Amino Acids (2014) 46:1441–1448
DOI 10.1007/s00726-014-1703-5

REVIEW ARTICLE

# Supraspinal metabotropic glutamate receptor subtype 8: a switch to turn off pain

**Enza Palazzo · Vito de Novellis · Francesco Rossi · Sabatino Maione**

Received: 22 January 2014 / Accepted: 18 February 2014 / Published online: 13 March 2014


**Abstract** Glutamate is the main excitatory neurotransmitter in the central nervous system and as such controls the majority of synapses. Glutamatergic neurotransmission is mediated via ionotropic and metabotropic glutamate receptors (iGluRs and mGluRs). Signaling via mGluRs permits to finely tune, rather than turning on/off, the excitatory neurotransmission as the iGluRs do. Eight mGluRs (mGluR1-8) have been cloned so far, which have been divided into three groups based on sequence homology, pharmacological properties and second messenger signaling. mGluRs are widely expressed both on glia and neurons. On neurons they are located both at postsynaptic (group I) and presynaptic sites (group II and III). Group II and III mGluR stimulation reduces glutamate release, which can prove useful in pathological conditions characterized by elevated glutamatergic neurotransmission which include chronic pain. Indeed, mGluRs are widely distributed on pain neuraxis. The recent development of selective mGluR ligands has permitted investigating the individual role of each mGluR on pain control. The development of (*S*)-3,4-dicarboxyphenylglycine, a selective mGluR8 agonist, has revealed the mGluR8 role in inhibiting pain and its related affective consequences in chronic pain conditions. mGluR8 proved also to be overexpressed in pain controlling areas during pathological pain guaranteeing the availability of a switch for turning off abnormal pain. Thus, mGluR8 corresponds to an ideal target in designing novel analgesics. This review will focus on the novel insights into the mGluR8 role on pain control, with particular emphasis on the supraspinal descending pathway, an antinociceptive endogenous source, whose activation or disinhibition (via mGluR8) induces analgesia.

**Keywords** Glutamate · Chronic pain · Metabotropic glutamate receptor 8 · Antinociceptive descending pathway · DHPG

E. Palazzo (✉)
Department of Anaesthesiology, Surgery and Emergency, The Second University of Naples, Piazza Luigi Miraglia 2, 80138 Naples, Italy
e-mail: enza.palazzo@unina2.it

V. de Novellis · F. Rossi · S. Maione (✉)
Pharmacology Division, Department of Experimental Medicine, The Second University of Naples, Via Costantinopoli 16, 80138 Naples, Italy
e-mail: sabatino.maione@unina2.it

## Introduction

The amino acid glutamate is the main excitatory neurotransmitter in the mammalian central nervous system (CNS) (Swanson et al. 2005) whose actions are mediated by ionotropic and metabotropic glutamate receptors (iGluRs and mGluRs) (Conn and Pin 1997). iGluRs receptors are in turn divided into *N*-methyl-D-aspartate (NMDA), α-amino-3-hydroxy-5-methylisoxazole-4-propionic acid (AMPA) and kainic acid (KA) receptors and are involved in the fast responses of the transmission of nervous signals. mGluRs are G-protein coupled receptors which play a modulation on the synaptic transmission and neuronal excitability and are divided into three groups based on sequence homology, pharmacological profile and transduction mechanism (Ferraguti and Shigemoto 2006). Eight mGluRs (mGluR1-8) have been identified so far. mGluR1 and mGluR5 belong to group I and are coupled to phospholipase C (PLC) activation. mGluR2 and mGluR3 belong to group II and mGluR4, mGluR6, mGluR7 and mGluR8 to group III. Group II and III mGluRs are both

associated with adenylate cyclase inhibition (Ferraguti and Shigemoto 2006; Ritter and Hall 2009). mGluRs are expressed at nerve terminals, postsynaptic sites and on glia modulating the glutamate transmission at several levels (Shigemoto et al. 1997; D'Antoni et al. 2008). Group I is mainly localized at the postsynaptic density. Their activation increases neural excitability, whereas group II and group III are primarily located presynaptically where they serve as auto- and hetero-receptors (Ohishi et al. 1995; Petralia et al. 1996). mGluRs are widely distributed throughout the nervous system with the only exception of the mGluR6, which is exclusively expressed in the retina (Vardi et al. 2000). Group III is the largest and less characterized among mGluR groups owing to the lack of selective pharmacological agents. Selective orthosteric and allosteric ligands for group III mGluR subtypes were recently discovered, thus permitting elucidating the role of each of these receptors in CNS disorders. mGluR4, mGluR7 and mGluR8 are able to modulate both the main inhibitory and excitatory neurotransmitter in the CNS and their wide expression in nearly all nervous tissues involved in nociception suggest that these receptors may be involved in the molecular mechanisms at the basis of neurological disorders, characterized by glutamate/GABA neurotransmission imbalance, including chronic pain (al-Ghoul et al. 1993; Hudson et al. 2002; Dmitrieva et al. 2004; Pitcher et al. 2007; Zeilhofer and Zeilhofer 2008; Santangelo et al. 2012). By this subject, mGluRs may be exploited to correct the biochemical abnormality at the base of these neuropathological conditions.

## mGluR8

mGluR8 is alternatively spliced into mGluR8a, 8b and 8c variant isoforms (Corti et al. 1998; Malherbe et al. 1999). mGluR8a and 8b differ in the last 16 amino acids and mGluR8c contains only the *N*-terminal domain (and it is likely to be a secreted protein). Its pre-synaptic localization has been confirmed using immunohistochemistry, electron microscopy and immunogold labeling owing to the absence of selective ligands for mGluR8 (Kinoshita et al. 1996; Shigemoto et al. 1997). The mGluR8 is widely distributed in the CNS. In situ hybridization of mGluR8 mRNA showed high mGluR8 levels in the lateral septum, amygdala, olfactory bulb, pontine gray, lateral reticular nucleus of the thalamus and piriform cortex. Less abundant expression was detected in the cerebral cortex, hippocampus (particularly, lateral perforant path to dentate gyrus), cerebellum and mammillary body of the hypothalamus (Saugstad et al. 1997; Ferraguti et al. 2005). In human brain a higher expression of mGluR8 mRNA was detected in the nucleus caudate. Low levels of mGluR8 were found in the spinal cord in both rats and humans (Robbins et al. 2007). Pharmacological evidence reports mGluR8 expression on the central terminals of primary afferents (Thomas et al. 2001). Immunohistochemistry revealed also the presence of mGluR8 in the retina (Koulen et al. 1999) as well as in the periphery (Tong et al. 2002; Tong and Kirchgessner 2003). mGluR8 seems to be exclusively located on the presynaptic sites of glutamatergic, GABAergic and monoaminergic terminals (Shigemoto et al. 1997; Ferraguti et al. 2005; Palazzo et al. 2011; Rossi et al. 2013). Synaptic glutamate can therefore activate mGluR8, as well as mGluR4 and mGluR7, and negatively regulate its own or other neurotransmitter release (Marabese et al. 2005; Niswender and Conn 2010; Palazzo et al. 2011) (Fig. 1). mGluR8 shows a 1,000 times higher affinity to glutamate than mGluR7 (Schoepp et al. 1999), which may allow its activation onto neighboring synapses by spillover of glutamate (Fig. 1). mGluR8 feedback inhibition may thus have a crucial importance in conditions characterized by glutamate hyperactivity.

## mGluR8 knockout phenotype

Due to the lack of specific antagonist for mGluR8, the characterization of mGluR8-deficient (−/−) mice is currently the only method available to investigate the physiological role of mGluR8. First report using mGluR8 −/− mice stated that this gene deficiency was not associated with any overt pathological phenotype (Gerlai et al. 2002). However, when mGluR8 −/− mice were investigated in different behavioral tests, they showed anxiogenic-like phenotype (Linden et al. 2002, 2003; Duvoisin et al. 2005, 2010a; Robbins et al. 2007). Indeed, the mGluR8-deficient mice exhibited a significant reduction in activity in the inner zone in the open field test and a significant reduction in the entries into the open arms in the elevated plus maze test (Linden et al. 2002; Duvoisin et al. 2005, 2010a; Robbins et al. 2007). mGluR8 −/− mice showed also increased acoustic startle response (Duvoisin et al. 2010a). Other studies using mGluR8 −/− mice expressed no anxiogenic phenotype in unconditioned model of anxiety, including the elevated plus maze, the zero maze and the light–dark box, and more surprisingly a contextual fear deficit, which is consistent with anxiolytic-like phenotype (Gerlai et al. 2002; Fendt et al. 2010). The mGluR8 −/− mice showed also weight gain, being 8 % heavier than their wild-type age-matched controls (Duvoisin et al. 2005). The mGluR8-deficient mice did not show cognitive impairments in some studies (Fendt et al. 2010) and impairments in novel location recognition and in spatial memory retention in the water maze in another study (Duvoisin et al. 2010b). The detrimental effects of mGluR8 −/− on

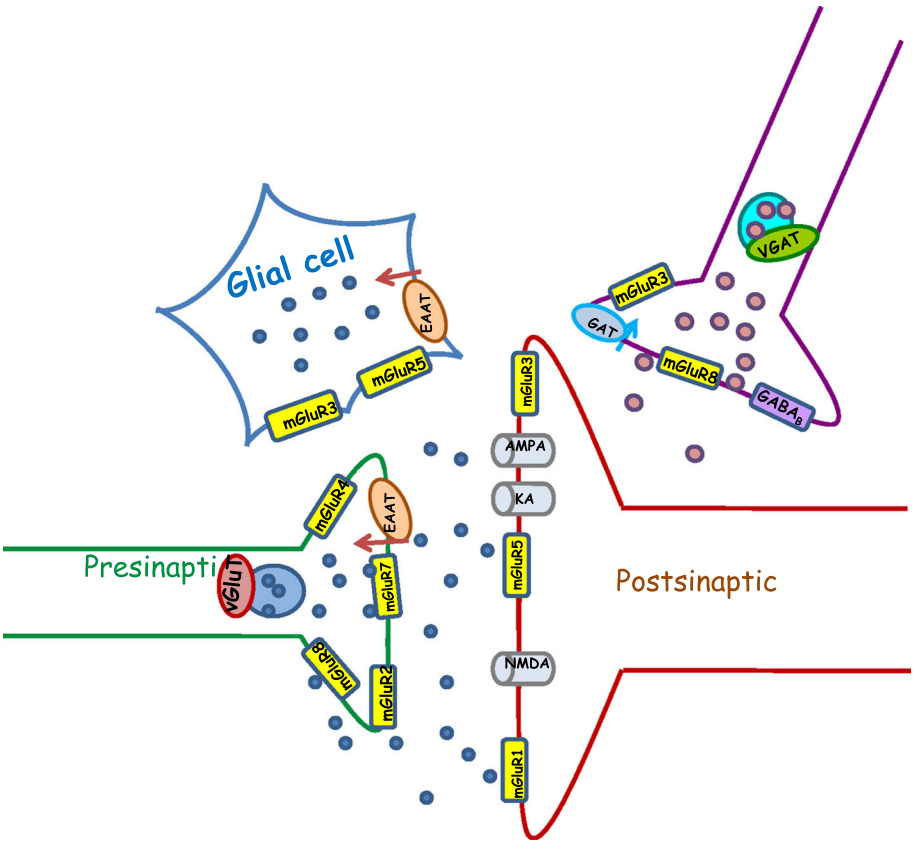


**Fig. 1** Synaptic distribution of mGluRs. Generally, group I mGluRs are localized postsynaptically, and group II and III presynaptically. Group III mGluRs are found both on glutamate and GABA neurons where they inhibit neurotransmitter release. Together with excitatory amino acid transporters (EAAT), presynaptic mGluRs contribute to low glutamate synaptic level and avoid glutamate-driven excitotoxicity at the base of several neurological disorders, included chronic pain. *VGAT* vesicular GABA transporter, *vGluT* vesicular glutamate transporter, *EAAT* excitatory amino acid transporter, *NMDA* *N*-methyl-D-aspartate receptor, *AMPA* α-amino-3-hydroxy-5-methylisoxazole-4-propionic acid receptor, *KA* kainic acid receptor, *GABA$_B$* GABA type B receptor

spatial memory retention in the water maze were more pronounced in female than male mice (Duvoisin et al. 2010b). Notwithstanding the relevant contribution of these studies, the limit of knockout mice strategies is based on the different background strain used and the compensatory effects raised during the development (Crawley 1999; Cryan and Holmes 2005) which may lead to age-dependent phenotype differences (Fendt et al. 2010). Evidence on mGluR8 knockout phenotype is summarized in Table 1.

## mGluR8 selective agonists

L-2-amino-4-phosphonobutyric acid (L-AP4) and L-serine-*O*-phosphate (L-SOP) are endogenous products of phospholipid cleavage which show non-subtype selective orthosteric agonist action on group III mGluRs (Schoepp et al. 1999). (1*S*,3*R*,4*S*)-1-amino-cyclopentane-1,3,4-tricarboxylic acid (ACTP-I) and (*R*,*S*)-4-phosphonophenylglycine (PPG) also behave as non-subtype selective orthosteric agonists of group III mGlu receptors (Schoepp et al. 1999). The available orthosteric antagonists of mGluRs lack specificity on group III mGluR subtypes. MAP4 and MSOP are α-methyl derivatives of the group III receptor agonists L-AP4 and L-SOP and show orthosteric antagonist activity, although they may show agonist activity at mGluR2 (MAP4, MSOP) or mGluR4 and mGluR6 (MAP4) (Schoepp et al. 1999; Swanson et al. 2005). (*R*, *S*)-α-cyclopropyl-4-phosphonophenylglycine (CPPG) is an orthosteric group III mGluR antagonist, which can also antagonize group II mGluRs with nanomolar affinity (Toms et al. 1996). (*S*)-3,4-dicarboxyphenylglycine (DCPG) is the only orthosteric agonist which shows a >100-fold selectivity for mGluR8 over other group III mGluRs, representing a useful tool to probe the physiological roles of mGluR8 and its involvement in the pathophysiology of several neurological and psychiatric disorders (Thomas et al. 2001). DCPG has been shown to play an anticonvulsant effect (Moldrich et al. 2001; Lee et al. 2003; Jiang et al. 2007; Folbergrová et al. 2008), reduce conditioned fear when injected in the amygdala (Schmid and Fendt 2006), inhibit alcohol self-intake (Bäckström and Hyytiä 2005) and prevent amphetamine-induced locomotor hyperactivity in mice (Robbins et al. 2007). Because the doses used for testing alcohol self-intake and amphetamine-induced hyperactivity also reduced spontaneous motor activity, the role of mGluR8 on drug addiction and schizophrenia is still to be determined. The compound AZ12216052 has shown to potentiate the activity of glutamate in human mGluR8b, thus behaving likely as a positive allosteric modulator (PAM) of mGluR8. Its specificity remains to be determined (Duvoisin et al.

**Table 1** A simplified table describing the behavioral mGluR8 knock out phenotype. The strain and the behavioral model used, together with related references, are also indicated

| Strain | Phenotype | Test | References |
|---|---|---|---|
| ICR | No significant difference | Morris water maze (MWM) | Gerlai et al. (2002) |
| | Locomotor hyperactivity | Context and cue-dependent fear conditioning | |
| ICR | Anxiogenic | Elevated plus maze in low illumination conditions | Linden et al. (2002) |
| ICR | Anxiogenic | Elevated plus maze | Linden et al. (2003) |
| | c-Fos-positive cells in the centromedial nucleus of the thalamus | c-Fos immunohistochemistry | |
| C57BL/6 | Heavier than age-matched wild type | Open field | Duvoisin et al. (2005) |
| | Anxiogenic | Elevated plus maze | |
| | Locomotor hypoactivity | | |
| C57BL/6 | Anxiogenic | Elevated plus maze | Robbins et al. (2007) |
| | Reduced sensorimotor gating | Open field | |
| | | Pre-pulse inhibition | |
| C57BL/6 | No anxiogenic | Elevated plus maze | Fendt et al. (2010) |
| | Contextual fear deficit | Elevated zero maze | |
| | Declarative memory impairment | Light–dark box | |
| | Reduced GABAergic activity | Conditioned freezing | |
| | Reduced acoustic startle response (reduced sensorimotor gating, anxiolytic) | Object recognition | |
| | | Chlordiazepoxide-facilitated extinction of operant conditioning | |
| | | Startle system | |
| C57BL/6J | Anxiogenic | Open field | Duvoisin et al. (2010a) |
| | Anxiongenic (male) | Elevated plus maze | |
| | Anxiolytic (female) | Acoustic startle response | |
| | Cognitive impairments (more pronounced in female) | Novel location recognition | |
| | | Water maze | |

2011). Consistently with the mGluR8 −/− mice anxiogenic phenotype, mGluR8 stimulation by DCPG or AZ12216052 reduced anxiety-like behavior in mice (Duvoisin et al. 2010a). Moreover, DCPG, administered in the central nucleus of the amygdala (CeA), reduced anxiety-like behavior in the elevated plus maze in arthritic rats, but not in the controls indicating an anxiolytic activity of DCPG only upon elevated anxiety-like behavior caused by arthritis (Palazzo et al. 2008). Interestingly, DCPG did not affect baseline synaptic transmission in hippocampal slices from adult rats (Ayala et al. 2008) and failed to modify anxiety-like behavior in normal rats after administration into the basolateral amygdala (Stachowicz et al. 2005). Further studies are warranted to assess the effect of site-specific mGluR8 stimulation on the measure of anxiety and conditioned fear in normal and pathological conditions. It seems that mGluR8 requires a pathological state to become effective. Thus mGluR8 proved to be different from non-subtype selective group III mGluR agonists such as LAP4 that inhibited not only pain-related plasticity, but also baseline transmission in the amygdala (Han et al. 2004; Li and Neugebauer 2006). Therefore, the anxiolytic-like effects of mGluR8 activation may depend on neuroplasticity that affects group III mGluR sensitivity (Neugebauer 2008).

## mGluR8 and pain control

The presence of mGluRs within the periaqueductal gray (PAG) (Catania et al. 1994; Leyva et al. 1995) has a strategic importance, with PAG being a crucial area within the antinociceptive descending pathway, whose stimulation induces analgesia (Reynolds 1969; Behbehani 1995). First evidence that mGluR8 in the PAG regulated pain transmission came from a study in which the effect of systemic and intra-PAG administration of DCPG was investigated in inflammatory and neuropathic pain models in mice. Intra-PAG and systemic DCPG alleviated pain responses. The effect proved to be dependent on the time of administration with respect to the peripheral insult. When DCPG was administered 15 min before the peripheral injection of formalin and carrageenan, it decreased pain responses. When the same treatment was carried out 15 min after the

peripheral injection of formalin/carrageenan, it still reduced the late hyperalgesic phase of the formalin test, but did not prevent (neither reduced) the development of carrageenan-induced inflammatory pain. MSOP, a group III receptor antagonist, antagonized the analgesic effect induced by the systemic administration of DCPG when microinjected into the PAG, suggesting that the analgesic effect of systemic DCPG involved mGuR8 within the PAG. Systemic DCPG was ineffective in alleviating thermal hyperalgesia and mechanical allodynia 7 days after the chronic constriction injury of the sciatic nerve, whereas it proved effective 3 days after neuropathic pain induction. Altogether, it seems that within the PAG mGluR8 inhibits the development of formalin and carrageenan-induced hyperalgesia, whereas it would seem less effective in established inflammatory or neuropathic pain (Marabese et al. 2007a). The antinociceptive effect of intra-PAG DCPG was also associated with changes in the rostral ventromedial medulla (RVM) activity (Marabese et al. 2007b), a relay station within the pain descending system, characterized by neurons which respond differently to pain stimuli: ON cells which are activated, OFF cells which are inhibited and neutral cells which are unaffected by nociceptive stimuli (Fields et al. 1983; Heinricher et al. 1989). Systemic or local administration of centrally acting analgesics (such as morphine) inhibits ON cell and increases OFF cell activity; thus, changes in these cell activities are predictive of pain response modulation (Heinricher and Tortorici 1994; Fields et al. 1995; de Novellis et al. 2005). Intra-PAG DCPG reduced thermoceptive responses in the tail flick test, consistently reduced the tail flick-evoked pause duration and delayed the onset of the pause of the OFF cells. A disinhibition of the pain descending system, played by mGluR8 through a decrease of GABA release at PAG level, has been hypothesized in this study and later on demonstrated (Palazzo et al. 2011). This study also evidenced opposite roles played by intra-PAG mGluR7 and mGluR8 stimulation on pain responses and related electrophysiological parameters (Marabese et al. 2007b). The opposite roles played by mGluR7 and mGluR8 on pain were later on evidenced also in the CeA and in the nucleus tractus solitarius (NTS) (Palazzo et al. 2008; Liu et al. 2012). In the CeA the activation of mGluR7 had pronociceptive and anxiety-like effects in normal animals, whereas mGluR8 activation was anti-nociceptive and anxiolytic in the arthritic animals (Palazzo et al. 2008). These results emphasize the importance of mGluR8 and mGluR7 in modulating pain responses, although their roles in normal conditions and different chronic pain models remain to be determined. The better understanding of mechanisms underlying the differential effects of group III mGluR subtypes may not only provide insights into the pathophysiology of chronic pain, but also lead to novel therapeutic strategies for the relief of pain and related affective consequences such as anxiety. Indeed, the facilitatory effect of mGluR7 on pain responses has led to the outcome that its negative modulation may generate analgesia, as recently demonstrated (Palazzo et al. 2013). Opposite actions of mGluR7 and mGluR8 in controlling cardiac nociception have been also evidenced in the NTS, which in contrast to the PAG, plays a facilitatory role in cardiac-evoked muscle hyperalgesia (Liu et al. 2012). Oppositely to what was found in the PAG and CeA, intra-NTS mGluR7-selective non-selective stimulation decreased cardiac nociception, whereas DCPG facilitated it. Both effects were antagonized by intra-NTS MSOP and by vagal deafferentation suggesting that group III mGluRs and mGluR7 in the NTS display an inhibitory effect, while mGluR8 displays a facilitatory effect in modulating cardiac nociception (Liu et al. 2012). Interestingly, the activation of mGluR7, located on the glutamatergic presynaptic terminals at NTS level, inhibits glutamate release, which attenuates NTS-descending facilitatory activity and depresses nociceptive information transmission at the spinal cord level. Activation of mGluR8 on GABAergic presynaptic terminals depresses instead the inhibitory action of GABAergic inter-neurons on the output neurons projecting to the spinal dorsal horn (disinhibition), which enhances NTS-descending facilitatory activity and increases nociceptive information transmission at the spinal cord level (Liu et al. 2012). Thus, the opposite actions of mGluR7 and mGluR8 observed in the NTS with respect to PAG and CeA depend on the proalgesic (NTS) versus analgesic areas (PAG and CeA) and the location of mGluR7 and mGluR8 on glutamatergic/GABAergic terminals. Activation of mGluR8 has shown to inhibit GABA release and simultaneously inhibit pain responses also at the CeA level (Palazzo et al. 2011). In particular, DCPG administered into the CeA increased serotonin and glutamate release, whereas it decreased GABA release in carrageenan-given rats but did not change serotonin, glutamate and GABA release in normal animals. DCPG also increased the thermoceptive threshold and modified the activity of ON and OFF cells of the RVM consistently with behavioral analgesia in rats treated with carrageenan. DCPG was ineffective on pain responses and ON and OFF cell activity in control rats. An increase in mGluR8 expression has been found in the CaA of carrageenan-given rats. Moreover, the double immune-staining revealed that mGluR8 was coexpressed with vesicular GABA transporter (vGAT)-positive profiles. Thus mGluR8 within the CeA, which proved to be overexpressed on GABAergic terminals in chronic pain conditions, seems similar to a switch to be turned on available to inhibit pain when necessary. Recently, the role of the mGluR8 has been investigated also in the dorsal striatum (DS). Consistently

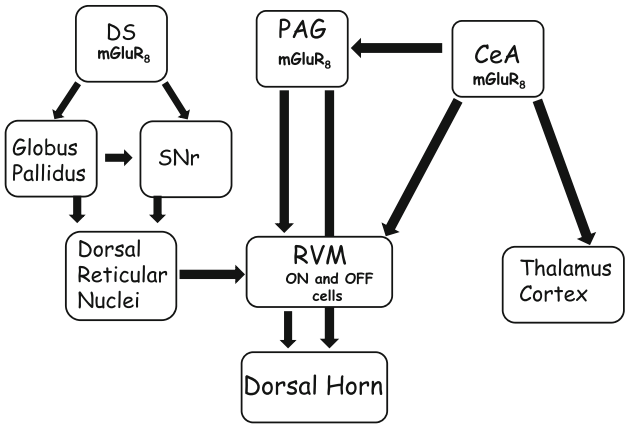


**Fig. 2** A simplified scheme illustrating the major pathways from DS, PAG and CeA to the RVM, which represents the converging point within the antinociceptive descending pathway. PAG has few direct connections to the dorsal horn of the spinal cord and uses the RVM as a relay station. The connection from DS to the RVM is far from being direct and the converging point is the dorsal reticular nuclei area. Modulation of pain by CeA is regulated through projections to the PAG and through this to the RVM and dorsal horn or direct connections to RVM. *DS* dorsal striatum, *PAG* periaqueductal gray, *CeA* central nucleus of the amygdala, *RVM* rostral ventromedial medulla

with the previous studies, the intra-DS DCPG did not modify the activity of the ON and OFF cells in sham rats, but reduced the mechanical withdrawal threshold, the ongoing and tail flick-evoked activity of the ON cells, while it increased the activity of the OFF cells in the neuropathic pain model induced by the spare nerve injury (SNI) of the sciatic nerve in rats. The AZ12216052 compound behaves as DCPG in increasing tail flick latency, the OFF cell activity and decreasing the ON cell activity in SNI rats only, but was less potent. VU0155041, a selective mGluR4 PAM, was instead ineffective in changing thermal nociception and ON and OFF cell activity in both sham and SNI rats. In the DS the mGluR8 gene and immunoreactivity were decreased and proved to be expressed on GABAergic terminals, associated with a protein increase in SNI rats. These results suggest that DS mGluR8 stimulation inhibits pain responses via RVM and confirm that DCPG is effective in chronic pain condition only (Rossi et al. 2013). Mechanisms, meaning and strategies to exploit mGluR8 plasticity in chronic pain conditions in pain-modulating sites deserve further investigation. The antinociceptive responses produced by mGluR8 stimulation in supraspinal areas such as PAG, CeA and DS pass through the modulation of RVM activity (Fig. 2) and the activation of the antinociceptive descending pathway. mGluR8-mediated inhibition of GABA release drive to the antinociceptive descending pathway disinhibition and consequent behavioral analgesia.

## Conclusions

The systemic and site-specific supraspinal stimulation of mGluR8 was shown to reduce pain responses. A particularity of this receptor is the plasticity shown in chronic pain conditions. Indeed, several studies have shown that selective mGluR8 stimulation is effective in pathological pain conditions only, thus assuring a switch to turn off pain only when pain is pathological (and no more useful). mGluR8 has been shown to be often expressed on GABAergic terminals whose stimulation leads to GABA release reduction. Thus, in antinociceptive areas such as PAG, DS and CeA the disinhibition leads to an enhancement of the antinociceptive descending pathway and behavioral analgesia. The development of more and more selective orthosteric and allosteric agonist for mGluR8 able to cross the blood brain barrier may hopefully lead to the development of novel and more effective analgesics.

**Conflict of interest** The authors declare that they have no conflict of interest.

## References

al-Ghoul WM, Volsi GL, Weinberg RJ, Rustioni A (1993) Glutamate immunocytochemistry in the dorsal horn after injury or stimulation of the sciatic nerve of rats. Brain Res Bull 30:453–459

Ayala JE, Niswender CM, Luo Q, Banko JL, Conn PJ (2008) Group III mGluR regulation of synaptic transmission at the SC-CA1 synapse is developmentally regulated. Neuropharmacology 54:804–814

Bäckström P, Hyytiä P (2005) Suppression of alcohol self-administration and cue-induced reinstatement of alcohol seeking by the mGlu2/3 receptor agonist LY379268 and the mGlu8 receptor agonist (*S*)-3,4-DCPG. Eur J Pharmacol 528:110–118

Behbehani MM (1995) Functional characteristics of the midbrain periaqueductal gray. Prog Neurobiol 46:575–605

Catania MV, De Socarraz H, Penney JB, Young AB (1994) Metabotropic glutamate receptor heterogeneity in rat brain. Mol Pharmacol 45:626–636

Conn PJ, Pin JP (1997) Pharmacology and functions of metabotropic glutamate receptors. Annu Rev Pharmacol Toxicol 37:205–237

Corti C, Restituito S, Rimland JM, Brabet I, Corsi M, Pin JP, Ferraguti F (1998) Cloning and characterization of alternative mRNA forms for the rat metabotropic glutamate receptors mGluR7 and mGluR8. Eur J Neurosci 10:3629–3641

Crawley JN (1999) Behavioral phenotyping of transgenic and knockout mice: experimental design and evaluation of general health, sensory functions, motor abilities, and specific behavioral tests. Brain Res 835:18–26

Cryan JF, Holmes A (2005) The ascent of mouse: advances in modelling human depression and anxiety. Nat Rev Drug Discov 4:775–790

D'Antoni S, Berretta A, Bonaccorso CM, Bruno V, Aronica E, Nicoletti F, Catania MV (2008) Metabotropic glutamate receptors in glial cells. Neurochem Res 33:2436–2443

de Novellis V, Mariani L, Palazzo E, Vita D, Marabese I, Scafuro M, Rossi F, Maione S (2005) Periaqueductal grey CB1 cannabinoid and metabotropic glutamate subtype 5 receptors modulate

changes in rostral ventromedial medulla neuronal activities induced by subcutaneous formalin in the rat. Neuroscience 134:269–281

Dmitrieva N, Rodríguez-Malaver AJ, Pérez J, Hernández L (2004) Differential release of neurotransmitters from superficial and deep layers of the dorsal horn in response to acute noxious stimulation and inflammation of the rat paw. Eur J Pain 8:245–252

Duvoisin RM, Zhang C, Pfankuch TF, O'Connor H, Gayet-Primo J, Quraishi S, Raber J (2005) Increased measures of anxiety and weight gain in mice lacking the group III metabotropic glutamate receptor mGluR8. Eur J Neurosci 22:425–436

Duvoisin RM, Pfankuch T, Wilson JM, Grabell J, Chhajlani V, Brown DG, Johnson E, Raber J (2010a) Acute pharmacological modulation of mGluR8 reduces measures of anxiety. Behav Brain Res 212:168–173

Duvoisin RM, Villasana L, Pfankuch T, Raber J (2010b) Sex-dependent cognitive phenotype of mice lacking mGluR8. Behav Brain Res 209:21–26

Duvoisin RM, Villasana L, Davis MJ, Winder DG, Raber J (2011) Opposing roles of mGluR8 in measures of anxiety involving non-social and social challenges. Behav Brain Res 221:50–54

Fendt M, Bürki H, Imobersteg S, van der Putten H, McAllister K, Leslie JC, Shaw D, Hölscher C (2010) The effect of mGlu8 deficiency in animal models of psychiatric diseases. Genes Brain Behav 9:33–44

Ferraguti F, Shigemoto R (2006) Metabotropic glutamate receptors. Cell Tissue Res 326:483–504

Ferraguti F, Klausberger T, Cobden P, Baude A, Roberts JD, Szucs P, Kinoshita A, Shigemoto R, Somogyi P, Dalezios Y (2005) Metabotropic glutamate receptor 8-expressing nerve terminals target subsets of GABAergic neurons in the hippocampus. J Neurosci 25:10520–10536

Fields HL, Bry J, Hentall I, Zorman G (1983) The activity of neurons in the rostral medulla of the rat during withdrawal from noxious heat. J Neurosci 3:2545–2552

Fields HL, Malick A, Burstein R (1995) Dorsal horn projection targets of ON and OFF cells in the rostral ventromedial medulla. J Neurophysiol 74:1742–1759

Folbergrová J, Druga R, Haugvicová R, Mares P, Otáhal J (2008) Anticonvulsant and neuroprotective effect of (*S*)-3,4-dicarboxyphenylglycine against seizures induced in immature rats by homocysteic acid. Neuropharmacology 54:665–675

Gerlai R, Adams B, Fitch T, Chaney S, Baez M (2002) Performance deficits of mGluR8 knockout mice in learning tasks: the effects of null mutation and the background genotype. Neuropharmacology 43:235–249

Han JS, Bird GC, Neugebauer V (2004) Enhanced group III mGluR-mediated inhibition of pain-related synaptic plasticity in the amygdala. Neuropharmacology 46:918–926

Heinricher MM, Tortorici V (1994) Interference with GABA transmission in the rostral ventromedial medulla: disinhibition of off-cells as a central mechanism in nociceptive modulation. Neuroscience 63:533–546

Heinricher MM, Barbaro NM, Fields HL (1989) Putative nociceptive modulating neurons in the rostral ventromedial medulla of the rat: firing of on- and off-cells is related to nociceptive responsiveness. Somatosens Mot Res 6:427–439

Hudson LJ, Bevan S, McNair K, Gentry C, Fox A, Kuhn R, Winter J (2002) Metabotropic glutamate receptor 5 upregulation in A-fibers after spinal nerve injury: 2-methyl-6-(phenylethynyl)-pyridine (MPEP) reverses the induced thermal hyperalgesia. J Neurosci 22:2660–2668

Jiang FL, Tang YC, Chia SC, Jay TM, Tang FR (2007) Anticonvulsive effect of a selective mGluR8 agonist (*S*)-3,4-dicarboxyphenylglycine (*S*-3,4-DCPG) in the mouse pilocarpine model of status epilepticus. Epilepsia 48:783–792

Kinoshita A, Ohishi H, Neki A, Nomura S, Shigemoto R, Takada M, Nakanishi S, Mizuno N (1996) Presynaptic localization of a metabotropic glutamate receptor, mGluR8, in the rhinencephalic areas: a light and electron microscope study in the rat. Neurosci Lett 207:61–64

Koulen P, Kuhn R, Wässle H, Brandstätter JH (1999) Modulation of the intracellular calcium concentration in photoreceptor terminals by a presynaptic metabotropic glutamate receptor. Proc Natl Acad Sci USA 96:9909–9914

Lee JJ, Jane DE, Croucher MJ (2003) Anticonvulsant dicarboxyphenylglycines differentially modulate excitatory amino acid release in the rat cerebral cortex. Brain Res 977:119–123

Leyva J, Maione S, Pallotta M, Berrino L, Rossi F (1995) Metabotropic and ionotropic glutamate receptors mediate opposite effects on periaqueductal gray matter. Eur J Pharmacol 285:123–126

Li W, Neugebauer V (2006) Differential changes of group II and group III mGluR function in central amygdala neurons in a model of arthritic pain. J Neurophysiol 96:1803–1815

Linden AM, Johnson BG, Peters SC, Shannon HE, Tian M, Wang Y, Yu JL, Köster A, Baez M, Schoepp DD (2002) Increased anxiety-related behavior in mice deficient for metabotropic glutamate 8 (mGlu8) receptor. Neuropharmacology 43:251–259

Linden AM, Baez M, Bergeron M, Schoepp DD (2003) Increased c-Fos expression in the centromedial nucleus of the thalamus in metabotropic glutamate 8 receptor knockout mice following the elevated plus maze test. Neuroscience 121:167–178

Liu XH, Han M, Zhu JX, Sun N, Tang JS, Huo FQ, Li J, Xu FY, Du JQ (2012) Metabotropic glutamate subtype 7 and 8 receptors oppositely modulate cardiac nociception in the rat nucleus tractus solitarius. Neuroscience 220:322–329

Malherbe P, Kratzeisen C, Lundstrom K, Richards JG, Faull RL, Mutel V (1999) Cloning and functional expression of alternative spliced variants of the human metabotropic glutamate receptor 8. Brain Res Mol Brain Res 67:201–210

Marabese I, de Novellis V, Palazzo E, Mariani L, Siniscalco D, Rodella L, Rossi F, Maione S (2005) Differential roles of mGlu8 receptors in the regulation of glutamate and gamma-aminobutyric acid release at periaqueductal grey level. Neuropharmacology 49:157–166

Marabese I, de Novellis V, Palazzo E, Scafuro MA, Vita D, Rossi F, Maione S (2007a) Effects of (*S*)-3,4-DCPG, an mGlu8 receptor agonist, on inflammatory and neuropathic pain in mice. Neuropharmacology 52:253–262

Marabese I, Rossi F, Palazzo E, de Novellis V, Starowicz K, Cristino L, Vita D, Gatta L, Guida F, Di Marzo V, Rossi F, Maione S (2007b) Periaqueductal gray metabotropic glutamate receptor subtype 7 and 8 mediate opposite effects on amino acid release, rostral ventromedial medulla cell activities, and thermal nociception. J Neurophysiol 98:43–53

Moldrich RX, Beart PM, Jane DE, Chapman AG, Meldrum BS (2001) Anticonvulsant activity of 3,4-dicarboxyphenylglycines in DBA/2 mice. Neuropharmacology 40:732–735

Neugebauer V (2008) Group III metabotropic glutamate receptors (mGlu4, mGlu6, mGlu7, and mGlu8). In: Gereau RW, Swanson GT (eds) The glutamate receptors. Humana Press, Springer, Clifton, pp 489–508

Niswender CM, Conn PJ (2010) Metabotropic glutamate receptors: physiology, pharmacology, and disease. Annu Rev Pharmacol Toxicol 50:295–322

Ohishi H, Akazawa C, Shigemoto R, Nakanishi S, Mizuno N (1995) Distributions of the mRNAs for L-2-amino-4-phosphonobutyrate-sensitive metabotropic glutamate receptors, mGluR4 and mGluR7, in the rat brain. J Comp Neurol 360:555–570

Palazzo E, Fu Y, Ji G, Maione S, Neugebauer V (2008) Group III mGluR7 and mGluR8 in the amygdala differentially modulate

nocifensive and affective pain behaviors. Neuropharmacology 55:537–545

Palazzo E, Marabese I, Soukupova M, Luongo L, Boccella S, Giordano C, de Novellis V, Rossi F, Maione S (2011) Metabotropic glutamate receptor subtype 8 in the amygdala modulates thermal threshold, neurotransmitter release, and rostral ventromedial medulla cell activity in inflammatory pain. J Neurosci 31:4687–4697

Palazzo E, Marabese I, Luongo L, Boccella S, Bellini G, Giordano ME, Rossi F, Scafuro M, de Novellis V, Maione S (2013) Effects of a metabotropic glutamate receptor subtype 7 negative allosteric modulator in the periaqueductal grey on pain responses and rostral ventromedial medulla cell activity in rat. Mol Pain 9:44–55

Petralia RS, Wang YX, Niedzielski AS, Wenthold RJ (1996) The metabotropic glutamate receptors, mGluR2 and mGluR3, show unique postsynaptic, presynaptic and glial localizations. Neuroscience 71:949–976

Pitcher MH, Ribeiro-da-Silva A, Coderre TJ (2007) Effects of inflammation on the ultrastructural localization of spinal cord dorsal horn group I metabotropic glutamate receptors. J Comp Neurol 505:412–423

Reynolds DV (1969) Surgery in the rat during electrical analgesia induced by focal brain stimulation. Science 164:444–445

Ritter SL, Hall RA (2009) Fine-tuning of GPCR activity by receptor-interacting proteins. Nat Rev Mol Cell Biol 10:819–830

Robbins MJ, Starr KR, Honey A, Soffin EM, Rourke C, Jones GA, Kelly FM, Strum J, Melarange RA, Harris AJ, Rocheville M, Rupniak T, Murdock PR, Jones DN, Kew JN, Maycox PR (2007) Evaluation of the mGlu8 receptor as a putative therapeutic target in schizophrenia. Brain Res 1152:215–227

Rossi F, Marabese I, De Chiaro M, Boccella S, Luongo L, Guida F, De Gregorio D, Giordano C, de Novellis V, Palazzo E, Maione S (2013) Dorsal striatum metabotropic glutamate receptor 8 affects nocifensive responses and rostral ventromedial medulla cell activity in neuropathic pain conditions. J Neurophysiol

Santangelo RM, Acker TM, Zimmerman SS, Katzman BM, Strong KL, Traynelis SF, Liotta DC (2012) Novel NMDA receptor modulators: an update. Expert Opin Ther Pat 22:1337–1352

Saugstad JA, Kinzie JM, Shinohara MM, Segerson TP, Westbrook GL (1997) Cloning and expression of rat metabotropic glutamate receptor 8 reveals a distinct pharmacological profile. Mol Pharmacol 51:119–125

Schmid S, Fendt M (2006) Effects of the mGluR8 agonist (*S*)-3,4-DCPG in the lateral amygdala on acquisition/expression of fear-potentiated startle, synaptic transmission, and plasticity. Neuropharmacology 50:154–164

Schoepp DD, Jane DE, Monn JA (1999) Pharmacological agents acting at subtypes of metabotropic glutamate receptors. Neuropharmacology 38:1431–1476

Shigemoto R, Kinoshita A, Wada E, Nomura S, Ohishi H, Takada M, Flor PJ, Neki A, Abe T, Nakanishi S, Mizuno N (1997) Differential presynaptic localization of metabotropic glutamate receptor subtypes in the rat hippocampus. J Neurosci 17:7503–7522

Stachowicz K, Kłak K, Pilc A, Chojnacka-Wójcik E (2005) Lack of the antianxiety-like effect of (*S*)-3,4-DCPG, an mGlu8 receptor agonist, after central administration in rats. Pharmacol Rep 57:856–860

Swanson CJ, Bures M, Johnson MP, Linden AM, Monn JA, Schoepp DD (2005) Metabotropic glutamate receptors as novel targets for anxiety and stress disorders. Nat Rev Drug Discov 4:131–144

Thomas NK, Wright RA, Howson PA, Kingston AE, Schoepp DD, Jane DE (2001) (*S*)-3,4-DCPG, a potent and selective mGlu8a receptor agonist, activates metabotropic glutamate receptors on primary afferent terminals in the neonatal rat spinal cord. Neuropharmacology 40:311–318

Toms NJ, Jane DE, Kemp MC, Bedingfield JS, Roberts PJ (1996) The effects of (RS)-alpha-cyclopropyl-4-phosphonophenylglycine ((RS)-CPPG), a potent and selective metabotropic glutamate receptor antagonist. Br J Pharmacol 119:851–854

Tong Q, Kirchgessner AL (2003) Localization and function of metabotropic glutamate receptor 8 in the enteric nervous system. Am J Physiol Gastrointest Liver Physiol 285:992–1003

Tong Q, Ouedraogo R, Kirchgessner AL (2002) Localization and function of group III metabotropic glutamate receptors in rat pancreatic islets. Am J Physiol Endocrinol Metab 282:E1324–E1333

Vardi N, Duvoisin R, Wu G, Sterling P (2000) Localization of mGluR6 to dendrites of ON bipolar cells in primate retina. J Comp Neurol 423:402–412

Zeilhofer HU, Zeilhofer UB (2008) Spinal dis-inhibition in inflammatory pain. Neurosci Lett 437:170–174