Which subset of C-fibres contributes to the TTX resistant component of the spinal evoked field potential in rodents?

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Introduction

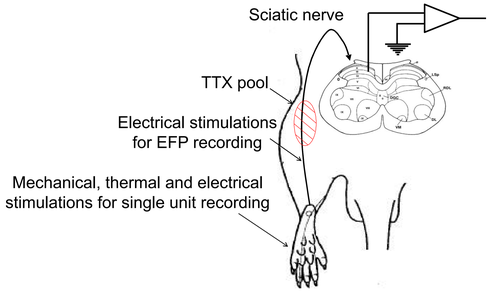
The expression of the voltage gated sodium channel 1.7 (Nav1.7) in nociceptors is mandatory for the generation of pain sensation[1]. Nav1.7 is supposed to play a role in the generation and transport of action potential at the peripheral end, and in synaptic transmission at the central end[2].

Compound action potentials measures performed in vitro and in vivo suggest that the conduction of action potentials in C-fibres along the sciatic nerve is essentially tetrodotoxin (**TTX**) sensitive[3,4]. This is in agreement with the view that Nav1.7 is essential for action potential propagation in C-fibre, as Nav1.7 is the predominant TTX sensitive Nav in nociceptors.

In contrast, Steffens and co-workers have demonstrated that the spinal C-fibre induced evoked field potential (**EFP**) in response to electrical stimulation (**ES**) of the sciatic nerve was insensitive to TTX applied on the sciatic nerve[5].

The present work attempted to explain how TTX, when applied on the sciatic nerve, can suppress the C-fibre compound action potential triggered by a distal ES while leaving intact the corresponding spinal EFP.

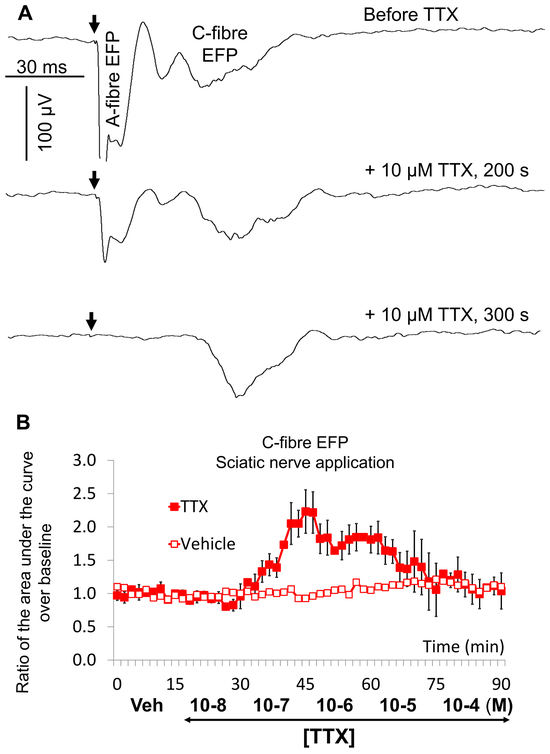
Experimental set up



- EFP and single unit activity in the spinal cord are measured using the same set up with different band pass filters.
- EFP is measured in response to ES of the sciatic nerve. Single unit activity is measured in response to thermal and mechanical as well as well as ES of the receptive field on the hind paw.

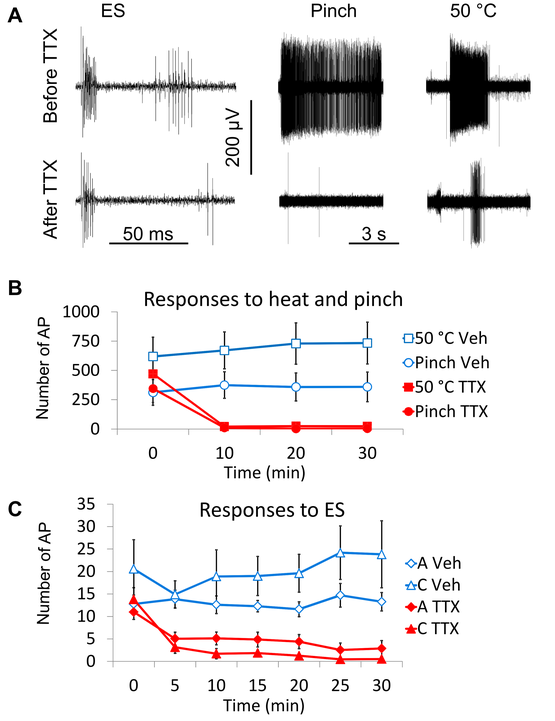
Results

Figure 1 : Spinal cord EFP induced by ES of the sciatic nerve



- **A**: spinal EFP recordings obtained before and at 2 different time points after application of 10 µM TTX on the sciatic nerve. Arrow: time of ES.
- **B**: quantification of the area under the curve of the C-fibre related EFP upon application of increasing TTX concentration every 15 min on the sciatic nerve (TTX, n=3; Vehicle, n=2).
- Note in **A** the persistence of the C-fibre related deflexion, in contrast with the disappearance of the A-fibre related deflexion.

Figure 2: Spinal cord single unit responses to noxious stimuli of the receptive field



- **A**: response of a spinal single unit to noxious stimuli (ES, pinch and 50 °C water jet) before and 15 min after 1 µM TTX application on the sciatic nerve. Note in this example the existence of a residual response after TTX application.
- **B**: mean responses to 50 °C water jet and pinch every 10 min after application of 1 µM TTX on the sciatic nerve (TTX, n=6; Vehicle, n= 7).
- **C**: mean responses to ES of the receptive field in the same experiment as in **B**. A and C refer to the action potentials generated by A- and C-fibres, respectively.

Methods

- Male C57BL6/J mice (20-30 g, Janvier Labs) were terminally anesthetized with urethane, paralyzed, and artificially ventilated.
- End tidal CO2, blood pressure and body temperature were monitored throughout the experiments.
- Electrical activity was measured using a Neurolog set up (Amplification, NL100AK and NL104A; Filter, NL125/126) connected to 2 MΩ tungsten electrode.
- Recording parameters were 1 and 20 K gain, 1 Hz-1 KHz and 3-5 KHz band pass for EFP and single unit recording, respectively.
- For EFP, ES of the sciatic nerve were performed with bipolar Ag wire electrodes (0.5 ms, 0.1 to 2.0 mA). ES were conducted at 1/20 Hz and the average of 5 successive responses used for quantification.
- For single units, mechanical stimuli consisted of pinch with mini haemostats clamp (applied for 5 s), and thermal stimuli consisted of water jet at 50 °C. Electrical stimulations (single square wave pulse, 2 ms, 5 mA) were delivered with 2 stainless steel needles inserted in the receptive field.

Conclusions

- In anesthetized mice, the C-fibre component of the electrically induced EFP was insensitive to TTX applied on the sciatic nerve, confirming the original finding of Steffens and co-workers obtained in rat. The same treatment dramatically reduced or abolished responses to noxious stimuli of all single units recorded in the spinal cord.
- We hypothesize that ES of the sciatic nerve recruits a subset of TTX resistant fibres that are not recruited by stimulation of the receptive field on the glabrous skin.
- The physiological function of the TTX resistant fibre innervating the hind paw and contributing to the C-fibres EFP remains to be determined.

References

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