MRI-compatible chronic neural monopolar carbon fiber electrodes for a longitudinal neuromodulation intervention in a Wistar rat model of chronic ethanol consumption.

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Objective

There is a need for MRI-compatible electrodes that allow for MRI acquisition with minimal distortion of the magnetic field to study focal repeated stimulation in rats. The most commonly used electrodes for neurostimulation in rats are made of metal which creates susceptibility artifacts (Dunn et al., 2009) and leads to the loss of signal around the region where the electrode is placed, as well as distortion and lower signal-to-noise ratio (SNR) (Redpath 1998). One option is to use carbon fiber electrodes that are commonly used in neuroscience for neural recording (Joshi-Imre et al. 2019; Chuapoco et al. 2019) and have been shown to work well in stimulating brain regions (Gallino et al. 2019). These electrodes have proven to be a better choice for the improvement of SNR with little to no MRI susceptibility artifacts, due to their physical properties like high resistance, lightweight, and low density, (Zhao et al. 2019). However, the viability of its use in the treatment of substance use disorders and in longitudinal MRI studies in rat models has not been tested. This work aims to validate a low-cost approach for assembling chronically implantable monopolar carbon electrodes with longitudinal MRI follow-ups and their use for repetitive focal stimulation alcohol use disorder (AUD).

Methods

We included 12 (male n = 6, female n = 6) Wistar rats (*Rattus norvegicus albinus*) since postnatal day 21 (P21) from the vivarium of the Institute of Neurobiology in Queretaro, Mexico. We used the *Intermittent access two-bottle choice (IA2BC)* (Carnicella, Ron, and Barak 2014; Simms et al. 2008; Wise 1973) as the *ethanol self-administration model*. Electrode implantation (Santana-Chávez, et al., 2020) was performed at P100 after the end of IA2BC at the beginning of a 10-day ethanol withdrawal period. The construction of the carbon monopolar electrodes was based on the design of Gallino et al., (2019). At P110 the IA2BC was restored, in accordance with that corresponding to the relapse. Between P120 and P132, we started the stimulation protocol for 10 sessions of 10 min for 10 consecutive days with 300 pulses (duration = 0.2 ms, intensity = 400 μA) at 20 Hz in 10 pulses per train of 2 s, and an inter-train interval of 20 s (Figure 4) (Levy et al. 2007; Gersner et al. 2010). After the stimulation protocol, 5 more sessions of IA2BC were performed. MRI sequences GE-EPI and 3D FLASH were performed at two-time points, after the implantation and after IA2BC the relapse sessions.

Magnetic Resonance Imaging

Magnetic Resonance Imaging (MRI) scanning was performed at the Unidad de Resonancia para Roedores y Otros Animales (URRA), Laboratorio Nacional de Imagenología por Resonancia Magnética (LANIREM) at the Instituto de Neurobiología, UNAM campus Juriquilla, located in Querétaro, Qro, Mexico. Prior to the acquisition, rats were injected subcutaneously with dexmedetomidine 0.012 mg/kg (Sirmpilatze, Baudewig, and Boretius 2019). During imaging, rats were anesthetized with 5% vaporized isoflurane at induction and 0.5% in a 50/50 mixture of oxygen and vaporized anesthesia. Image acquisition was

performed using a 7T Bruker Pharmascan (Bruker Pharmasan 70/16, US) with a 2x2 surface coil and acquired using Paravision 6.0.1. A 3D FLASH T2w sequence with 2 repetitions TR = 30.76ms TE = 5ms, flip angle = 10°, and FOV = 25.6 x 19.098 x 25.6 mm and an isometric voxel of 160 microns, and GE EPI sequences were 1) TR = 1000 ms, TE = 20 ms, flip angle = 60, slice thickness = 1 mm, FOV = 30 x 30, number of slices = 24, volumes = 600 and axial as primary slice orientation, 2) TR = 1800 ms, TE = 20 ms, flip angle = 60, slice thickness = 0.75 mm FOV = 30 x 30, number of slices = 32, volumes = 334 and axial as primary slice orientation. Both sequences were performed at P110 to verify electrode placement and at P144 at the end of the stimulation protocol. All images were converted from Bruker to nifti format using the brkraw v0.3.3 tool (Lee, Ban, and Shih 2020). Anatomical images were preprocessed using a proprietary pipeline based on MINC-toolkit-v2 and ANTs that performed the following steps: intensity normalization, central imaging, denoising, and registration in LSQ6 alignment. (https://github.com/psilantrolab/Documentation/wiki/Preprocessing-Rat-Structural-in-vivo).

Results

Twelve rats conserved both stimulation electrodes and remained available for the stimulation protocol for the longitudinal follow-up. All the electrodes held their original resistance values (2kΩ-8kΩ). Around the electrode area, there were no displacement artifacts, geometric distortion, or signal loss (Hargreaves et al., 2011). In the longitudinal follow-up of a rat, there were no structural changes measured with the 3D FLASH sequence. After the stimulation, there were no group differences in alcohol consumption. However, the individual data showed high variability of consumption. Our results showed that 66% of the rats in the active group had a reduction in consumption, while 50% of the rats in the sham group also showed a reduction in consumption.

Conclusions

Carbon fiber monopolar electrodes are a good option for repetitive focal electrical stimulation with longitudinal MRI follow-up in rodent models of AUD.

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