Chapter 4
Sensory representation of a cued tactile stimulus in the posterior parietal cortex of the mouse

(submitted)
Sensory representation of a cued tactile stimulus in the posterior parietal cortex of the mouse

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Abstract

The cognitive process of attention allows for the selective processing of salient stimuli over others, when we are constantly challenged with multiple stimuli that engage our senses. The fronto-parietal attention circuit in the mammalian neocortex, of which the posterior parietal cortex (PPC) is part of, plays a role in the selection of pertinent stimuli. However, how a sequence of sensory information, where one stimulus is cued by another, is represented in PPC during attention is poorly understood. Here, using 2-photon imaging in awake behaving mice, we show that neurons in layer 2/3 of the mouse PPC can reliably represent texture touch events, in addition to auditory cues that predict the incoming tactile stimulus. Notably, omission of the cued texture touch elicited a large response in a subset of neurons that were largely non-responsive to or inhibited by the tactile stimuli. A higher proportion of PPC neurons were able to detect the stimulus omission, as compared to discriminating stimulus features (texture graininess). Our results suggest that the layer 2/3 network in PPC participates in stimulus encoding and could potentially further function as a reporter of mismatch signals under conditions of stimulus omission.

Significance statement

The posterior parietal cortex (PPC) is implicated in a variety of functions, ranging from spatial navigation and attention to multisensory processing and perceptual decision making. Here we show that in addition to discriminating stimulus features, the PPC can signal the omission of a cued stimulus and potentially function as a reporter of stimulus mismatch.

Introduction

Sensory processing carried out by the mammalian neocortex allows for the perception of the external environment. However, the brain is not capable of equally treating the multitude of sensory stimuli detected by the sensory organs. It therefore selectively shifts attention to salient or relevant stimuli over others, to achieve current internal goals. Top-down stimulus selection in the attention network is distributed across the fronto-parietal-
collicular areas (Womelsdorf and Everling, 2015), of which the posterior parietal cortex (PPC) is part of. In rats it is reciprocally connected to the orbitofrontal cortex, anterior cingulate cortex and the frontal eye field (FEF) (Kolb and Walkey, 1987), and together PPC and FEF form the fronto-parietal attention network. PPC in addition has dense reciprocal connections with the surrounding sensorimotor and visual cortices (Kolb and Walkey, 1987; Reep et al., 1994; Wang et al., 2012) and is capable of processing somatosensory, visual and auditory stimuli, as well as combined stimuli resulting in multisensory enhancement (Wallace et al., 2004; Olcese et al., 2013). PPC is also implicated in sensorimotor decision making in both primates and rodents (Shadlen and Newsome, 2001; Harvey et al., 2012; Raposo et al., 2014; Goard et al., 2016) and more recently in goal distance estimation with auditory cues (Funamizu et al., 2016). Lesion studies show that damage to the PPC results in failure to attend and respond to contralesional sensory stimuli. This hemispatial neglect occurs despite intact sensory processing at the primary sensory cortices (Corbetta and Shulman, 2011). It can be alleviated partially by the presentation of cues to direct attention to the contralesional side (Riddoch and Humphreys, 1983; Robertson et al., 1998). Little is known about how multiple sensory stimuli are encoded in the neuronal microcircuits of PPC, in particular when the presentation of one salient stimulus occurs in sequence with distinct preceding cues and thus might be expected. In mouse primary visual cortex (V1), mismatch between the actual and expected visual feedback generates mismatch signals in a subset of L2/3 neurons (Keller et al., 2012; Fiser et al., 2016). Interestingly, these mismatch signals are also observed in the axonal projections of the thalamic lateral posterior nucleus (LP) to V1 (Roth et al., 2015), which is the main source of thalamic input to the PPC (Krieg, 1947; Reep et al., 1994). Hence the PPC, as part of the fronto-parietal attention network, might also participate in the representation of sensory mismatch signals.

Here we presented naïve awake mice with tactile stimuli in the form of texture touches to the intact whisker pad (Chen et al., 2013), presaged by auditory cues. We report that layer 2/3 neurons of PPC respond to both the tactile stimuli and the auditory cues that predict their delivery. Moreover, we find that omission of the textures elicits responses in a subset of neurons that were largely non-responsive to or suppressed by the texture stimuli. This omission response was present from the first omission trial, suggesting the rapid encoding and binding of sensory information within the PPC. Stimulus omission was detected by a higher proportion of PPC neurons compared to those discriminating texture graininess. We show for the first time single-cell neural correlates of an omission response in the PPC, as a potential form of mismatch signal to the missing stimulus.
Materials and Methods

**Viral and tracer injections.**

Experimental procedures followed the guidelines of the Veterinary Office of Switzerland and were approved by the Cantonal Veterinary Office in Zurich. Intrinsic signal optical (IOS) imaging was performed on young adult (P35-42) male wild-type C57/BL6 mice to identify the location of PPC by exclusion. In brief, to avoid the activation of surrounding whiskers, all whiskers except the γ-barrel-column whisker on the right whisker pad of the mice were trimmed and their locations were mapped using IOS on the exposed skull during whisker stimulation (rostrocaudal deflections at 10 Hz). The location of primary visual cortex (V1) was similarly mapped using a full-field stimulation with a green LED placed 5 mm in front of the contralateral eye. Subsequent viral or tracer injections were targeted to the non-activated region between these two identified sites, within 500-μm from the border of the IOS corresponding to the γ-barrel-column location (white dotted lines in Figure 1C). To verify that we are able to localize PPC by IOS exclusion as described above, we went on to verify the established anatomical input and output connections to PPC (n = 4 mice). To identify the sites that send their axons to PPC, the retrograde tracer CTB-Alexa594 (Molecular Probes, Invitrogen; 200 nl, 1% wt/vol) was injected into PPC ~300 μm and 500 μm below the pial surface targeting layer 2/3 and layer 5, respectively. A similar injection with AAV8-hSyn-Jaws-GFP-ER2 (200 nl, UPenn) was performed to identify output regions that PPC projects to, by following anterogradely labelled axons. For calcium imaging, AAV1-EF1α-YC-Nano140 (300 nl, ~1 x 10⁹ vg/μl) was injected into the PPC (n = 4 mice), targeting layer 2/3 (~300 μm below the pial surface) to induce expression of the calcium indicator yellow cameleon Nano140 (YC-Nano140) (Horikawa et al., 2010).

**Verification of PPC location.**

Following 5-7 days after CTB-Alexa594 injection to allow for uptake (Conte et al., 2009), and 3 weeks in the case for virus expression of GFP, mice were anesthetized (ketamine/xylazine; 100/20 mg/kg body weight) and perfused transcardially with 4% paraformaldehyde in phosphate buffer, pH 7.4. Coronal cortical sections (50-μm thickness) were cut using a vibratome (VT100; Leica). Slices were stained with 2.5 μM DAPI for 10 min and mounted on glass slides. Images were acquired with a confocal microscope (Fluoview 1000, 20x and 10x objectives; Olympus) with green (GFP), red (CTB-Alexa594), and blue (DAPI) excitation/emission filters.
Cranial window implantation and habituation.

In order to carry out long-term in vivo calcium imaging, a cranial window was implanted 24 hrs after virus injections over PPC as described previously (Margolis et al., 2012). Briefly, a craniotomy was performed at the injection site. A cover glass (4 mm diameter) was placed directly over the exposed dura mater and sealed to the skull with dental acrylic. A metal post was fixed to the skull with dental acrylic, contralateral to the cranial window, to allow for subsequent head fixation. One week following chronic window implantation, mice were handled daily for 1 week, and gradually habituated to head fixation. This also gave time for the trimmed whiskers to re-grow. Mice were subsequently used for texture presentation only if their whisker pads were fully intact.

Texture presentation.

Behaviour experiments were performed using a data acquisition interface (USB-6008; National Instruments) and custom-written LabVIEW software (National Instruments) to control devices required for texture presentation. Commercial-grade sandpaper (3M) was used, with a rough sandpaper (P100) and smooth sandpaper (P1200) as stimuli. Sandpapers were mounted onto panels attached to a stepper motor (T-NM17A04; Zaber) mounted onto a motorized linear stage (T-LSM100A; Zaber) to move textures in and out of reach of whiskers of head-restrained mice. The textures were presented in a randomized order, with no more than 3 consecutive presentations of the same texture. Trials without texture presentation (“Blanks”) but with linear stage translation were interleaved. A trial consisted of a 2-s pre-stimulus period, a 2-s translation period of the linear stage to bring the texture in contact with the whiskers where it remained for 1-s before retraction. A 2093-Hz auditory cue tone (1 ms) signalled the start of stage translation and when it stopped. Stage translation itself generated a sound, and served as an additional auditory cue that indicated the arrival of the incoming textures. Mice were not required to perform any task during the trials and were not rewarded during texture presentation.

Two-photon calcium imaging.

We used a custom-built two-photon microscope controlled by HelioScan (Langer et al., 2013), equipped with a Ti:sapphire laser system (~100-fs laser pulses; Mai Tai HP; Newport Spectra Physics), a water-immersion objective (16×LWDPF, 0.8 NA; Nikon), galvanometric scan mirrors (model 6210; Cambridge Technology), and a Pockel’s Cell (Conoptics) for laser intensity modulation. For calcium imaging, YC-Nano140 was excited at 840 nm and fluorescence collected with blue (480/60 nm) and yellow (542/50 nm) emission filters for CFP and YFP fluorescence detection, respectively. Images were
acquired at 15.6 Hz with 128 x 64-pixel resolution. Single trials of 9 to 10-s duration were recorded at a time with 1-s breaks in between trials to allow for hard disk storage during inter-trial interval periods.

**Whisker tracking.**

The whisker field was illuminated with 940-nm infrared LED light and movies were acquired at 200 Hz (500x500 pixels) using a high-speed CMOS camera (A504k; Basler). The time course of the whisker angle across all imaged whiskers was measured using automated whisker tracking software (Clack et al., 2012). Whisking amplitude, defined as the angle between the whisker shaft and sagittal plane from nose tip to the back of the head, was used as a measure to represent both rhythmic and non-rhythmic forms of whisking behavior. We used custom written MATLAB (Mathworks) scripts to extract whisking episodes by first applying a fourier transform on traced theta to identify whisking events with frequencies between 4 Hz and 20 Hz. Events with power greater than a predefined threshold were used to segregate significant whisking episodes. Time periods from these whisking episodes was used to define trial events with or without whisking behavior during the stimulus window. For all trials, the first and last time point for whisker-to-texture contact was quantified manually through visual inspection. In Figure 3C-D, the presented data is from 34 omission-selective neurons and 12 FOVs (instead of 38 neurons and 13 FOVs) as whisker tracking could not be performed on one of the sessions.

**Calcium imaging analysis.**

The CFP and YFP fluorescence channel data were imported into MATLAB for processing. First, background was subtracted on each channel (bottom 1st percentile fluorescence signal across entire video). Motion correction to both channels was performed by using image registration with TurboReg (Thevenaz et al., 1998). Regions of interest (ROIs) corresponding to individual neurons were manually selected from the average intensity projections of a single-trial time series using ImageJ (National Institute of Health) and custom scripts in MATLAB. Mean pixel value for each ROI was extracted for both channels. Calcium signals were expressed as relative YFP/CFP ratio change \( \Delta R/R = (R - R_0)/R_0 \). \( R_0 \) was calculated for each trial as the bottom 12th percentile of the ratio for the trial. Active neurons were identified by two-way ANOVA with repeated measures of the neuronal calcium signal against the neuropil signal, defined by all pixels not assigned to neuronal soma of the overall ROI annotation (significance value, \( P < 0.05 \)).
Neuronal-response classification.

Active neurons were first checked against their baseline for significant responses during the time windows corresponding to the auditory cues generated by the translation stage (from auditory tone onset to 1 s into stage translation) and the window where the textures can potentially come into contact with the whiskers as the stage moves into position (650 ms before stage stop to the time before stage retraction) for all trials. Responses that occurred during texture retraction were not considered in the classification. Active neurons were further cross-correlated with a binary vector corresponding to these time durations for the auditory cues and tactile stimulus. Neurons with significant responses and positive correlation (> 0.3) within the corresponding windows of the binary vectors were classified as either auditory cue- (hereby referred to as cue) or touch-responsive. Neurons were classified as mixed-responsive if they fell into both categories. Neurons with significant responses but negative correlation within the auditory cue- or touch-stimulus vectors were classified collectively as suppressed neurons. Neurons that did not have significant activity during the cue and touch-stimulus windows during texture trials were termed as unclassified. Neurons with a significant response during “Blank” trials in the window and showed a positive correlation with the tactile-stimulus vector were additionally classified as omission-selective neurons. Neurons that were cue-responsive during the “Blank” trials were not considered as omission-selective neurons if the calcium transient fell by more than 50% of the peak response within the cue window once the stage stopped moving, and failed to show a rise during the

Figure 1. Stimulation setup and localization of PPC in mice.

A, Schematic of the texture presentation set-up that allows for 2-photon imaging of the PPC using a cranial window, in a head restrained mouse. B, Schematic of the trial structure, where the translation stage presenting the textures slides in and out of position. The loudspeaker symbols indicate the 1-ms auditory tones just before the start and at the end of the stage translation, respectively. The shaded beige rectangles indicate the time windows with auditory cues from the translation stage movement and the green rectangle indicates the time win-
Touch-stimulus window. Based on this classification, neurons can be placed into 1 of the 5 classes, and be additionally termed as omission-selective neurons.

**Trial type analysis.**

The performance of single neurons in discriminating two distinct trial types was assessed using a receiver operating characteristic (ROC) analysis (Green and Swets, 1966; O’Connor et al., 2010). Classification of stimulus X versus stimulus Y (P100 vs P1200, Blank vs P100 and Blank vs P1200) was based on the similarity of the calcium transient in each trial to the mean calcium transient for trial type X compared to trial type Y. This analysis was confined to the possible touch-stimulus window. Each trial

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**Figure 2. Neural dynamics of sensory representation in the PPC.**

**A,** Left, 2-photon image of a FOV with neurons expressing the GECI YC-Nano140. Right, calcium transients from the ROIs selected in the left panel in response to the presented auditory cues and textures, with the corresponding whisker angles (Wh) in green. The transients represent the very first 5 trials that the mouse has been exposed to. The averaged responses correspond to: P100, n = 57; P1200, n = 26; Blank, n = 17 trials. **B,** Example average calcium transients of neurons classified as cue-responsive, touch-responsive, mixed-responsive, suppressed and unclassified. Traces are averages of 40 to 60 P100 trials.
was assigned a “touch-window stimulus discrimination variable” score \((DV)\) equal to the dot-product similarity to the mean calcium transient for trial type \(X\) minus the dot-product similarity to the mean for trial type \(Y\). Thus, for trial type \(X\)

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DV_X = X_i (\bar{X}_{\forall j \neq i} - \bar{Y})
\]

and for trial type \(Y\)

\[
DV_Y = Y_i (\bar{X} - \bar{Y}_{\forall j \neq i})
\]

where \(X_i\) and \(Y_i\) are the single-trial calcium transients for the \(i\)-th trial. Trials were classified as belonging to trial type \(X\) or \(Y\) if \(DV_X\) or \(DV_Y\) was greater than a given criterion, respectively. To determine the fraction of trials an ideal observer could correctly classify, an ROC curve was constructed by varying this criterion value across the entire range of \(DV_X\) or \(DV_Y\). At each criterion value, the probability that a trial of type \(X\) exceeded the criterion value was plotted against the probability that a trial of type \(Y\) exceeded the criterion value. The area under the ROC curve was then calculated to represent the single-neuron performance (“fraction correct”) as the fraction of trials correctly discriminat-

**Figure 3. Omission response is robust and stable.**

**A**, Example average omission responses from the first 5 and last 5 Blank trials. **B**, Left, mean peak \(\Delta R/R\) of the first 5 and last 5 omission responses from the first imaging session of each mouse \((n = 18\) omission-selective neurons from 4 mice). Right, mean peak \(\Delta R/R\) of the very first Blank trial and the mean peak omission response from all Blank trials of the first imaging session. **C**, Example single-trial calcium transients of omission-selective neurons in Blank trials, with the corresponding whisker angles below, and their average traces \((n = 24)\) compared to their corresponding response to P100 trials \((n = 54)\). Trials 2, 4 and 5 display whisking during the touch-stimulus window. **D**, mean peak \(\Delta R/R\) of the omission response in Blank trials with no-whisking and whisking. Lines represent individual neurons. Filled circles with error bars represent group averages shown as mean ± sem. Statistical significance is indicated by *** for \(P < 0.005\). No statistically significant differences were found comparing trials in **B**.
ed by an ideal observer using the DV (Green and Swets, 1966). Neurons discriminating above chance were identified using repeated permutation tests where trial type or stimulus labels were randomly shuffled. For each permutation test, a threshold corresponding to 2 standard deviations of the shuffled distribution was calculated. Neurons, whose performance values were above the mean value of this threshold across 1000 permutation tests, were considered to be discriminating above chance.

**Statistical analysis.**

All data are presented as mean ± s.e.m. unless stated otherwise. Non-parametric tests were performed for all analyses, and the Wilcoxon signed-rank paired test and the Wilcoxon-Mann-Whitney test for paired and unpaired group comparisons respectively and the Wilcoxon signed-rank test for single distributions.

**Results**

In order to determine how sensory information is represented in the neocortical microcircuit of PPC, we presented one of two textured surfaces (sandpaper of grade P100 and P1200, respectively) to the mouse whisker pad (Chen et al., 2013). We adopted a motorized linear stage that randomly presented the texture to the whisker pad contralateral to the imaging site (Figure 1A) in naïve awake mice pre-implanted with cranial windows and habituated to head restraint in the setup, but otherwise not exposed to the textures. Two auditory tones signaled the start of the trial and when the texture was in position, respectively. Additional auditory cues were generated by the translation mo-
tor as it moved in and out of the stimulus position (Figure 1A-B). To determine if PPC carries mismatch signals to omitted expected stimuli, as observed in V1 (Keller et al., 2012; Fiser et al., 2016), we further interleaved the texture trials with ‘Blank’ trials where textures were omitted and the preceding auditory cues were present (Figure 1A; Methods).

In order to localize the position of PPC in mice (Wang et al., 2012; Olcese et al., 2013), we performed IOS imaging (Grinvald et al., 1986; Petersen, 2007) to visualize the boundaries between the γ barrel column in the mouse whisker barrel cortex and the primary visual cortex V1 (see Methods and Materials) (Figure 1C). This allowed us to target our recordings to posterior parietal area A (Wang and Burkhalter, 2007; Wang et al., 2012) in the lateral parietal association cortex (LPtA) (Franklin and Paxinos, 2008), located in between the somatosensory barrel cortex and V1. In four mice, we carried out retrograde and anterograde labelling experiments with tracer injections targeted to the area between the γ and V1 boundaries and determined that this region received axonal projections from known PPC input sites, particularly LP (Figure 1G-H) and premotor cortex M2 (Figure 1D-E), as well as sent its axons to known PPC output sites, including M2, the dorsal striatum (Figure 1D-F) and LP (Figure 1G-H) (Reep and Corwin, 2009; Harvey et al., 2012). Hence, we identify this region between barrel cortex and V1 as PPC and subsequently used IOS to localize it via exclusion in all experiments.

We performed in vivo calcium imaging of layer 2/3 neurons (150 to 220 µm in depth) of the PPC through a cranial window using YC-Nano140, a genetically encoded calcium indicator that reliably reports action potential discharge (Chen et al., 2013) (Figure 2A). We imaged from four mice and 13 fields of views (FOV), and recorded a total of 358 neurons, where 126 were active during the trials (see Methods and Materials). For each FOV, mice were presented with 100 trials, which comprised of approximately 40% trials with presentation of each texture and 20% Blank trials. PPC neurons displayed a diversity of responses during trials, with some neurons responding to auditory cues, textures, or both, while others showed suppressed activity during cue and texture presentation (Figure 2A-B). We classified the trial-active neurons (n = 126) as cue-responsive (20.6%), touch-responsive (13.5%), mixed-responsive (11.1%) and suppressed (18.3%) based on the dynamics of their calcium responses to the trial structure. The remaining 36.5% of ‘unclassified’ active neurons responded neither to auditory cues nor to texture presentation (Figure 2B; see Methods and Materials). Furthermore, a subset of these ‘unclassified’ neurons (n = 12) as well as a subset of classified neurons (n = 26) showed an enhanced response in the touch-stimulus window during Blank trials, where the expected texture, as cued by the auditory sounds, was omitted (see average traces of neurons 1, 3-5, Figure 2A). These neurons were additionally termed as ‘omission-selective’ neurons and made up 30.2% of the active neurons. Interestingly, 55.3% of omission-selective neurons were suppressed or non-responsive during tex-
ture presentation trials, suggesting that they could be potentially acoustically driven in the Blank trials.

We went on to further characterize these omission responses by examining their neural dynamics during the course of the experiment. To determine if their activity was experience-dependent and gradually developed over time, we compared the mean peak $\Delta R/R$ response during the touch-stimulus window between the first 5 and the last 5 Blank trials of all omission-selective neurons recorded during the first session, in which the mouse was confronted with the setup (i.e. we only used data from the first imaging area measured). We found that the peak amplitudes of the evoked responses in omission-selective neurons were comparable for both subsets of trials (Figure 3A-B; peak $\Delta R/R$: first 5 trials $21.2 \pm 2.4\%$, last 5 trials $24.9 \pm 2.6\%$; $p = 0.09$; $n = 17$ omission-selective neurons in 4 FOVs, Wilcoxon signed paired test). Moreover, the peak calcium transient amplitude in response to the very first omission response was comparable to the mean peak omission response for all Blank trials (Figure 3B; peak $\Delta R/R$: first trial $24.6 \pm 4.5\%$, trial mean $23.5 \pm 2.3\%$; $p = 0.43$, $n = 17$ omission-selective neurons in 4 FOVs, Wilcoxon signed paired test). This suggests that the PPC is able to report the omission of a tactile stimulus reliably and stably over time. Moreover, for all 4 FOVs, the first Blank trial was presented as the second trial in the sequence of trials, further suggesting that the PPC is able to rapidly encode these sequences of salient multisensory stimuli.

We further verified if the omission response could also be an efferent copy of a motor command such as whisking as the PPC has strong reciprocal connections with the motor and premotor cortices (Kolb and Walkey, 1987; Reep et al., 1994; Wang et al., 2012). The omission response was observed in Blank trials both in the absence and presence of whisking behavior (see Methods) during the touch-stimulus window, indicating that it is not solely dependent on whisking (Figure 3C-D; peak $\Delta R/R$: no whisking $9.7 \pm 1.1\%$; $p = 3.65 \times 10^{-7}$, $n = 34$ omission-selective neurons in 12 FOVs, Wilcoxon signed rank test). A comparison of the peak of the evoked omission responses in the presence and absence of whisking during this window revealed that the omission response was enhanced by whisking (Figure 3D; peak $\Delta R/R$: no whisking $9.7 \pm 1.1\%$, whisking $16.4 \pm 1.4\%$; $p = 1.47 \times 10^{-7}$, $n = 34$ omission-selective neurons in 12 FOVs, Wilcoxon signed paired test). To verify if the omission response was acoustically driven in omission-selective neurons that were suppressed or non-responsive during texture presentation trials, we determined the onset time of the omission response of these neurons in non-whisking trials. Apart from one neuron (onset $120 \text{ ms after texture stop tone}$), the others had onset times well before the translation stage came to rest (onset $790 \pm 100 \text{ ms before texture stop tone}$, $n = 18$ neurons). Hence the omission response could represent a mismatch signal that can be further heightened by whisking motor output.

We then characterized how well tactile stimulus features or the absence of the tactile
stimulus are represented in the PPC. Texture-evoked peak $\Delta R/R$ responses during the touch-stimulus window in touch-responsive and mixed-responsive neurons were comparable to the omission response in omission-selective neurons (Figure 4A; mean peak $\Delta R/R$: P100 20.7 ± 2.7%, P1200 19.7 ± 2.3%, Blank 20.5 ± 1.3%, Wilcoxon rank test p = 0.27 for P100 vs Blank and p = 0.13 for P1200 vs Blank, n = 31 touch responsive and mixed neurons, n = 38 omission-selective neurons). We carried out an ROC analysis to identify the fraction of neurons that could significantly discriminate between the two presented textures and the fraction of neurons that could discriminate between texture-presentation trials and stimulus omission in Blank trials (compared to shuffled controls, Methods). Only 8.7% of trial-active PPC neurons (11 out of 126 neurons) were able to discriminate between the graininess of the presented textures, whereas 23.8% of them (30 out of 126) could discriminate between texture presence and omission (Example traces in Figure 4B). This subset of omission-selective neurons was able to discriminate a comparable fraction of trials compared to texture-discriminating neurons (Figure 4C; fraction of trials discriminated: P100 vs P1200 70.5 ± 1.7, Blank vs P100 74.8 ± 1.7, Blank vs P1200 76.9 ± 0.02, Wilcoxon-Mann-Whitney test p = 0.13 for P100 vs P1200 and Blank vs P100 and p = 0.26 for P100 vs P1200 and Blank vs P1200, n = 11 texture-discriminating neurons, n = 30 (Blank vs P100) and n = 19 (Blank vs p1200) omission-selective neurons. Note that the latter 19 neurons are part of the 30 omission-selective neurons). These results suggest that while a subset of neurons within the PPC can discriminate stimulus features (texture graininess), a larger fraction of PPC neurons can reliably represent the omission of the cued stimulus.

**Discussion**

We found that layer 2/3 neuronal populations in the mouse PPC display a diversity of responses in a given context during repeated trials, with neurons responding to auditory cues and/or tactile stimuli, as well as other neurons being suppressed by these stimuli. Few PPC neurons (8.7%) were able to discriminate the two textures that were presented. This low discrimination power could be expected as the experimental sessions were the first time that the naïve mice were exposed to these textures, without the explicit association of a relevant context to the stimulus features or any reinforcement training. Indeed in the somatosensory barrel cortex the proportion of neurons that can discriminate the same texture stimuli as applied here in a reward-based go/no-go task is higher in trained compared to naive animals (Chen et al., 2013; Chen et al., 2015). More interestingly, we found that about 30% of trial-active neurons responded to omission of a cue-predicted stimuli. This was observed in the very first omission trial, suggesting that the PPC is potentially capable of single-trial (rapid) stimulus binding and association (Winters and Reid, 2010). Our stimulation paradigm incorporated an auditory sound
that signaled the arrival of a tactile stimulus, hence a salient event. Prior stimulus binding and association of salient events acquired during home cage experience based on natural statistics could very well account for the presence of the omission response in the very first Blank trial. An alternative interpretation is that omission-selective neurons are acoustically driven and the presence of texture-touches merely suppresses their response. Indeed, more than half of these neurons were non-responsive to or suppressed by texture-touches. However, these responses had onset times well before (with the exception of one neuron) the tone signaling stage stop, suggesting that something other than sound is driving the omission response. The remaining cue and touch responsive omission-selective neurons could be potentially sound offset neurons and not purely sound responsive (Scholl et al., 2010). Nonetheless, such neurons could also convey the temporal dynamics of stimulus sequences that might serve towards stimulus binding and association.

Omission-selective neurons also displayed responses that were comparable in amplitude to texture-evoked responses in touch-responsive and mixed neurons. Hence these neurons are well equipped to play the role of reporters of mismatch signals, which could reliably convey information about unexpected changes in the sequence of sensory stimuli to the prefrontal cortex, FEF and pulvinar, brain nuclei implicated in the allocation of attentional resources (Womelsdorf and Everling, 2015). Indeed stimulus-omission responses have also been described in the auditory (Raij et al., 1997; Yabe et al., 1997) and visual systems (Bullock et al., 1990; Bullock et al., 1994; Schwartz et al., 2007), in the context of mismatch negativity (MMN) and oddball responses. However, the stimulus presentation paradigms employed in MMN are different compared to those used in our study. More recently, mismatch responses have been reported in mouse V1 (Keller et al., 2012; Fiser et al., 2016), when visual flow is not correlated with self-initiated locomotion. These mismatch signals were also present in the LP thalamic inputs to layer 1 of V1 (Roth et al., 2015), and not in the axons of the anterior cingulate cortex projecting to V1 (Fiser et al., 2016). LP is the main source of thalamic input to the PPC, which does not receive any direct input from primary sensory thalamus (Krieg, 1946; Reep et al., 1994). Hence LP could be a potential source for the omission responses that we have observed in layer 2/3 of the PPC. This could be in turn modulated by top-down feedback, potentially from the FEF, providing the prediction signal in the proposed framework of predictive coding (Kanai et al., 2015).
References


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