated the noise correlation between each pair of recorded neurons for each FOV. This reveled that noise correlation was greater for within-type pairs than cross-type pairs for the outcome-selective neurons (ENs /CNs, Figure 6A), indicating that neurons that showed similar neural encodings were more likely to coactivate"

In the original Figure 6D-K, we only included data from specific group of neurons for each of the analysis. We defined different groups of neurons based on their outcome selectivity in two successive learning days. We have specified the neuron data in both the result section and the figure legend.

Which time window did you use? As you have effects of previous trial outcome represented in some of the time windows, you would expect some correlation due to this signal. How did you take this into account and investigate/eliminate this effect on correlations?

Response: We calculated noise correlation using neural activity from frames 1–8 (1.1 s) following reward delivery. To minimize potential confounds from outcome encoding, noise

correlations were computed separately for correct and error trials, and a weighted average was used as the estimated noise correlation. Since most neurons did not exhibit significant encoding of outcome history during this period, we believe that previous outcome encoding did not substantially influence the findings presented in Figure 6. As a control, we also calculated noise correlation using neural activity during the fixation period (8 frames prior to cue onset), applying a weighted average of noise correlations across trials with different outcome histories. This analysis produced similar results.

Figure 6F,G,J,K need clearers description in the figure legends. What data am I looking at and what is the critical piece of data shown.

Response: We realized that the description in the original manuscript might not be clear. In the revised figure legend, we have added more detailed description of the results, shown as follows:

"(F) An ROC analysis was used for quantify the difference in noise correlation between within-future-type pairs and across-future-type pairs. The area under curve (AUC) value quantifies the predictive power of noise correlation for neuron ensemble evolution. The red and gray lines indicate the ROC curves for outcome-selective and outcome-non-selective neurons, respectively. Data are from the same example session shown in (E). (G). Comparison of the predictive power for neuron ensemble evolution between noise correlation and signal correlation. Each dashed line connects data from the same day. Red and gray dots represent results from outcome-selective and outcome-non-selective neurons, respectively. The red horizontal line represents the average value. The green dots denote data from the example session. Noise correlation was significantly different between within-future-type and across-future-type pairs for neurons which were outcome-selective on the second day, but no such difference was observed for neurons which were outcome-non-selective on the second day."

"Data in (J), (K) were shown in the same format as in (F), (G)."

Minor:

Abbreviations: the many abbreviations make the text not very readable, e.g. AL, ISAL could be written out as they are not standardly abbreviated terms.

Response: We thank the reviewer for pointing this out and agree that the original manuscript contained an excessive number of abbreviations, which may have led to confusion. In the revised version, we have substantially reduced the use of abbreviations to improve readability. However, we have retained a few key abbreviations—such as AL (associative learning), ISAL (image-saccade associative learning), and PPC (posterior parietal cortex)—as they are frequently used throughout the manuscript and help maintain conciseness.

Introduction: l51: sentence is unclear and grammatically incorrect.

Response: Thanks for pointing this out. In the revised manuscript, we have corrected it as

follows:

"And what factors govern these changes?"

Figure 1D. Legends states data are from each monkey. What part is from what monkey?

Response: We realized that the description in the original manuscript might not be clear. Figure 1D-1E included data from both monkeys. In the revised manuscript, we have distinguished the data from the two monkeys in different colors, shown as follows:

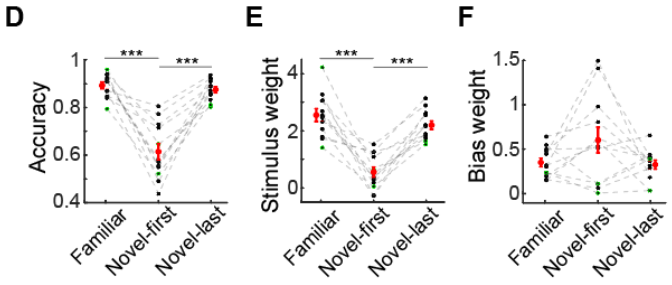


Figure: the labelling by letter seems haphazard in most figures and does not follow a clear pattern. Some panels are not really visible, like e.g. the bottom of 1G

Response: We have reorganized the panels in each main figure to improve clarity, and we hope the lettering is now more intuitive in the revised versions.

Figure legends: it would be helpful if the main outcome for each panel is summarised as well as only describing what is plotted.

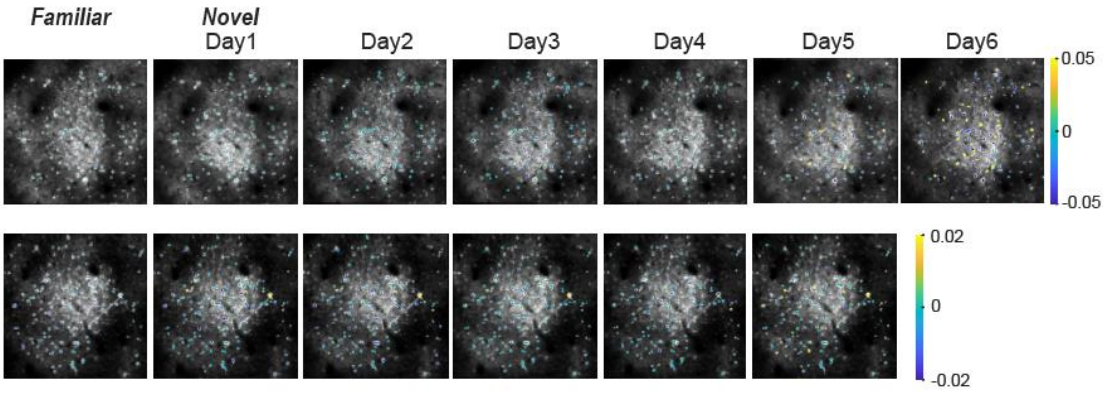
Response: We thank the reviewer for pointing this out. Following the reviewer' suggestion, we have added short summaries for most of the important findings in the revised figure legend.

P-values for stats are sometimes "p" (Methods) and sometimes "P" (Results). Very confusing as you also look at proportions. I suggest to standardise to "p =" and always write directly after naming the test.

Response: Thanks for the correction. We have changed them all to 'P' in the revised manuscript.

Labelled neurons in Figure 4A and 4F are not easily discernible.

Response: We realize that our original description may have been unclear and may have led to confusion for the reviewer. In the original Figures 4A and 4F, we presented the averaged images of two example FOVs without labeling individual neurons. The purpose of these images was to illustrate the stability of recordings across days. In the revised figures, we have now labeled individual neurons and added a color code indicating outcome encoding to improve clarity, as shown below:



Reviewer #3 (Remarks to the Author):

This study investigates the changes in the activity of cells in the primate area 7a, measured with two-photon calcium imaging, during an associative learning task that demands new visual-motor associations over multiple days. The results showed a strong neural representation of the trial outcome, which was closely correlated with monkeys' learning. This neural outcome signal was reorganized as monkeys transitioned from exploitation to exploration and showed a gradual evolution while the animals learned new visual-motor associations. In addition, the encoding of trail outcome was not closely related to the cell encoding of the output saccadic movements. Finally, the modulation in outcome encoding was constrained by the local network connectivity.
This is a very interesting and well performed study, providing novel information about how the PPC incorporates local outcome signals to promote visual-motor associations along multiple days. An important strength of this manuscript is the use of calcium imaging with two-photon that allows the measurement of the activity of the same population of neurons during periods of learning that last days. Another strength in the study is the use of a simple task where two images are associated with two saccade directions over a period of days, which permits to measure the reconfiguration of the cell and network responses during the process. Nevertheless, a have some concerns.

Response: We appreciate the reviewer's positive feedback on our work. As detailed below, we have addressed all comments, which have been very helpful in improving the manuscript.

First, it is difficult to know whether the data of the figures are from the monkey with stable saccadic behaviors or some of them are from the monkey with high frequency of fixation breaks. A direct comparison of the functional properties of 7a neurons between monkeys is in place.

Response: We realize that our original description may have been unclear and may have caused confusion for the reviewer. We also agree that a direct comparison of the functional properties of area 7a neurons between the two monkeys could provide valuable insights

to the manuscript. In the original version, most of the population-level results presented in the main figures included data from both monkeys, with a few exceptions noted below.

First, the original Figures 2N to 2R included data only from the monkey that exhibited stable saccadic behavior. In these panels, we demonstrated that PPC neuronal activity encoded outcome history from the previous trial. The other monkey showed a high frequency of fixation breaks (fix-break ratio = 0.57 ± 0.08) during task performance, resulting in an excessive number of trials with no choice responses interspersed between completed trials. Consequently, data from this monkey were excluded from the main figures (2N to 2R) but presented separately in a supplementary figure (Figure S6) in the original manuscript. These supplementary data also revealed significant encoding of previous-trial outcomes in area 7a neurons, although the encoding strength was weaker compared to the monkey with stable saccadic behavior.

Second, the original Figures 3A to 3D included data only from the monkey with a high frequency of fixation breaks. This is because we were able to record both the learning task and the control saccade task exclusively in this monkey. Conversely, the original Figures 3E to 3G included data only from the monkey with stable saccadic behavior, as only this monkey experienced learning of associations both with and without punishment for error trials across different recording sessions. These details have been clarified in both the Results section and the figure legends in the revised manuscript.

Following the reviewer's suggestion, we now present data from both monkeys separately for most results in the supplementary figures, except for the analysis of the evolution of outcome encoding during learning. This exception is because the monkey with stable saccadic behavior was successfully recorded across a complete learning course in only two FOVs. Comparisons of neuronal encoding between the two monkeys are provided in the revised Figures S1, S2, S6, S11, S13, and S15.

Furthermore, we have clearly specified the data corresponding to each monkey in the legends of all relevant figures.

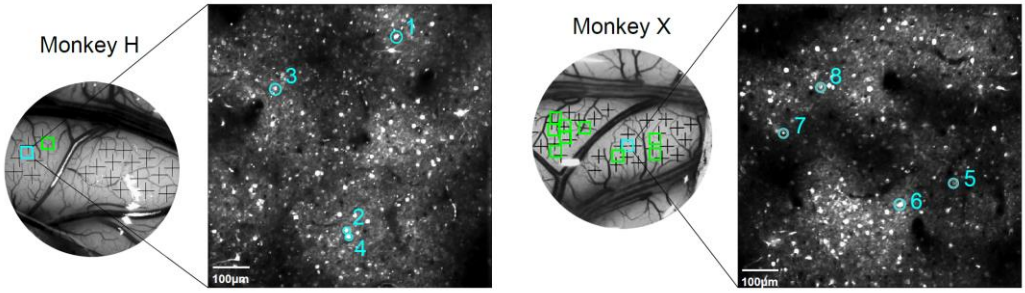
The heterogeneity in the outcome signal modulation across FOVs is large, that should be clear in the paper and something needs to be say about what is the roles of such heterogeneity in AL. In addition, it is important to know where the FOV where within area 7a, which is a large structure and limits with 7ab and 7m that have different connectivity and functional properties.

Response: We agree with the reviewer that the heterogeneity of outcome representation across FOVs is worth discussing. We speculate that two main factors may contribute to this variability. First, our findings suggest that outcome encoding in area 7a is linked to learning content. It is therefore possible that some 7a neurons exhibit stronger outcome encoding when the monkeys learn certain associations, but show weaker encoding when learning different ones. This heterogeneity in outcome representation may reflect the storage of distinct learning experiences or facilitate the learning of different content. Second, previous studies have shown that neurons with similar functions tend to be

clustered in higher cortical areas. Thus, outcome-encoding neurons in area 7a may not be uniformly distributed but rather spatially clustered in subregions within area 7a. In the revised manuscript, we have added a sentence in the Discussion section to address this potential heterogeneity of outcome representation across FOVs, as shown below:

'This content-dependent outcome encoding may also explain the heterogeneous evolution of outcome signals across different 7a subregions (FOVs) throughout the learning process. Beyond potential clustering of outcome-encoding neurons, such heterogeneity might reflect the storage of distinct learning experiences or support the acquisition of different types of content.'.

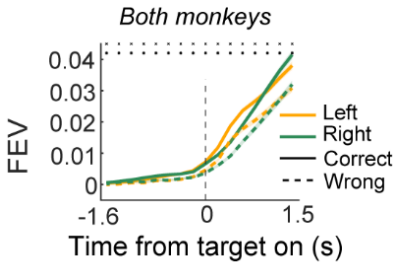
Furthermore, we have labeled all recorded FOVs in the revised Figure 1 (shown in below), clearly showing that they are all located within area 7a. However, it is important to note that our recordings did not cover the entire extent of area 7a in either monkey, due to technical limitations inherent to two-photon calcium imaging in behaving monkeys.



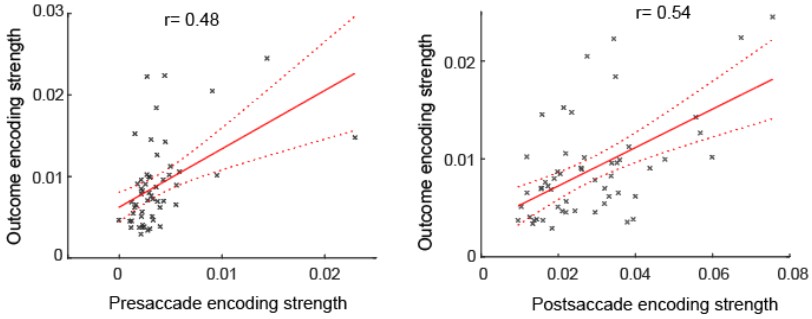
The present results describe how outcome encoding changes during long-term AL, not how the outcome signals strengths the visuomotor associations during learning. The same question is valid for the observation that neurons coactivated more closely were more likely to develop strong connections over time, leading to similar outcome encoding, but the question remains of how this encoding signal directly affects the association between image-saccade direction across days.

Response: The reviewer raised a good point which did not clearly address in the original manuscript. To fill this gap, we have included two new analysis on the saccade direction encoding during the delay period-which likely represent the image-saccade associations-in the revised manuscript.

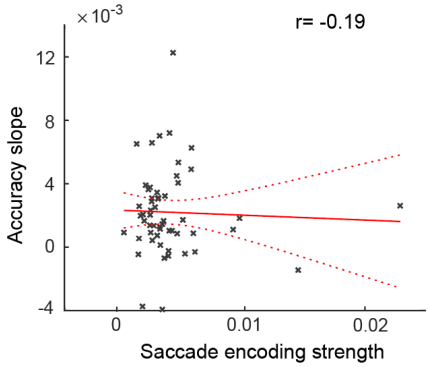
First, we examined whether the saccade direction encoding during the delay interval (which potentially encoded the image-saccade association) was influenced by the outcome history (outcome in the preceding trial). We found that saccade direction encoding—which likely reflects image-saccade associations—was very modestly, but significantly, stronger when the monkey's saccade choice was correct in the preceding trial compared to when it was incorrect. This finding suggests that outcome signals itself may modulate saccade direction encoding in the subsequent trial, potentially contributing to the learning of new associations. These results are included in the revised manuscript as Figure S16, and shown in below:



Second, we examined whether the average strength of outcome encoding correlated with the average strength of saccade direction encoding across different learning days for all the neurons within each FOV. On the population level, we found a significant positive correlation between strength of outcome encoding correlated and strength of saccade direction encoding across different learning days. This result is now included as revised Figure S10, shown as bellow:



However, although the encoding of image-saccade association (saccade direction selectivity during the delay period) within area 7a was influenced by the outcome history, the saccade direction encoding in area 7a did not exhibited systematic relationship with the monkeys' behavior performance across learning days (shown in below).



Notably, as described earlier, we conducted new analyses demonstrating that outcome selectivity in area 7a may directly influence the monkeys' learning behavior in subsequent trials. Specifically, stronger outcome selectivity led to increased exploration behavior (i.e., switching choice direction for the same association), as shown in new Figure 4E-4F and Figure S15. This effect was observed in both correct-preferring and error-preferring neurons across monkeys. These results suggest that outcome signals in area 7a may not directly contribute to forming sensorimotor associations during learning but

instead primarily facilitate exploratory behavior. We have substantially revised the discussion section to discuss the potential roles of outcome representations in area 7a played during AL, which is shown in below:

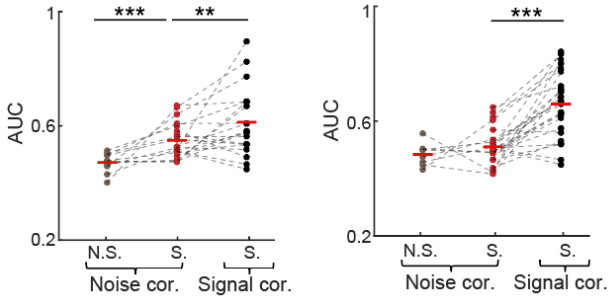
"Previous studies have identified outcome representation in brain areas outside PPC, emphasizing the significance of the frontal-striatum network in evaluating reward reinforcement. Outcome signal has been proposed to serves as a teaching signal, reinforcing correct associations while weakening incorrect ones. However, the observed outcome representation in PPC differs markedly from these earlier findings, instead highlighting a predominant sensitivity to salient, negative outcomes that may drive exploratory choices during AL. This suggests a potential complementary role of PPC in monitoring trial outcomes during sensorimotor AL, relative to the role of the frontal-striatum network. While the frontal-striatum circuitry primarily evaluates reward reinforcement and supports the acquisition and exploitation of learned associations during AL, PPC may instead play a predominant role in detecting salient 'error signals' and facilitating exploratory behavior throughout the learning process. The outcome signal in area 7a may be primarily relayed to brain regions such as the frontal pole, which are known to drive exploratory behavior during learning. This is also in line with opponent-process theories, which propose that emotional experiences and motivated behavior are regulated by pairs of opposing processes.

The critical roles of the fronto-striatal network in AL have been demonstrated in previous studies using causal approaches, such lesion studies. However, the causal involvement of the primate PPC in long-term AL remains to be tested in future studies. Our findings suggest that area 7a may not play a central role in forming or encoding sensorimotor associations during AL. First, only a small fraction of 7a neurons showed significant encoding of the visual cue during either the cue presentation (<4%) or delay (20%) periods of the ISA task. Second, encoding of saccade direction prior to the monkeys' choices was notably weak. Third, this pre-saccadic encoding did not show systematic changes across learning days that aligned with the monkeys' learning performance. The relative lack of attention to the primate PPC in earlier AL studies aligns with our observation that area 7a may not be directly involved in acquiring new associations. Instead, it may contribute to outcome monitoring—specifically by encoding salient outcome signals that promote exploratory behavior during AL. Nonetheless, the role of the primate PPC in outcome monitoring and its potential contribution to exploration-driven learning processes warranting further investigation with causal manipulations. Previous rodent studies have reported that PPC was causally involved in processing sensory-stimulus history and history-based action selection bias to guide decision behavior. Moreover, PPC was also shown to represent discrepancies between the current model and observed state transitions in reinforcement learning, as well as reward-related belief updating during flexible decision-making. Collectively, these results indicate that PPC likely plays an important role in updating the estimation of the current state by incorporating past experience, to guide decision-making and learning."

Finally, for Figure 6 I highly recommend computing the mutual information of the neural activity during the task period between cells to generate robust graphical models of functional connectivity across cell types and learning epochs.

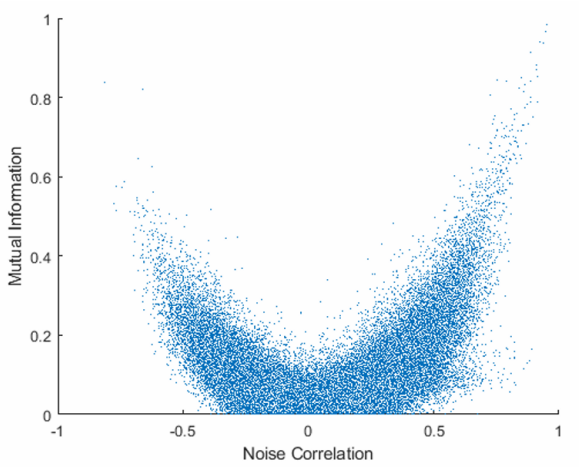
Response: We agree with the reviewer that applying graph theoretical analysis to the functional neuronal network could yield valuable insights. In a separate manuscript, we constructed functional neuron networks using partial noise correlation and applied graph theoretical analysis to the 7a neuron network. This approach provided novel insights into how network topology constrains both the functional roles and the evolution of neuronal encoding across learning days.

In response to the reviewer's suggestion, we also performed the analysis in Figure 6 using mutual information instead of noise correlation. However, this approach did not produce satisfactory results compared to the original analysis, as functional connectivity based on mutual information failed to predict the evolution of neuronal encoding during learning. This result is shown below:



As shown in the figures above, while functional activity measured by mutual information could barely predict changes in neural encoding across learning days (left panel, same group of neurons in day 1 but different groups of neuros in day2, mean AUC = 0.55; right panel, different group of neurons in day 1 but same group of neurons in day2, mean AUC = 0.51), its predictive power was not greater than that of signal correlation. Most notably, this predictive power was substantially weaker than that derived from noise correlation (Figure 7G, mean AUC = 0.75; Figure 7K, mean AUC =0.76).

We believe this difference arises primarily because mutual information lacks a critical feature necessary for our analysis—its inability to distinguish between positive and negative connectivity. Although mutual information and noise correlation values are highly correlated (as shown in the figure below), mutual information only yields positive values, whereas noise correlation captures both positive and negative values, allowing us to differentiate between excitatory and inhibitory (or anti-correlated) relationships. In our data, neurons with opposite encoding preferences tended to exhibit negative functional connectivity. Therefore, mutual information may not be an appropriate metric to fully represent the connectivity between neurons in this study.



Minor comments

Please describe what the PsyTrack parameters are and why this model is better than others to describe AL.

Response: To capture the dynamics of the monkeys' behavioral strategies during learning, we fitted their performance accuracy using the 'PsyTrack' model. This model estimates time-varying weights that characterize the animals' decision-making strategy on a trial-by-trial basis, modeled as a linear combination of available task variables. As such, it allows for trial-resolution tracking of how decision strategies (reflected by different weights) evolve throughout the learning process. Specifically, we used the task condition (cue identity) as the model input, coding each trial as either -1 or 1, while the monkeys' saccade choices served as the output, coded as 0 or 1 per trial.

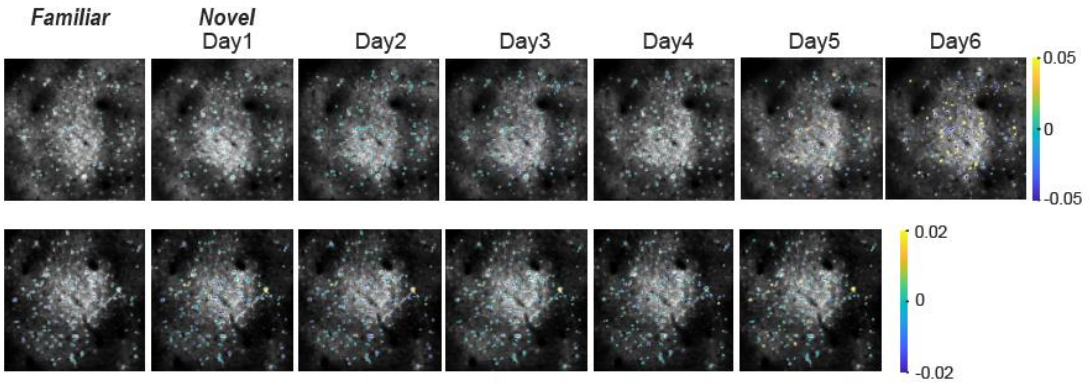
For model fitting, we applied three hyperparameters: 'sigInit' = 2^4 (controlling the prior variance of initial weights), 'sigma' = 2^-4 (determining the smoothness of weight evolution across trials), and 'sigDay' = 2^-1 (capturing potential day-to-day shifts in weights). The model yielded three behavioral weights: bias weight, previous answer weight, and stimulus weight, reflecting the degree to which the monkeys' saccade choices depended on internal bias, the previous trial's outcome (the extent to which the monkeys' saccade choice was dependent on the win-stay, lose-shift strategy), and the visual cue, respectively. These details were provided in the Methods section; we have now also added one sentence in the Results section to describe the PsyTrack model fitting procedure and its advantages, as shown below:

'This model describes choice behavior at the resolution of single trials, allowing us to quantify the evolution of animal's decision strategies (represented by different weights) across the learning course.'.

The change of outcome signaling in the population showed in Figure 4 is astonishing. Please add the color code for outcome encoding of Figure 2C in Panels 4A and F.

Response: Following the reviewer's suggestion, we have added the color code for outcome

encoding in the revised Figure 5A and F, shown as follows:



Is the outcome signal in PPC used locally for AL or is also used in other areas?

Response: This is an interesting and important question; however, the current study cannot directly address it. As we detailed in the previous response,-area 7a might not be the key brain region for acquiring and representing the sensorimotor associations, and outcome encoding in area 7a may primarily drive exploration behavior during learning. Therefore, we believe it is more plausible that the outcome signals are transmitted to other subregions such as the frontal pole, where they may directly contribute to drive exploratory choices. However, because our recordings were primarily limited to cortical layers 2/3, without access to deeper layers, it remains possible that outcome signals in deeper cortical layers of area 7a could participate in learning and presenting sensorimotor associations. We have discussed this possibility in the revised manuscript as follows:

'While the frontal-striatum circuitry primarily evaluates reward reinforcement and supports the acquisition and exploitation of learned associations during AL, PPC may instead play a predominant role in detecting salient 'error signals' and facilitating exploratory behavior throughout the learning process. The outcome signal in area 7a may be primarily relayed to brain regions such as the frontal pole, which are known to drive exploratory behavior during learning.'

How the complex remapping of outcome encoding linked to new associations is used to generate the AL. How the previous and actual trial outcome signal affects the image-saccade association, when the mapping of outcome encoding remapping is to complex. What is the outcome signal doing to generate the association between the image and the saccade direction?

Response: We thank the reviewer for these insightful questions. In this study, we found that outcome encoding in area 7a underwent significant remapping as the monkeys transitioned from familiar to novel associations. This suggests that outcome encoding in area 7a is not abstract but closely tied to the specific content of learning. Consequently, distinct neuronal ensembles may contribute to representing trial outcomes depending on the specific associations being learned. We speculate that PPC neurons exhibiting strong

outcome encoding during the learning of specific associations may become functionally linked to neurons representing these associations, thereby influencing the formation of sensorimotor associations during learning. Such content-dependent outcome representations may provide an advantage over abstract outcome signals by enabling outcome-encoding neurons to directly influence the neuronal ensembles representing specific associations, without requiring more complex targeting mechanisms. Consistent with the notion that the outcome encoding in area 7a was related to learning content, we found that stronger outcome selectivity was specifically linked to switching saccade choice direction for the same imaging-association rather than simply switching saccade directions irrespective to the associations. We have added several sentences in the revised Discussion section to discuss this point as follows:

"Outcome signal has been proposed to serves as a teaching signal, reinforcing correct associations while weakening incorrect ones."

"We speculate that PPC neurons exhibiting strong outcome encoding during the learning of specific associations may become functionally coupled with neurons representing these associations, thereby influencing the learning of sensorimotor associations. Such content-dependent outcome representations may provide a computational advantage over abstract, content-independent outcome signals by enabling outcome-encoding neurons to directly influence the neuronal ensembles representing specific associations, without requiring more complex targeting mechanisms."

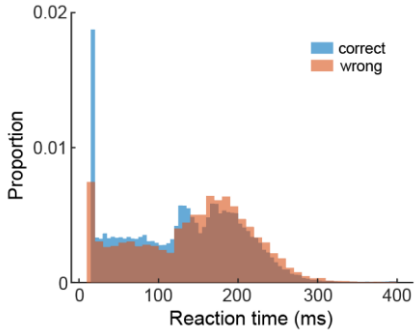
An intriguing question concerns the precise role of the outcome signal during the learning of novel associations. Previous studies have proposed that the outcome signal serves as a teaching signal, reinforcing correct associations while weakening incorrect ones. This mechanism enables the brain to determine, through trial-and-error learning, which saccade direction is correctly linked to a given visual image. As noted in our previous response, we speculate that this learning process may primarily occur in other brain regions that are directly responsible for acquiring and storing sensorimotor associations. Instead, the outcome encoding in area 7a might primarily facilitate the exploration choice during learning, as we demonstrated that greater outcome encoding in area 7a lead to saccade choice switches during learning.

The outcome-history encoding of the majority 7a neurons predominantly emerged and localized within the fixation period of the ISAL task, so these signals have an effect on how the cue images are progressed or kept in memory? Or is a signal that changes the reaction time of the saccades or the saccade direction?

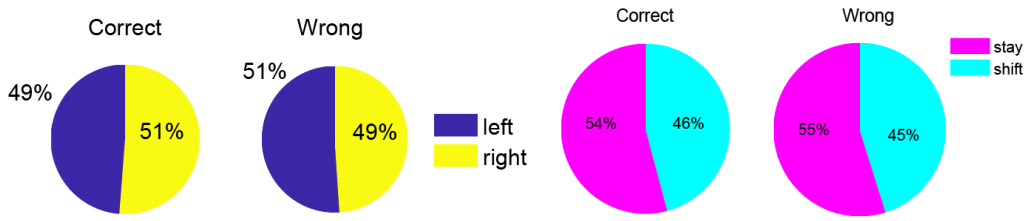
Response: This is a good question. We conducted five analyses to address it. First, as described in previous response, we examined whether outcome history influenced the encoding of visual cues during the cue presentation and delay periods of the learning task. Our analysis revealed that saccade direction encoding—which likely reflects image-saccade associations—was slightly stronger when the monkey's saccade choice was correct in the preceding trial compared to when it was incorrect. This effect was observed

in both left-preferring and right-preferring neurons. These findings suggest that outcome signals may modulate saccade direction encoding in the subsequently trial, potentially contributing to the learning of new associations.

Second, we tested whether the outcome history of the previous trial influenced the monkeys' reaction time (RT) in the current trial. This analysis revealed no significant difference in RT during most recording sessions(46/59, the averaged result is shown below）between trials preceded by a positive outcome and those preceded by an negative outcome. This result is not surprising, given that our task design included a fixed delay period between stimulus offset and target onset, which likely minimized the influence of prior outcomes on reaction time.



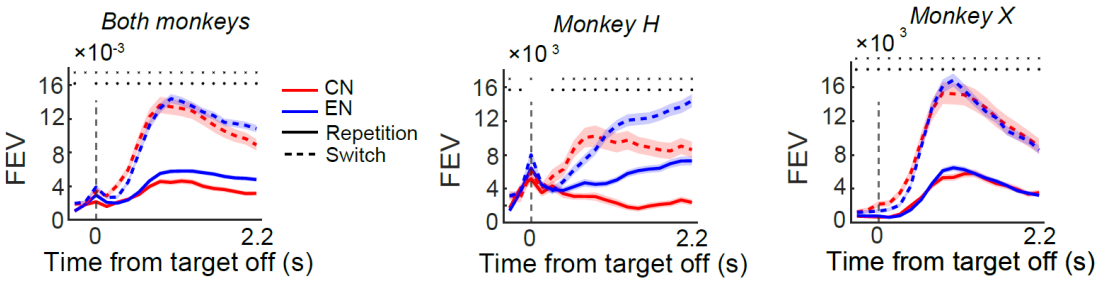
Third, we examined whether outcome history in the previous trial influenced the monkeys' saccade direction in the current trial in two different ways. First, we tested whether the monkeys exhibited saccade direction bias when previous trial was correct or incorrect. This revealed no significant bias in the majority of learning sessions (40/59). Second, we tested whether the outcome history effected the monkeys saccade choice strategy, i.e.' win-stay, lose-shift. Our results showed that the outcome history significantly influenced the monkeys' choice strategy in a minority of the learning sessions (21/59). The averaged results are shown in bellow:



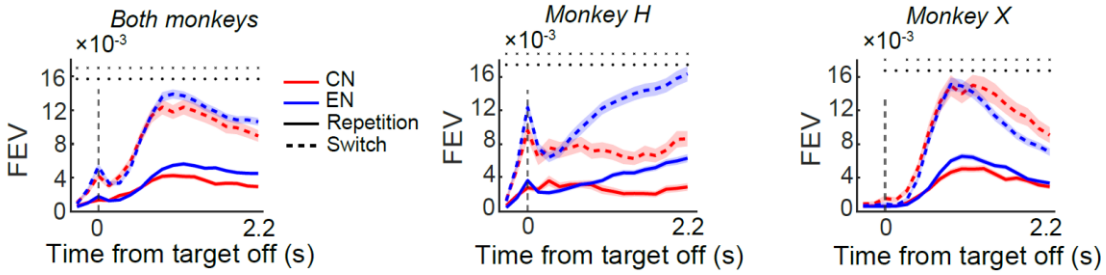
Left two panels: the percentages of trial that the monkeys chose different saccade directions followed correct or incorrect outcomes; right two panels: the percentages of trials using different choice strategies followed correct or incorrect outcomes.

Fourth, we have included new analysis to investigate whether outcome encoding in area 7a was modulated by outcome history, specifically the outcome of the preceding trial. We quantified the encoding strength separately for trials following outcome switches (correct→error or error→correct transitions) versus outcome repetitions (correct→correct or error→error). Notably, outcome encoding was significantly stronger following outcome

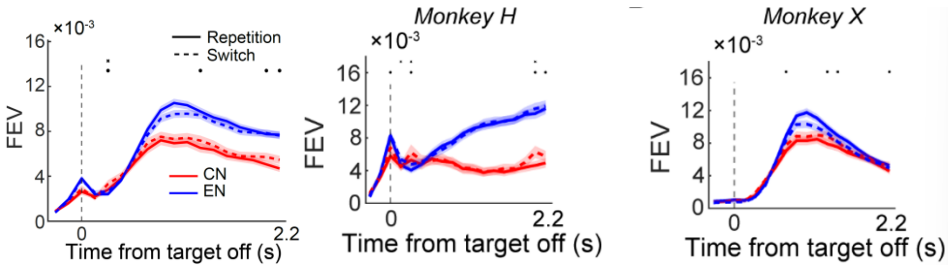
switches compared to outcome repetitions (new Figure 3H). This effect was observed for both error-preferring and correct-preferring neurons across two monkeys (new Figure S13). These findings suggest that outcome encoding strength in area 7a was remarkably influenced by the outcome history during learning, and that area 7a is particularly sensitive to outcome changes. This observation aligns with the well-established role of the primate PPC in salience representation. Detailed results are shown in bellow:



Lastly, we conducted new analyses to test whether outcome selectivity in area 7a directly influenced the monkeys' learning behavior in the subsequent trial. Trials were categorized into two conditions: exploration, in which monkeys switched saccade direction relative to the previous trial for a given image-saccade association, and exploitation, in which they repeated the same saccade direction for the same association. For each condition, we measured outcome selectivity in the preceding trial. Outcome selectivity was significantly stronger prior to exploratory trials compared to exploitative ones (new Figure 4E and Figure S15), suggesting that enhanced outcome encoding in area 7a predicted subsequent exploratory behavior. The phenomenon was found for both correct-preferring and error-preferring neurons across two monkeys. In contrast, area 7a neurons did not show increased outcome selectivity prior to trials in which the monkeys switched saccade direction independently of the image-saccade association (new Figure 4F and Figure S15). These findings suggest that outcome representations in area 7a may contribute specifically to strategy shifts during sensorimotor association learning, rather than reflecting general switching behavior. The detailed results were shown as follows:



These figures plot the strength of outcome encoding (quantified by FEV) for trials prior to the exploration and exploitation trials respectively.



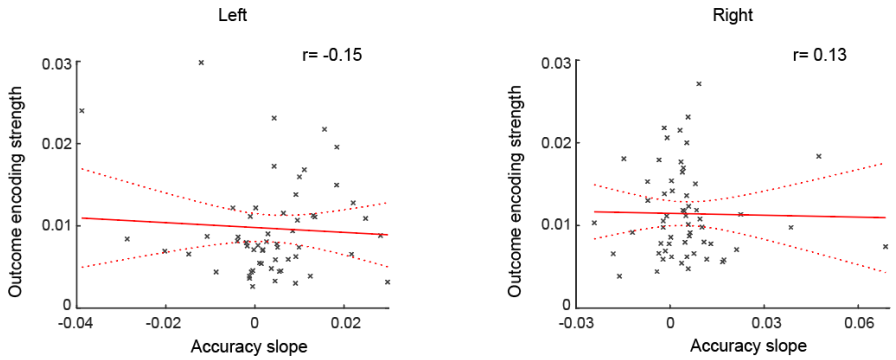
These figures plot the strength of outcome encoding (quantified by FEV) for trials prior to saccade direction switching and repeating trials independently of the image-saccade association. The left panel shows the averaged results from two monkeys.

What is the functional meaning of having outcome signals that are different between the actual and the previous trial?

Response: As the reviewer noted, a substantial fraction of neurons exhibited inconsistent encoding of trial outcomes or outcome history. We speculate that this variability may not reflect specific functional roles, but rather the dynamic nature of outcome encoding within the network, either within area 7a or across interconnected brain regions. The maintenance of outcome information may rely not only on persistent firing during the inter-trial interval but also on the overall population activity state and potentially short-term synaptic plasticity. Similar dynamic encoding of information during delay periods has been reported at both the single-neuron and population levels in previous studies (Spaak et al. ,2017; Stokes，2015).

What is the effect of the speed of learning (e.i. 3 vs 7 days) on the magnitude of the encoding of outcome by neurons and on dynamics across days.

Response: This is an excellent point. Following the reviewer's suggestion, we calculated the correlation between the magnitude of outcome encoding and the monkeys' learning speed. Given the heterogeneity of outcome representation across different FOVs, we applied a multiple-class linear regression using the average strength of outcome encoding for each recording session, with the corresponding average learning speed as a regressor while controlling for FOV differences. We calculated the daily learning speed for each image-saccade association separately for each learning session. This analysis revealed no systematic correlation patterns between learning speed and the strength of outcome encoding across different 7a subregions (FOVs). The detailed results are shown in below:



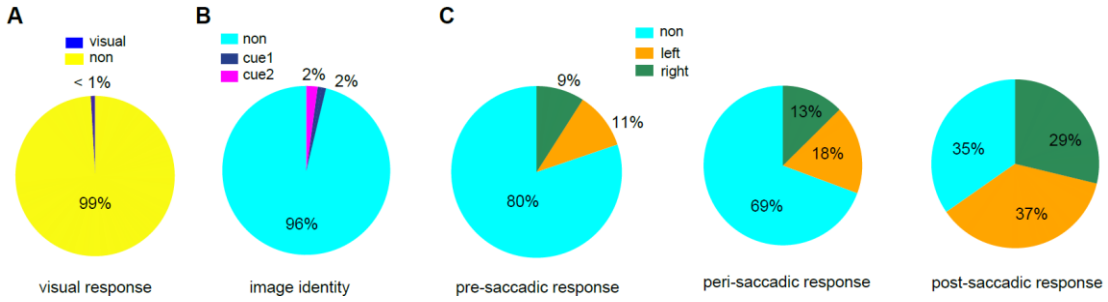
However, we believe that the absence of a clear correlation does not rule out a functional role for outcome encoding in associative learning. This is because outcome encoding in area 7a appears to be content-dependent—that is, the recorded neurons in each session may have contributed to real-time learning to varying degrees (fully, partially, or minimally). Such variability could account for the inconsistent relationship observed between outcome encoding strength and learning speed. Moreover, as outlined in our previous response, outcome encoding in area 7a may primarily drive exploratory behavior during learning. Since exploratory behavior may not consistently influence learning speed across different learning stages, this could further obscure a systematic relationship between outcome encoding strength and behavioral improvement.

What is the proportion of cells that encode the stimulus onset, the image identity, the saccade movement preparation and execution and the outcome. How many neurons encoded more than one parameter, and how the outcome signal changes during AL modified the encoding of the image and the saccade direction in the same cells or in other populations.

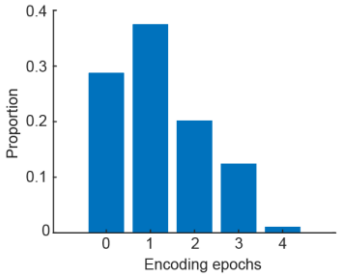
How many neurons encode both the previous and the actual outcome as a function of learning.

Response: In the revised manuscript, we have included additional analyses to illustrate these results, summarized as follows. First, beyond reporting the proportion of neurons showing significant responses to stimulus onset (visual response), saccade preparation (pre-saccadic response), and post-saccadic response, we have now added the proportion of neurons that exhibited significant encoding of image identity and saccade execution (peri-saccadic activity) in the revised Figure S9. Second, we quantified the proportion of neurons exhibiting various forms of mixed selectivity, and these results are also presented in the revised Figure S9. Third, we conducted a new analysis to examine how changes in outcome signals during AL influenced the encoding of image identity and saccade direction. Specifically, we assessed the correlation between the strength of outcome encoding and the strength of saccade direction encoding during the delay period across different learning days. This analysis revealed a positive correlation at the population level ($r = 0.48$), as shown in the new Figure S10. Fourth, we quantified the proportion of neurons encoding both the previous and current trial outcomes as a function of learning progress. This revealed a decreasing trend of neuron encoding both trial outcome and outcome history after the monkeys fully learned the associations.

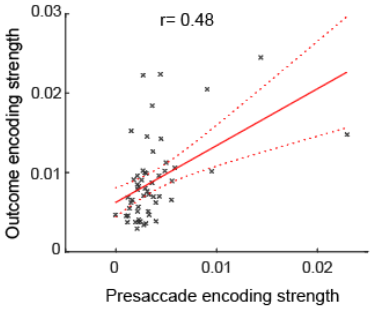
All of these revised and additional results are presented in the figures below.



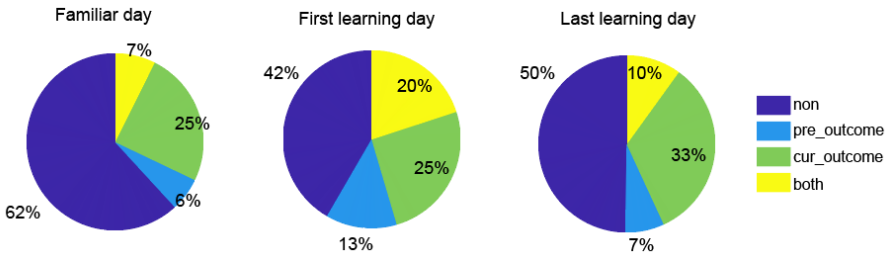
These figures show the proportion of neurons exhibiting different selectivity.



This figure plots the proportion of neurons exhibiting different levels of mixed encoding.



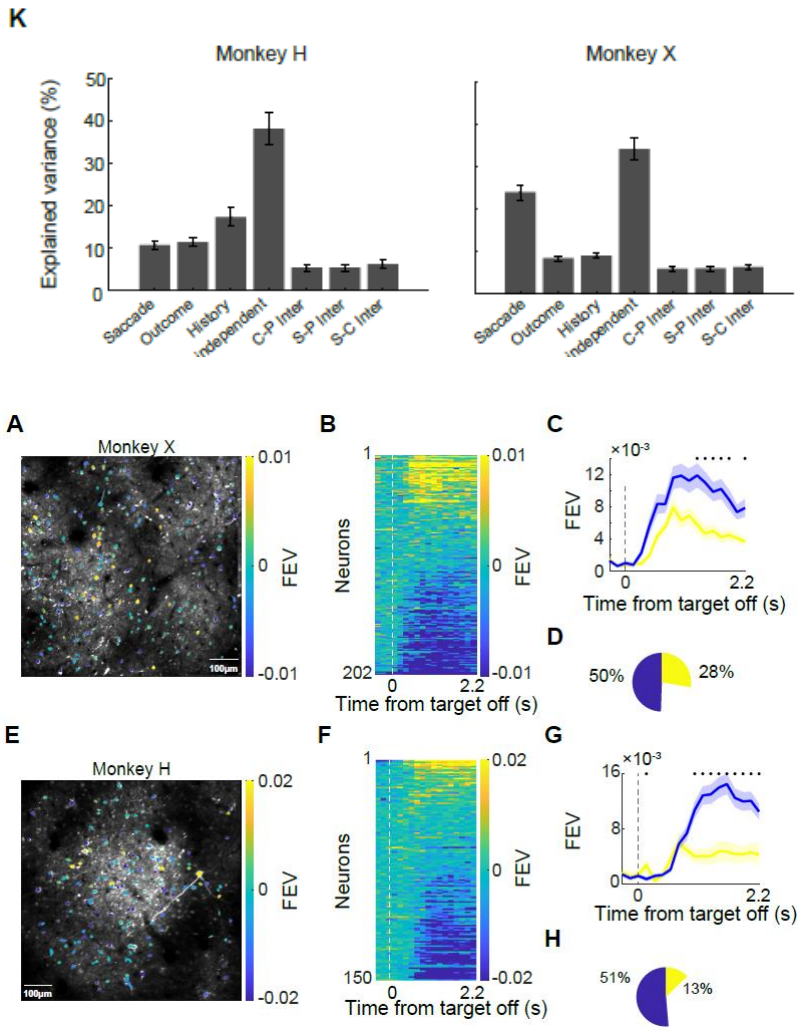
This figure plots the correlation between outcome encoding and saccade direction encoding during the delay period.



These figures show the proportion of neurons exhibiting significant encoding on current and previous outcomes across different learning epochs..

Please provide Figures of the FOV with pronounced outcome selectivity for the second monkey as in Figure S1 and S9.

Response: We apologize for the unclear description in the original manuscript. In the revised version, we have provided more detailed annotations distinguishing data from the two monkeys in the figures. Actually, the upper and lower panels in Figure S1 show data from the two monkeys respectively. Additionally, we have included the results for the second monkey in the revised Figure S11 (original Figure S9). The updated figures are shown below.



Typos.

There are many typos in the manuscript such as:

from two monkeys. (see Methods).

Figure.2C

Response: Thanks for the correction, we have corrected them all.

Authors' Response to Reviewers

We sincerely thank all reviewers for their thoughtful and detailed feedback on the earlier version of our manuscript. Their comments and questions have greatly helped us to improve the quality and clarity of our work. We also appreciate the reviewers' positive recognition of our previous revisions and responses. In this new version of the manuscript, we have carefully addressed the latest comments from the reviewers.

Specifically, Reviewer 1 suggested us to invite a native English speaker to improve the writing. Following the suggestion, we invited a native English speaker to polish the manuscript language to enhance the clarity.

Reviewer 2 expressed no further concerns or questions.

Reviewer 3 recommended enlarging and clarifying the black stars denoting significant effects in the figures and suggested adding new references. Accordingly, we have updated the corresponding figures and incorporated the suggested references in the introduction section of the revised manuscript.