TRPA1 channel mediates the analgesic action of dipyrone and pyrazolone derivatives

S. Materazzi[1], R. Nassini[1], C. Fusi[1], E. Coppi[1], I.M. Marone[1], F. De Logu[1], D. Preti[2], R. Tonello[1,3], A. Chiarugi[1], R. Patacchini[4], P. Geppetti[1], S. Benemei[1]

[1]Dept. of Health Sciences, Section of Clinical Pharmacology and Oncology, University of Florence, Italy
[2]Dept. of Pharmaceutical Sciences, University of Ferrara, Italy
[3]Programa de Pós-graduação em Ciências Biológicas: Bioquímica Toxicológica, Universidade Federal de Santa Maria, RS, Brazil
[4]Dept. of Corporate Drug Development, Chiesi Farmaceutici SpA, Italy

Since the seminal discovery of antipyrine (AntiP) by Ludwig Knorr in 1883, pyrazolone derivatives (PDs) have been one of the most successful classes of drugs in pain pharmacotherapy. However, although still used by hundreds of millions of people worldwide, the mechanism of the analgesic action of PDs, such as dipyrone (Dip), propyphenazone (PPh) and AntiP, remains unknown. PDs, like NSAIDs/coxibs, have been proposed to act through prostaglandin synthesis inhibition (Hinz et al., 2007). However, pain models responsive to Dip are clearly distinct from those inhibited by classical COX-inhibitors (Brune et al., 1983; Lorenzetti et al., 1985). In addition, COX inhibition by PDs is, however, weak, resulting in poor anti-inflammatory effects, which do not match the remarkable analgesic action. Higher efficiacy of Dip *vs.* NSAIDs in reducing pain with respect to prostaglandin-dependent inflammation and neuropathic pain conditions, such as sciatic pain or post-surgical pain and poor gastrointestinal toxicity, further discriminate PDs from NSAIDs with regard to their pharmacological activities.

The transient receptor potential ankyrin 1 (TRPA1) channel, expressed by a subset of capsaicin-sensitive primary sensory neurons, represents a major target for pain transduction. Emerging evidence indicates that oxidative and nitrative stress and the ensuing lipid peroxidation byproducts produce nociception and hyperalgesia by TRPA1 targeting (Bautista et al., 2006; Taylor-Clark et al., 2009; Trevisani et al., 2007). This novel pathway has been reported to contribute to models of both inflammatory and neuropathic pain (McNamara et al., 2007; Nassini et al., 2014; Trevisan et al., 2013).We hypothesized that PDs target the TRPA1 channel and by this mechanism produce their analgesic effect.

In this study, we demonstrated that PDs selectively inhibit acute nocifensor responses evoked by reactive TRPA1 agonists in mice. In line with recent results obtained with TRPA1 antagonists or with TRPA1 gene deletion, the two most largely used PDs, Dip and PPh, reduce TRPA1-mediated nociception and mechanical allodynia in models of inflammatory and neuropathic pain (such as formalin, carrageenan, partial sciatic nerve ligation, and the peripheral neuropathy evoked by chemotherapeutic drug, bortezomib). Notably, Dip and PPh attenuate carrageenan-evoked mechanical allodynia, without affecting prostaglandin E_2 levels and edema. In addition, PD main metabolites, 4-methylaminoantipyrine (MAA), N-demethylpropyphenazone (dm-PPh) and edaravone (Edar), do not target TRPA1 and do not affect TRPA1-dependent nociception and hyperalgesia.

Thus, present data suggest that attenuation of the pain-producing TRPA1-dependent pathway activated by oxidative stress by-products might also contribute to the analgesic action of PDs in various types of pain in humans (Derry et al., 2010; Ramacciotti et al., 2007). Moreover, present findings strongly support the rationale for the development of TRPA1 antagonists as new analgesics for the treatment of both inflammatory and neuropathic pain. The new chemical entities with TRPA1 antagonistic properties, while maintaining good efficacy in pain treatment and general safety profile of Dip or PPh, should be devoid of the life-threatening hematologic adverse reactions, presumably associable to the chemical structure of PDs.

Bautista et al. 2006. *Cell*. 124**:** 1269-82.
Brune 1983). *Agents Actions.* 13**:** 360-3.
Derry at al. 2010. *Cochrane Database Syst Rev*. 8**:** CD003227.
Hinz et al. 2007. *Faseb J.* 21**:** 2343-51.
Lorenzetti & Ferreira 1985. *Eur J Pharmacol*. 114**:** 375-81.
McNamara et al. 2007. *Proc Natl Acad Sci U S A*. 104**:** 13525-30.
Nassini at al. 2014. *Rev Physiol Biochem Pharmacol*. 2014**:** 26.
Ramacciotti at al. 2007. *Cochrane Database Syst Rev.* 18**:** CD004842.
Taylor-Clark et al. 2009. *Mol Pharmacol.* 75**:** 820-9.
Trevisan et al. 2013. *Cancer Res.* 73**:** 3120-3131.
Trevisani et al. 2007. *Proc Natl Acad Sci U S A.* 104**:** 13519-24.