Electronically induced contrast enhancement in whisker S1 cortical response fields

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***Abstract*— The ability of an organism to specifically attend to relevant sensory information during learning and subsequent performance of a task is highly dependent on the release of the neurotransmitter Acetylcholine (ACh). Electrophysiological studies have shown that pairing endogenous ACh with specific visual or auditory stimuli induces long lasting enhancements of subsequent cortical responses to the previously paired stimulus. In this study we present data suggesting that similar effects can be elicited in the rat whisker sensory system. Specifically, we show that pairing whisker deflection with electrical stimulation of the magnocellular basal nucleus (BN: a natural source of cortical ACh) causes an increase in the center-surround contrast of the treated whisker's cortical response field (CRF). Meanwhile, deflections of whiskers distant from the treated whisker show overall increased response magnitudes, but non-significant changes in contrast between principle vs. surround barrel responses. Control trials, in which BN stimulation was not paired with whisker deflection, showed similar lack of contrast enhancement. These results indicate that BN stimulation, paired with incoming whisker information, selectively increases the paired whisker's CRF center-surround contrast, while unpaired BN stimulation causes a more general increases in S1 responsiveness, without contrast modulation. Enhanced control over whisker sensory pathway attentional mechanisms has the potential to facilitate a more effective transfer of desired information to the animal's neural processing circuitry, thereby allowing experimental evaluation of more complex behavior and cognition than was previously possible.**

Keywords—whisker; acetylcholine; basal nucleus; cortical response field;

I. INTRODUCTION

Attention is highly correlated with learning ability [1,2], and its impairment has been argued to be responsible for cognitive disorders such as dementia, schizophrenia, and attention deficit disorder[3–5]. Studies of attention have focused primarily on the neurotransmitter Acetylcholine (ACh) which has been shown to increase in concentration, within several brain regions, in correlation with increased attention [6–11]. A primary source of ACh in the brain is the basal nucleus (BN) whose projections terminate diffusely throughout the cerebral cortex, primarily in layers V and VI [12,13]. Previous studies of the visual and auditory system have electrically stimulated the BN to induce release of endogenous ACh in the cortex. The pairing of BN stimulation with a specific visual or auditory stimulus has thus far been found to cause long lasting enhancements of subsequent neuronal responses to the previously paired sensory stimuli [14,15]. However, this pairing of endogenous ACh with sensory stimulation has not yet been thoroughly studied in the rodent whisker system, with most previous studies instead using either exogenous ACh or endogenous ACh unpaired with whisker stimulation [16–18]. The whisker sensory system is important because it is the primary sensory modality used by rats in navigating their environment [19,20]. The whisker S1 cortex is also referred to as the 'barrel' cortex due to the Cytochrome oxidase staining pattern in layer IV of distinct cylindrical regions, each corresponding to a single whisker on the rats mystacial pad. While neurons within a given barrel respond with shortest latency and highest spikes per second to a single 'principle' whisker, they also respond with greater latency and lower spikes/sec to the deflection of surround whiskers [21]. This characteristic allows experimenters to easily monitor varying degrees of 'center-surround' contrast between the responses of principle and surround barrels to tactile whisker stimulation.

II. METHODS

A. Surgery

Electrodes were implanted in a single hemisphere in both the basal nucleus and in layer II/III of the primary somatosensory cortex (S1) whisker region of eight rats (4 experimental and 4 control). The localization of electrodes (2x4 array of stainless steel wires, 0.025 mm diameter) within specific barrels of the S1 cortex was determined by analysis of peri-stimulus time histogram (PSTH) peak latencies in response to individual whisker deflections. Electrodes recording response latencies of less than or equal to 8ms were classified as being within the deflected whisker's principle barrel, while latencies greater than 8ms were classified as residing within principle-surround barrels (Fig. 1A) [21]. Proper depth positioning of BN electrodes (two stainless steel wires, 0.1 mm diameter) was achieved by applying bursts of high frequency electrical stimulation (30 biphasic pulses, 100 Hz, 400 uA) as the electrode's depth approached the stereotaxic z-coordinate of the Basal Nucleus [22]. Electrode lowering was stopped when stimulations elicited a >1 sec increase of S1 cortical activity [23] (Fig.

1B). Electrodes were fixed in place via titanium skull screws and dental cement. Rats were allowed to recover for a minimum of seven days post-surgery before any experimental testing was initiated.

B. Treatments

Rats were anaesthetized with pentobarbital (20 mg/kg) and placed in stereotax earbars. Anesthesia was maintained by 1-3% isoflurane-air mixture administered through a nose cone. The rat's chronically implanted S1 electrodes were connected to a Plexon MAP system to allow electrophysiological recordings. Depth of anesthesia was noted every 30 min by recording reflex responses to toe web clamping. Anesthetic depth was scored according to criteria previously established by Zandieh et al. and Jang et al.[24,25], and a score < 2 was maintained in all experiments. In the pre-treatment period (Fig. 1C, Left), two whiskers, separated by at least 2 whisker columns, contralateral to the implanted hemisphere were deflected along the horizontal plane by 250 μm at 0.15 Hz via an in-house constructed, voice coil actuator, triggered by a software controlled, hardware timed digital output card (National Instruments). The deflection rate of 0.15Hz used in the pre- and post-treatment periods was chosen to avoid potential effects of long duration stimulus trains [26]. Whiskers were chosen such that the implanted electrodes recorded both <8 ms and >8 ms latency responses to their

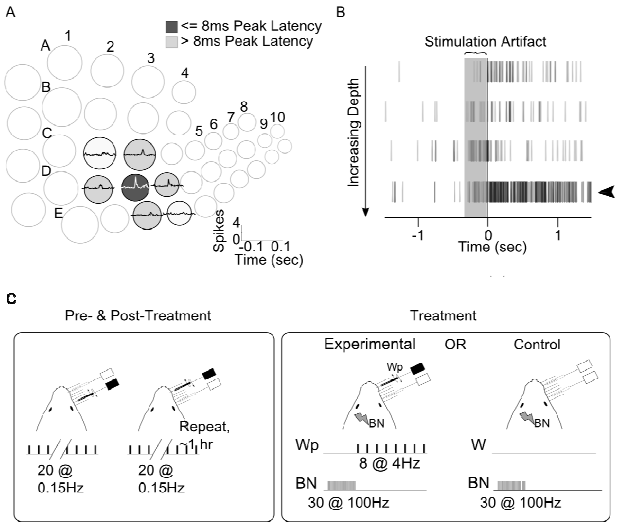

Fig. 1. A) A topographic illustration of the whisker S1 cortex showing recorded PSTHs from individual recorded units in response to a single whisker's deflection (whisker D3). B) A raster plot of spike responses recorded from a single S1 electrode after stimulation through electrodes being lowered towards the BN. Time = 0 represents the end of stimulus train. Decent of BN electrodes was stopped when stimulus trains elicited a >1 sec response at whisker S1 cortex electrodes (arrowhead). C) Timeline showing pre-treatment, treatment, and post-treatment periods. Pre- and post-treatment periods lasted on average one hour and involved horizontal deflections of individual whiskers by 250 μm at 0.15 Hz. During the treatment period, experimental treatments involved BN stimulation followed immediately by 8 whisker deflections (Wp) at 4 Hz, while control treatments involved only BN stimulation.

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deflections. In the pre-treatment period deflections were repeated at the specified rate until the magnitudes of evoked PSTH responses were observed to be stable, with SEM < 5 calculated from the previous 45 min of recording. At this point treatment was initiated (Fig. 1C, Right). Experimental treatments (N = 4) were constituted by 20 BN stimulations delivered across 10 minutes, each followed by 8 whisker deflections of a single whisker across a 2 second period. The whisker chosen to be paired with BN stimulation was randomly chosen from one of the two whiskers deflected during the pre-treatment period. In control treatments (N = 4) BN stimulations were presented without paired whisker deflections. The post-treatment period was conducted in the same way as the pre-treatment period for approximately one hour.

C. Analysis

Multi-unit activity was recorded using Plexon's Sort Client. Responses to whisker deflections were averaged in groups of 20 to account for inter-deflection variations in response magnitudes [27]. Measurements of PSTH peak heights were taken as baseline-to-peak magnitudes (spikes/bin, 5 ms bins) within a 100 ms post-stimulus window, with baseline being calculated as the average spikes/bin of spontaneous activity in the 50ms directly preceding the stimulus. Mann Whitney analysis was used to determine the presence of any post-treatment periods (of duration $\geqslant$30 min) which significantly deviated from the latest 45 minutes of pre-treatment values. Significant post-treatment changes were found in both principle and surround responses in all sessions. The magnitudes of significant changes were then calculated by subtracting the mean pre-treatment PSTH peak height from the mean of the significantly deviating post-treatment PSTH peak heights (Fig 3B).

III. RESULTS

Fig. 2A & B depict examples of spike PSTH waveforms and their magnitudes over the duration of individual trials. Fig. 2C summarizes the changes in PSTH peak heights induced by both experimental and control treatments (N=4). Experimental treatments caused enhancement of the paired whisker's principal barrel (Wp) vs. surround barrel (Wp-s) responses ($p < 0.05$), while whiskers distant from the paired whisker showed statistically equivalent increases in both principal (Wd) and surround barrel (Wd-s) responses ($p = 0.073$). Control treatments showed statistically equivalent increases in both principal (W) and surround barrel (W-s) responses to whisker deflection ($p=0.37$). Changes induced by both experimental and control treatments are depicted in conceptual form in Fig. 2C, Bottom.

IV. DISCUSSION

The pairing of acetylcholine with whisker deflection potentiated the paired whisker's principal barrel (Wp) response, while potentiating surround barrels (Wp-s) to a significantly lesser degree, thereby increasing the contrast between the Wp and Wp-s barrel responses (Fig. 2C).

Additionally, whiskers distant from the paired whisker showed statistically indistinguishable increases in responsiveness of both principal (Wd) and surround barrels (Wd-s). Finally, control treatments also resulted in statistically indistinguishable potentiation of both principal (W) and surround barrels (W-s). These results are similar to those from previous studies of ACh's effect on the whisker system. In 2001 Ego-Stengel et al. showed that whisker deflection paired with the iontophoresis of ACh caused potentiation of principal barrels, but not surround barrel responses to deflections of Ach-paired whiskers [16]. Meanwhile, in a study by Howard & Simons and Kuo et al., BN stimulation delivered alone eliciting increased responsiveness of whisker surround barrels [17,18]. Combined, the present and previously published results seem to indicate that the degree of contrast between principal and surround barrels may be adjusted as a function of ACh concentration and the presence of paired whisker deflection. In our data, Wp and Wd responses appear to be similarly

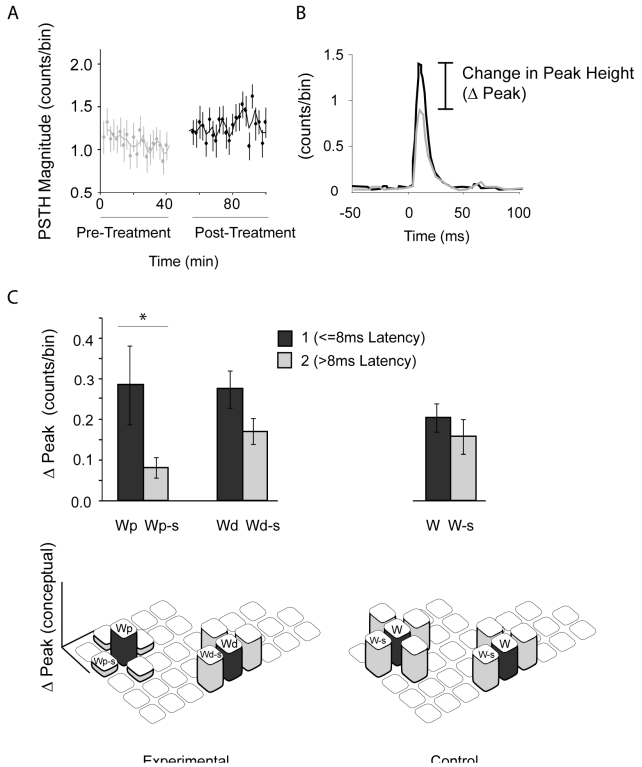

Fig. 2. A) An example of a recorded unit's PSTH peak heights over time, both pre-treatment (grey) and post-treatment (black). B) Overlaid examples of a recorded unit's mean pre-treatment (grey) and post-treatment (black) PSTHs, illustrating the difference in peak heights. C) Top Row: Changes in PSTH peak heights pre- vs. post-treatment of principle and surround barrels in response to deflections of the paired whisker (Wp), distant whisker (Wd), or unpaired control whisker (W). Asterix indicates significant difference between changes in Wp and Wp-s responses. No statistically significant difference was found between increases in Wd vs. Wd-s or W vs. W-s responses. Bottom Row: A cartoon of the topographically arranged whisker S1 cortex showing a conceptual model of changes in barrel PSTH peak heights in response to whisker deflections (Wp, Wd, or W).

enhanced, with the difference lying in the enhancement of their respective surround responses. The mechanism for this effect could be a potentiation of lateral inhibitory connections in the S1 cortex that have been shown to modulate specificity of stimulus responses [28,29]. While all anesthetics affect brain function, isoflurane's main effect is to strengthen GABAergic inhibition in thalamocortical connections [30]. Therefore its use in this study is most likely a cause of across the board dampening of response magnitudes, without differentiating the overall pattern of effects from an awake condition.

The meaning of increasing neural response contrast has been hypothesized to be a selective directing of attention to the sensors eliciting the largest neural responses, with little or no attention devoted to those sensory inputs with responses of lower magnitude. In the case of this experiment, this could suggests an ability of the rat to direct attention to a specific whisker (the Wp). However, given that rats cannot control individual whisker movements [31,32], it is unclear why they would possess the ability to direct attention to a single whisker. One reason could be that whiskers of particular lengths are better suited to detect specific ranges of surface roughness. Indeed, this has been suggested by studies in which the resonant properties of whiskers of varying lengths have been shown to differ [33]. However, while the whiskers of a rat's mystacial pad decrease in length along rows from caudal to rostral, each column of the rats mystacial array has whiskers of approximately equal length [34]. Therefore, assuming an advantage in directing attention to whiskers of particular length, this would predict that attention resolution in the whisker system would be limited to individual columns and not to individual whiskers. This possibility is somewhat supported by studies indicating that whiskers in separate columns can be controlled independently from each other; allowing those in one column to whisk while those in a neighboring columns remain still or move in an opposite direction [33,35,36]. This column-specific level of motor control would be capable of working in concert with column-specific attention mechanisms. Because multiple BN-paired whiskers within a single whisker column were not tested in the present study, this question of attentional resolution within the rat whisker system remains to be further tested.

In conclusion, we have observed specificity in the effects of Basal Nucleus stimulation on the contrast of principal vs. surround barrel responses to tactile stimuli in the whisker S1 cortex. Whether this specificity has a resolution of individual barrels, or barrel-columns remains to be tested. Further study of attention's effects on the whisker S1 cortex is important because knowledge gained will facilitate use of the whisker sensory system as a highly effective conduit for the delivery of information to the rat's brain, allowing more complex cognitive experiments to be conducted in the future.

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