Physiological Properties of Lamina I Spinoparabrachial Neurons Innervating the Hind Paw in the Mouse

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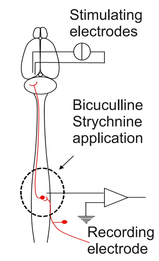
Aim

- Lamina I spinoparabrachial (SPB) neurons form the major ascending pain pathway in rodents, and contribute to both acute and chronic pain.
- Current research on lamina I SPB neurons is exclusively based on recording from spinal cord slice, with all limitations attached. Lamina I SPB neurons have never been studied in mice in vivo despite the growing and extensive use of this species in the field of pain research.
- The aim of the present study was to characterize lamina I SBP neurons in anaesthetized mice and to assess the ability of spinal application of GABAergic and glycinergic antagonists to allow input from light tactile afferences to reach nociceptive specific lamina I SPB neurons.

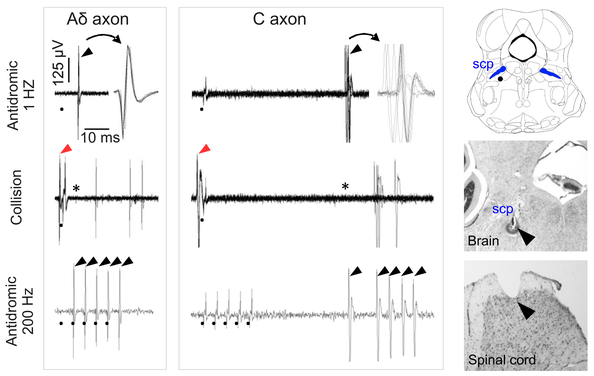
Set up and method



- Swiss mice under isoflurane anaesthesia, d-tubocurarine paralysis, artificial ventilation, with continuous measure of end tidal CO2, blood pressure and body temperature.
- Extracellular recording of single unit in the spinal cord using a tungsten microelectrode.
- Search of lamina I neurons based exclusively on antidromic stimulations using bipolar concentric electrodes implanted in the PB area.
- Responses to brush (Br.), Von Frey (VF) (force in mN), pinch with haemostat clamp (Pi.), and to water jet (WJ, 10 ml) at different temperatures measured as number of action potentials (AP) over 5 s and peak firing frequency (Hz) over 0.1 s.
- Peripheral electrical stimulations with two needles inserted in the receptive field (RF) or with bipolar electrodes hooked on the sciatic nerve (SN).
- Measure of brush response before and after the application of 100 µM bicuculline and 10 µM strychnine on the spinal cord.

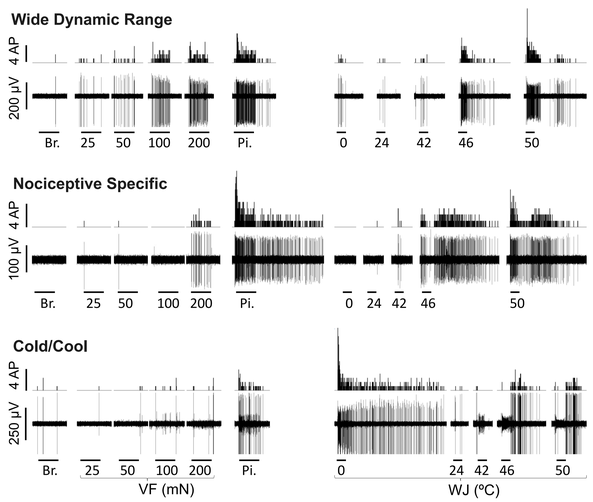
Results

1 Identification of lamina I SPB neurons



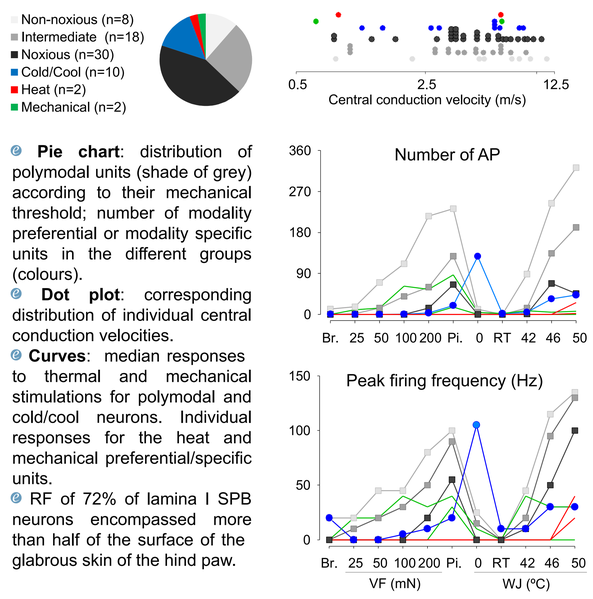
- Recordings from 2 lamina I SPB neurons with lightly myelinated (Aδ) and unmyelinated (C) projection axons. "Antidromic 1 Hz" and "Collision" correspond to the overlay of 10 successive responses.
- ▪ , antidromic stimulation artefact; ➤, orthodromic AP triggering the antidromic stimulation; ✱, missing AP; ➤, antidromic AP.
- Example of location of recording and stimulation sites (electrolytic lesion). Scp, superior cerebellar peduncle (brachium conjonctivum).

2 Example of responses to mechanical and thermal stimuli



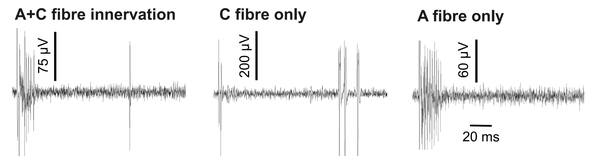
- Responses of 3 lamina I SPB neurons to stimulations of the RF on the hind paw during the initial characterization. Peristimulus histogram: number of AP/0.1 s bin. Time scale bar: mechanical stimuli, 6 s; thermal stimuli, 3 s.

3 Lamina I SPB neurons form a heterogeneous population with slow central conduction velocity



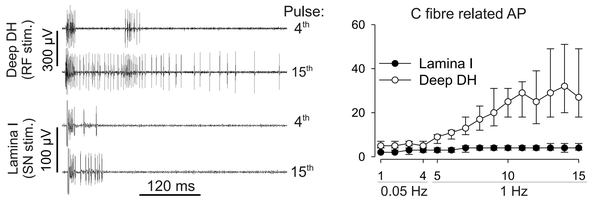
- **Pie chart**: distribution of polymodal units (shade of grey) according to their mechanical threshold; number of modality preferential or modality specific units in the different groups (colours).
- **Dot plot**: corresponding distribution of individual central conduction velocities.
- **Curves**: median responses to thermal and mechanical stimulations for polymodal and cold/cool neurons. Individual responses for the heat and mechanical preferential/specific units.
- RF of 72% of lamina I SPB neurons encompassed more than half of the surface of the glabrous skin of the hind paw.

4 Selective innervation of lamina I SPB neurons by peripheral A or C fibre



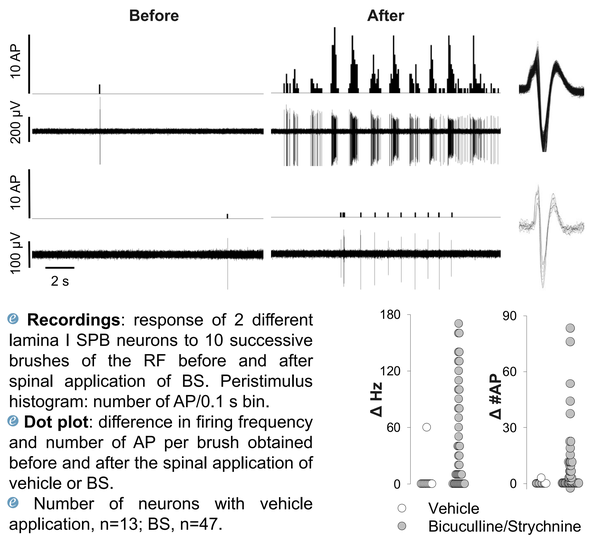
- Response to a single electrical stimulation (0.2 ms, 10 mA) of the RF in 3 different lamina I SPB neurons.
- Number of units with corresponding innervation: A+C, 25/35; A only, 6/35; C only, 4/35.

5 Reduced wind up of lamina I SPB neurons



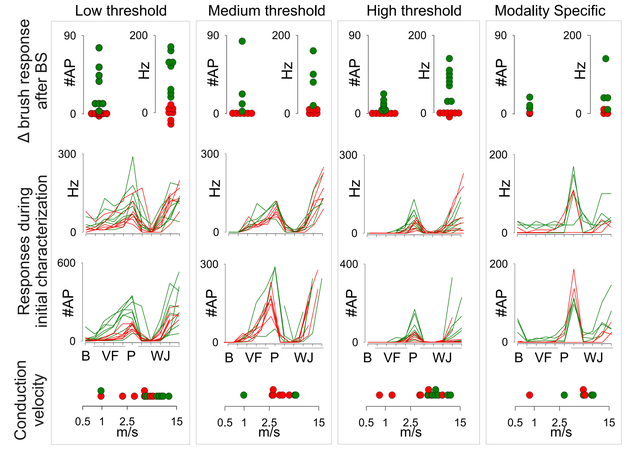
- Wind up was obtained from lamina I SPB and polymodal deep dorsal horn (DH) neurons (n=23 each).
- Example of recordings and median (25th-75th percentile) number of C fibre related AP during the successive electrical stimulations.

6 Variable enhancement of brush responses after spinal application of bicuculline and strychnine (BS)



- **Recordings**: response of 2 different lamina I SPB neurons to 10 successive brushes of the RF before and after spinal application of BS. Peristimulus histogram: number of AP/0.1 s bin.
- **Dot plot**: difference in firing frequency and number of AP per brush obtained before and after the spinal application of vehicle or BS.
- Number of neurons with vehicle application, n=13; BS, n=47.

7 Greater disinhibition of lamina I SPB neurons with initially low mechanical threshold



- All neurons tested with spinal bicuculline and strychnine were pooled according to their mechanical threshold (at least 1 AP in response to the stimulation) or as modality specific units. Low, threshold corresponding to VF 25 mN and brush; Medium, VF 50 mN; High, VF 100, 200 mN and pinch.
- Red: neurons with responses per brush enhanced by <2 AP. Green, neurons with responses per brush enhanced by ≥2 AP.
- Neurons with initially low mechanical threshold displayed the largest enhancement of responses to brush after spinal disinhibition.
- B, brush; P, pinch.

Conclusion

- Mice lamina I SPB neurons form a nociceptive and thermoreceptive pathway with slow conduction velocity more similar to what has been described in cat than rat:
  - polymodal neurons (n=56) responded preferentially to noxious mechanical and thermal (heat) stimuli (14 % responded to light touch).
  - modality specific neurons responded preferentially to cold (n=10) and heat (n=2). Two units were mechanical specific/preferential.
- The reduced wind up of lamina I SPB neurons compared to polymodal neurons located deeper in the spinal cord may be informative on the respective function of these 2 populations in the coding of pain.
- Pharmacological disinhibition confirmed that some, but not all lamina I SPB neurons could receive input from light touch afferences. This effect was highly heterogeneous, suggesting that some, but not all lamina I neurons might be "activated" in chronic pain condition.

References

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