Reproducible whisker stimulation for fMRI and neurophysiological studies

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INTRODUCTION

The use of animal models for simultaneous fMRI and neurophysiology [1] has become important for neuroscience studies. However concurrent fMRI and neurophysiology [2] is more the norm for high magnetic field rodent studies because the placement of the neurophysiology probe destroys the MRI signal from the voxel or region of interest. Concurrent experiments inside and outside the magnet requires reproduction of exact stimulus conditions. This can be easily achieved when the stimulus is of electrical origin, e.g., forepaw stimulation using electrical currents [3]. However for other non-electrical stimuli used in animal experiments (e.g., olfactory, visual, auditory, whisker) it is extremely important to generate stimuli with identical sensory effects both inside and outside the magnet. The rodent whisker-to-barrel cortex pathway has a number of properties that facilitate the experimental use of this model for understanding sensory perception [3,4]. Deflections can be applied to the whisker array in a precise fashion and the structure of receptive fields can be mapped in a highly quantitative manner [5]. In the rat, many studies have utilized different stimulation devices to characterize the whisker deflection to barrel response for fMRI and elctrophysiological studies [6-9]. The goal of this work was to design a simple stimulation paradigm for whisker deflections that provides consistent and reproducible neuronal activations in the rat cerebral cortex for fMRI and neurophysiological studies. Preliminary results from fMRI and neurophysiological studies show that stimulation of all whiskers with this air puff device increased activity in the contralateral barrel field with a maximum response at 8 Hz, in good agreement with prior findings [6-9].

MATERIALS and METHODS

Animal preparation: Sprague-Dawley rats were tracheotomized and artificially ventilated (70% N_2O, 30% O_2). During the animal preparation halothane (1 to 2%) used for induction. Intraperitoneal lines were inserted for administration of α-chloralose (46±4 mg/kg/hr) and D-tubocurarine chloride (1 mg/kg/hr). An arterial line was used for monitoring physiology (blood pH, pO_2, pCO_2) throughout the experiment. ***Whiskers stimulation:*** Contralateral whiskers were trimmed to a length of ~14 mm. Air puffs were used to stimulate the whiskers. Airpuffs were generated from pulses of compressed air, which could be delivered in a computer-controlled way by inbuilt solenoid unit (Solenoid valves, Cole-Parmer Instrument). Air puffs were applied through stiff micropipette tip with a 2 mm opening positioned 15-20mm lateral from whiskers. Under these conditions, the air puff stimuli deflected all the whiskers by 2 mm (rostral-caudal) which were glued with a tiny adhesive tape (Fig. 1A). All stimulus presentation was controlled through a µ-1401 (CED, Cambridge, UK) running custom-written script. Whiskers stimuli were presented at 4, 8 and 12 Hz for duration of 30 s and were interleaved with an inter-stimulus interval (ISI) of 300s (i.e., time form end of one stimulus to the start of next). This very long ISI was used to ensure that there were no interaction effects between adjacent stimuli. ***fMRI (n=8):*** All fMRI data were obtained on a modified 11.7T Bruker horizontal-bore spectrometer (Billerica, MA) using a ^{1}H surface coil (Ø = 2 cm). The images were acquired with gradient echo EPI sequence (TR/TE = 2500/15). ***Laser Doppler flow (LDF) and tissue pO_2 (n=10):*** The rat was placed in a stereotaxic holder on a vibration-free table inside a Faraday cage. Tiny burr holes above the contralateral and ipsilateral whisker barrel regions [5.5 mm lateral and 2.5mm posterior to bregma] were made for the LDF and pO_2 probes (Oxford Optronix) up to layer 4 depth with stereotaxic manipulators (Kopf). The signal was then digitized with a µ-1401 interface using SPIKE-2 software [2].

RESULTS and DISUSSION

Whisker stimulation (4 to 12 Hz) produced BOLD signal increases in the contralateral barrel cortex (Fig. 1B) with a maximum response at 8-12 Hz range (Fig. 1C). The fMRI results were in agreement with neurophysiology data from pO_2 (Fig. 1D) and LDF (Fig. 1E) probes reflecting tissue oxygenation and perfusion changes, which together give rise to the BOLD signal change. While this device provides easy control of frequency of stimulation, we are currently investigating the amplitude variations. In summary, unilateral whiskers stimulation with air puffs resulted in significant increases in neurophysiological signals in the contralateral barrel field in agreement with prior findings [6-9].

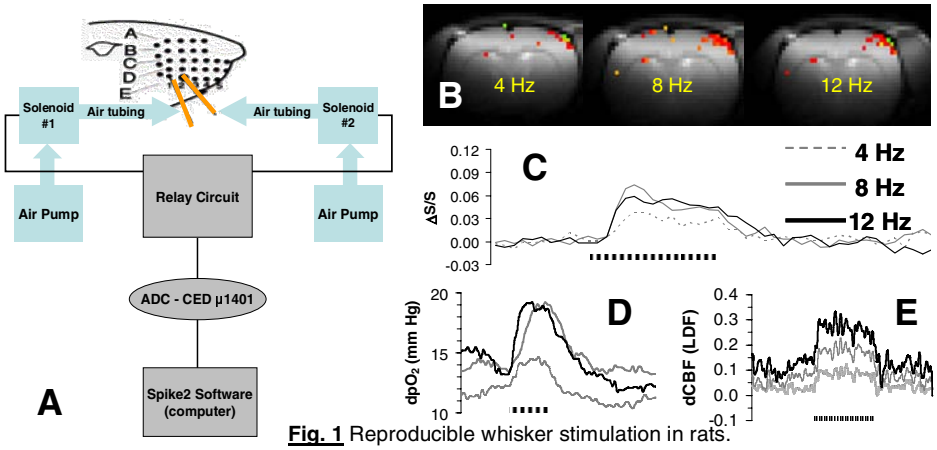

Fig. 1 Reproducible whisker stimulation in rats.

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