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 We use this protocol and it's working

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SPARC RNEL Bladder January 2019 protocol

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ABSTRACT

Protocol for dataset "RNEL Bladder January 2019"

Lower urinary track nerve responses to high-density epidural spinal cord stimulation (RNEL January 2019)

ATTACHMENTS

[SPARC_RNEL_Bladder_2019_Protocol.pdf](#)

MATERIALS

Nerve cuffs:

- Micro-Leads self-closing nerve cuffs (0.5, 0.75 and 1.0 mm inner diameter).
- Ardiem nerve cuff sized for sciatic nerve.

Stimulation array:

- custom 16-channel and 24-channel spinal cord arrays (Micro-Leads)

Drugs:

- Ketamine (induction),
- Isoflurane

Neural Recording and stimulation hardware:

- Ripple Grapevine neural interface processors,
- 32 channel high-current stimulation headstage,
- 32-channel recording headstage

Surgical Preparation

- 1 Anesthetize animal and maintain physiological parameters throughout procedure
- 2 Place animal in the supine position, place tracheostomy tube and place invasive blood pressure measurement catheter.
- 3 Make a midline abdominal incision to expose bladder.
- 4 Insert dual lumen catheter into the bladder dome and secure with purse string suture and other suture as necessary.
- 5 Identify and verify the pelvic nerve. Place nerve cuff (MicroLeads) on the left pelvic nerve. This nerve typically requires a 500 um nerve cuff.
- 6 Close abdominal incision in layers with suture or staples leaving catheter and electrode leads exposed.
- 7 Place animal in a prone position and make incision on the left side just lateral to the tail exposing the ischioanal fossa.
- 8 Identify and verify the left pudendal nerve, as well as the sensory branch, deep perineal branch, and caudal rectal branch. Place appropriately sized nerve cuff electrodes on all four nerves.
- 9 Place nerve cuff (Ardiem) on the sciatic nerve ipsilateral to the rest of the nerve cuffs.

- 10 Close incision with suture or staples.
- 11 Perform laminectomy removing the L6, L7, and S1 lamina. Mark dorsal process location with suture prior to laminectomy. Place epidural spinal cord array (MicroLeads) with 16 or 24 channels on the spinal cord for spinal cord stimulation.
- 12 Three epidural stimulation locations were tested. For the most rostral location, the array was placed such that the most rostral row of electrodes is aligned to the center L6 process. At the two more caudal locations, the array is placed such that the most caudal row of electrodes is aligned to the L7 and S1 spinous processes, respectively.
- 13 Connect nerve cuff electrode contacts to the recording headstage. Connect the epidural stimulation array contacts to the stimulation headstage.

Electrical stimulation

- 14 Stimulation is delivered using a Grapevine Neural Interface Processor through a Nano 2+Stim high-current headstage. Stimulation is controlled the MATLAB (Mathworks) XippMex API. All pulses are asymmetric charge-balanced with a 200 us cathodal phase, followed by a 66 us interphase period, followed by a 400 us anodal phase at half the current amplitude.
- 15 Deliver 50 stimulation pulses to all electrodes, one at a time, in a random order at a low frequency (20 Hz) and a uniform high current amplitude (typically 400-800 uA). The exact stimulation amplitude is empirically determined, using test pulse trains, and should be an amplitude that results in activation of the sciatic nerve and many or all of the other nerve cuffs. Since the ultimate goal is to identify stimulation thresholds at each nerve cuff, this high-amplitude is used to identify whether responses can be observed in the instrumented nerves. By using a low frequency in this single-amplitude survey, we observed the longest-latency responses, then added 5 ms to produce the minimum response latency of interest at a given electrode.
- 16 For all nerve cuff recordings, remove stimulus artifact, high-pass filter the data and perform stimulus-triggered averaging to find the mean nerve responses to stimulus pulses.

- 17 Determine the presence of a compound action potential (CAP) on each nerve. Taking the stimulation-triggered averages, compare the response to baseline nerve cuff recordings without stimulation.
- 18 Determine the threshold stimulus current amplitude for each electrode necessary to recruit activity on each nerve. Stimulation frequencies and amplitudes can be determined using a binary search procedure. In the binary search, include electrodes where stimulation produced CAPs on the pelvic and pudendal nerves. Use a stimulation frequency determined by the minimum response latency found during the single amplitude survey. Increasing the stimulus frequency minimized the amount of time required to perform the experiments.
- 19 For each electrode, use a binary search algorithm to identify the minimum stimulus threshold that results in a detectable response in each instrumented nerve. 300 individual stimulus pulses are typically sufficient for averaging.
- 20 Stimulation was applied to monopolar electrode configurations, as well as multipolar configurations, to localize current below the electrodes stimulated. Multipolar configurations included bipolar and tripolar sets of electrodes, with bipolar configurations of two adjacent electrodes both horizontally and vertically in the electrode array, and tripolar configurations of adjacent vertical electrodes in the array.