Direct Activation Induced by Gadolinium in Cultured Human DRG Neurons

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Introduction

Gadolinium based contrast agents (GBCA) are widely used in magnetic resonance imaging (MRI). All GBCAs are based on chelated Gd^{3+} and are considered to be non-toxic to human. However, in patients with renal insufficiency or renal failure, Gd^{3+} ions may accumulate in the body and induce toxicities. Patients with compromised kidney function can develop nephrogenic systemic fibrosis (NSF) after exposure to GBCA. In addition, many of the patients developing NSF, frequently report intense pain, apparently originating from deep tissues and joints. At present, it is not clear whether the pain is directly induced by Gd^{3+} exposure and/or is a consequence of NSF. In an attempt to elucidate the detailed mechanism of Gd^{3+}-induced pathology, we investigated the effects of Gd^{3+} in cultured human dorsal root ganglion (DRG) neurons.

Methods

Human DRGs were collected employing proprietary methods and reagents from ethically consented organ donors in the USA, and following the Ethical Guidelines for Pain Research and Clinical Practice Guidelines in the field of Pain [www.iasp-pain.org/guideline]. DRGs were collected within 2 h post cross-clamp in proprietary cold solution to preserve viability, transported to AnaBios laboratory and immediately dissociated and plated. Cells were tested 2-7 days post-plating. For calcium imaging, cells were loaded with Fluo8-AM. For whole cell patch clamp recordings, cells were held at -80mV and Gd^{3+} was applied to the cell body directly through a perfusion system.

Results

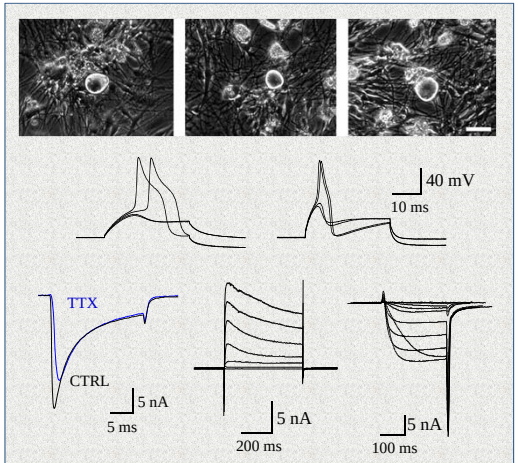


Figure 1. Human DRG Cells in Culture. Cells express both TTX-R as well as TTX-S sodium channels. This results in action potentials exhibiting different kinetics (Middle panel). Also shown, whole cell K^+ and Ca^{2+} currents. The basic electrophysiological properties of human DRG neurons in culture have recently been described in detail (Davidson et al., *PAIN* 2014)

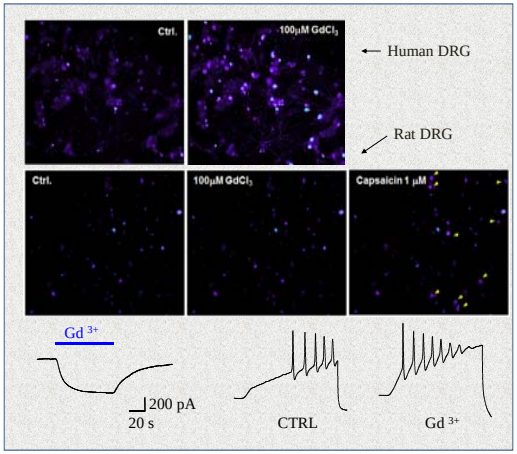
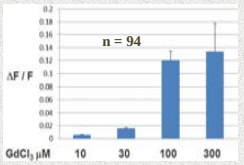


Figure 2. Gd $^{3+}$ Directly Activate Human DRG Neurons. A large population of human DRG neurons are activated by Gd $^{3+}$. In addition to inward currents (bottom panel, right), Gd $^{3+}$ increase the excitability of neurons, as shown by the reduced threshold for action potential generation following ramp current injection.



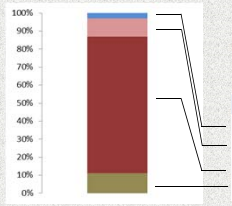
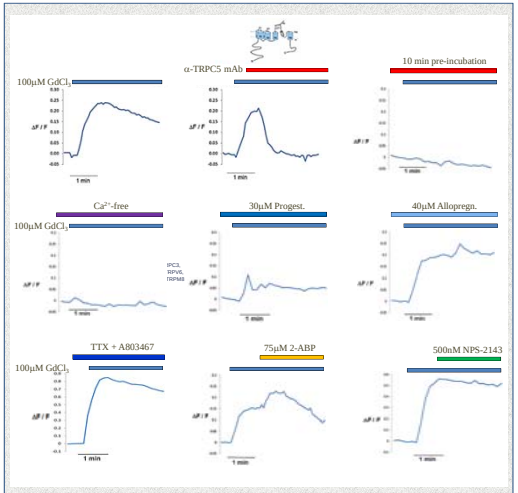


Figure 3. Activation of Human Nociceptors by Gd^{3+}. The activation of human DRGs neurons by Gd^{3+} is dose dependent. The population of Gd^{3+}-sensitive cells largely overlaps with capsaicin-sensitive cells.

%	Gd^{3+}	capsaicin
3	+	-
10	-	+
76	+	+
11	-	-



Agonist	Block	n
α-TRPC5 mAb (10 μg/mL)	+	16
Ca^{2+}-free buffer	+	25
Progesterone (30 μM)	+/-	21
Allopregnanolone (40 μM)	-	18
TTX (500 nM), A803467 (100 nM)	-	23
2-ABP (75 μM)	-	25
NPS-2143 (500 nM)	-	15

Figure 4. TRPC5 Channels Mediate the Effect of Gd $^{3+}$

Conclusions

Gadolinium can directly activate a large population of human nociceptive DRG neurons. This could explain the pain-related side effect of GBCA. The broad elevation of internal calcium level in DRG neurons can trigger various downstream events, such as abnormal sensations, and may contribute to the initiation of subsequent long-term physiological or pathological events, such as NSF.