

## Cochlear dopamine release is modulated by group II metabotropic glutamate receptors via GABAergic neurotransmission

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### Abstract

Dopamine (DA), released from the lateral olivocochlear efferent fibers, is suggested to be neuroprotective against ischemia and noise exposure in the mammalian cochlea because it can reduce the postsynaptic excitotoxic effect of glutamate on the dendrite of the afferent auditory neuron. Using *in vitro* microvolume superfusion method on isolated guinea pig cochlea preparation, we found that the selective mGluR2/3 agonist (2*R*,4*R*)-aminopyrrolidine-2,4-dicarboxylic acid (2*R*,4*R*-APDC) significantly increased the release of DA in a dose-dependent manner. Other mGluR agonists, acting on groups I and III receptors (3,5-dihydroxyphenylglycine, amino-4-phosphonobutyric acid) and antagonists (2-methyl-6-(phenylethynyl)pyridine), (2*S*)-2-amino-2-(1*S*,2*S*-2-carboxycyclopropan-1-yl-3-(xanth9-yl)propanoic acid,  $\alpha$ -methylserine-*O*-phosphate), were ineffective. The GABA<sub>A</sub> antagonist bicuculline (10  $\mu$ M) could antagonize the effect of 2*R*,4*R*-APDC suggesting that the mGluR-mediated enhancement of DA release was most likely attributable to a disinhibitory mechanism involving local GABAergic fibers. Bicuculline alone could also elevate the DA outflow indicating that cochlear GABA controls local DA release tonically. Our findings expand the view on the local effects of glutamate in the cochlea by showing the ability of the excitatory neurotransmitter to alleviate its own action on type I afferents via mGluRs and initiate a neuroprotective mechanism.

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Glutamate excitotoxicity is a well-known mechanism of ischemia- or noise-induced hearing loss involving postsynaptic overexcitation, subsequent swelling and degeneration of the afferent dendrites [26,28,29,30]. Glutamate is released from the inner hair cells and activates type I afferent fibers. Lateral olivocochlear fibers are forming en passant synapses on the dendrite of the type I afferent fibers and release transmitters, such as acetylcholine, dopamine (DA), gamma-amino butyric acid (GABA), and neuropeptides (CGRP, enkephalins) [7]. GABA receptors have been detected in this synapse [6,17]. DA is thought to have a protective role against glutamate excitotoxicity involving a postsynaptic antagonism on the effect of the released glutamate [11,25,27,31,35]. Pujol et al., [29] found that piribedil (a D2/D3 receptor agonist)

had a preventive effect on the ischemia-induced morphological changes in the cochlea. Moreover, intracochlear perfusion of piribedil could reduce the compound action potential (CAP) threshold shift during noise exposure [3]. DA had no effect on the spontaneous firing rate of the cochlear afferent nerve, while DA and both D1 and D2 agonists could depress the glutamate-induced firing [24]. In contrast, DA could enhance the natural stimulus of the cochlea as suggested by the DA-evoked increase in the spontaneous and sound-induced activity of the afferent cochlear nerve [31].

Previous studies showed that activation of GABA<sub>A</sub> receptors reduced the firing rate of the afferent cochlear nerve evoked by the glutamate and ionotropic glutamate receptor agonists, while the spontaneous activity was not affected [1,8]. Therefore, a neuroprotective effect of GABA can be assumed. The presence of GABA<sub>A</sub> [6,19] and GABA<sub>B</sub> receptors [17] on the spiral ganglion cells and on the afferent

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nerve dendrites [39] has been clearly shown. All kinds of ionotropic glutamate receptors (iGluRs) have been found in the dendrite of the afferent nerve [23,32]. Although several studies confirmed the presence of metabotropic glutamate receptors (mGluRs) on both the spiral ganglion cells and inner hair cells [2,22,33], little is known about their function in the cochlea. As they have slower and longer kinetics than iGluRs, they are more likely involved in modulatory actions in the cochlea [16]. It is particularly important that groups II and III mGluR agonists are considered as neuroprotective compounds [21]. In the present study, we aimed to explore the functional role of mGluRs in the cochlea. In the light of the known neuroprotective feature of mGluRs, we assumed that the cochlear DA release may be functionally linked to activation of mGluRs in the cochlea.

Male guinea pigs (Toxicoop, Hungary) weighing 200–300 g were used for stereomicroscopic separation of the cochlear tissue, as described previously [10,14]. All efforts were made to minimize the number of animals and pain. Experiments were carried out in accordance with the National Institute of Health Guide for the Care and Use of Laboratory Animals. Following isolation of the two cochleae, *in vitro* microvolume superfusion method [38] was used to measure dopamine release. To imitate physiological conditions our experiments were performed in perilymph-like solution [10] thermoregulated at 37 °C and continuously saturated with 100% O<sub>2</sub>. Isolated cochlea tissues were incubated for 35 min at 37 °C in 1 ml of perilymph-like solution containing 0.1 μM <sup>3</sup>H-dopamin (31.0 Ci/mmol, Amersham). After incubation, each cochlea was transferred to a thermoregulated (37 °C) plexiglas microvolume chamber (inside volume 100 μl) and superfused at a rate of 3 ml/min. After 60 min pre-perfusion, the outflow was collected in 3 min fractions. The released radioactivity was determined by assaying 500 μl-aliquot of each sample with liquid scintillation counter (Packard Tri-Carb 1900TR). Cochleae were electrically stimulated at 60 V, 2 Hz, 0.5 ms impulse duration for 3 min with a Grass S88 stimulator (West Warwick, USA) during the 3rd and, in some cases during the 13th collection periods (S<sub>1</sub> and S<sub>2</sub>) through a platinum electrode inserted at the top and bottom of the chamber. After collecting the samples each cochlea was transferred to 500 μl of 10% trichloroacetic acid for suspension for 24 h, then 100 μl was used to measure tissue content of radioactivity. To express the release of DA during one collecting period, the fractional release (FR) of tritium-outflow was calculated as the percentage of total radioactivity present in the tissue at the time of sample collection. The release of tritium evoked by field stimulations was calculated by the area-under-the-curve method [13–15]. The effects of drugs on the field stimulation-evoked [<sup>3</sup>H]DA release are expressed by the ratio of FR of S<sub>2</sub> to FR of S<sub>1</sub> (FRS<sub>2</sub>/FRS<sub>1</sub>). The effect of the drugs on the resting outflow is determined as the ratio of the sum of the two highest consecutive FR *R* values in the presence of the drug (FRR<sub>2</sub>) to the sum of

the two consecutive FR *R* values before the drug reached the cochleae (FRR<sub>1</sub>; FRR<sub>2</sub>/FRR<sub>1</sub>). ANOVA was used in all experiments' statistical analyses. Data are expressed as the mean ± S.E.M. (*n* = number of cochleae). Tukey's post hoc test was applied to determine the significance of data using the Statistica 6.0 program (Statsoft Inc., USA). Levels of significance are as follows: \**P* < 0.05, \*\**P* < 0.01, \*\*\**P* < 0.001. We purchased 3,5-dihydroxyphenylglycine (3,5-DHPG), 2-methyl-6-(phenylethynyl)pyridine (MPEP), (2*R*,4*R*)-Aminopyrrolidine-2,4-dicarboxylic acid (2*R*,4*R*-APDC), (2*S*)-2-amino-2-(1*S*,2*S*-2-carboxycyclopropan-1-yl-3-(xantho-9-yl)propanoic acid (LY-341495)), amino-4-phosphonobutyric acid (L-AP-4) α-methylserine-*O*-phosphate (MSOP) from Tocris. Bicuculline methiodid was from Sigma.

In order to explore the role of mGluRs in the regulation of cochlear DA release we tested the effect of selective agonists and antagonists of groups I–III mGluRs on the spontaneous and evoked release of DA from isolated guinea pig cochleae. Perfusion with agonists and antagonists of group I mGluRs (DHPG, 100 μM and MPEP 20 μM) failed to alter the release of DA from the cochlea (*n* = 6, 8, Fig. 1). In contrast, mGluRs belonging to group II were active in modulating DA release: administration of the agonist 2*R*,4*R*-APDC (100 μM) increased the release of DA at rest from isolated cochleae (Fig. 1A and D), as revealed by the 21% increase in the FRR<sub>2</sub>/FRR<sub>1</sub> value. This increase in the release of DA was highly significant compared with the control (*P* = 0.002, *n* = 12). The electrically evoked DA release did not change in the presence of 2*R*,4*R*-APDC (Fig. 1A and C). The selective group II antagonist LY-341495 (0.2 μM) caused no significant effect on either the spontaneous or the evoked release of DA (*n* = 7, Fig. 1). Group III agonist (L-AP-4, 100 μM) and antagonist (MSOP, 100 μM) caused no significant effect on the DA outflow (*n* = 5–6, Fig. 1). The lack of effect of 2*R*,4*R*-APDC on the field stimulation-evoked release suggests that those receptors, which were modulated by this drug, are functionally not present on dopaminergic terminals. To further investigate the apparent excitation by the mGluR group II agonist on the spontaneous DA release from the cochlea, we tested 2*R*,4*R*-APDC at 100 and 300 μM concentrations, in the absence of second electrical stimulation. These experiments revealed that the action of 2*R*,4*R*-APDC seems to be dose-dependent (Fig. 2A and D). The increase in the release lasted through the perfusion with the drugs suggesting no desensitization of the receptor and the release returned to the baseline at the end of the perfusion, indicating that the effect of 100 μM 2*R*,4*R*-APDC can be washed out (Fig. 2A). In contrast, the release by 300 μM 2*R*,4*R*-APDC was long-lasting and did not return to the baseline within the observation period, as revealed by the ratio of the fractional release values of the last three and the first two samples (control: 0.67 ± 0.02, 0.80 ± 0.02; *P* < 0.001, Fig. 2A). The specific group II mGluR antagonist LY-341495 could prevent the DA-releasing effect of 2*R*,4*R*-APDC proving the selective activation of group II receptors by 2*R*,4*R*-APDC (*P* < 0.01, Fig. 2B).

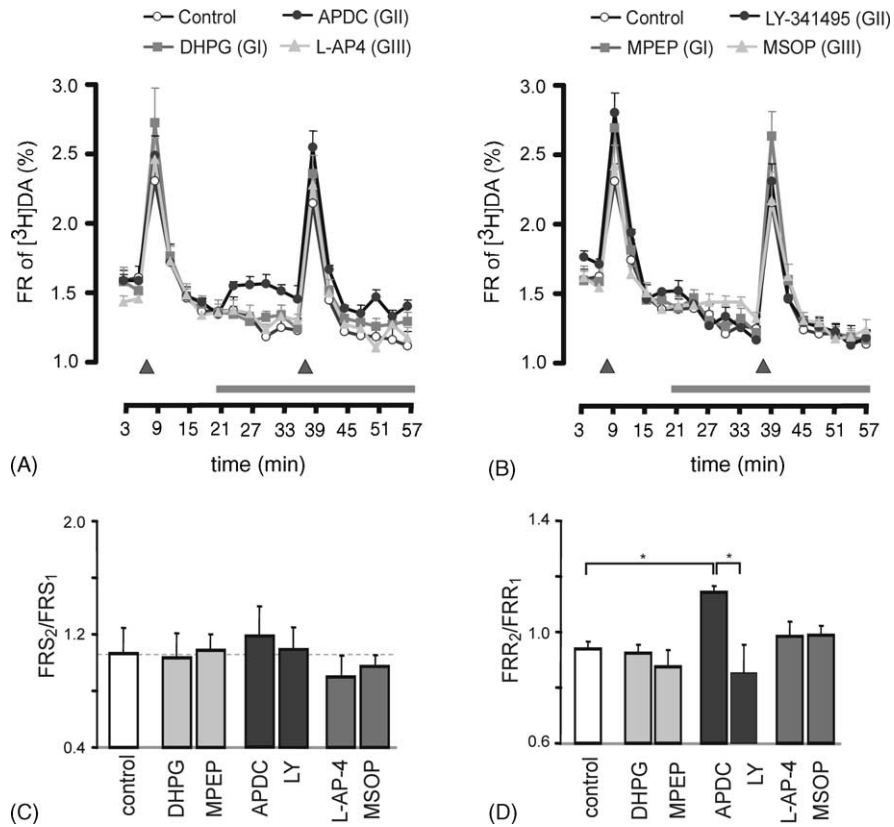


Fig. 1. Time-lapse changes in DA outflow from isolated guinea pig cochleae in the presence of agonists and antagonists of groups I–III metabotropic glutamate receptors. (A) The group II mGluR agonist 2*R*,4*R*-APDC (100  $\mu$ M, black circle) evoked DA release from the cochlea. Group I (DHPG, 100  $\mu$ M, gray rectangle) and III (L-AP4, 100  $\mu$ M, gray triangle) agonists caused no effect compared to control (open circle). Electrical field stimulations (black triangles) evoked reversible and reproducible increase in the fractional release (FR). Drugs were applied in the perfusion solution as the horizontal bar indicates. (B) The group II mGluR antagonist LY-341495 (0.2  $\mu$ M, black circle), the group I antagonist MPEP (20  $\mu$ M, gray rectangle) and the group III antagonist MSOP (100  $\mu$ M, gray triangle) caused no effect compared to control (open circle). Electrical field stimulations (black triangles) evoked reversible and reproducible increase in the fractional release (FR). Drugs were applied in the perfusion solution as the horizontal bar indicates. (C) Bar chart shows the effect of agonists and antagonists of different type of mGluRs on the electrical stimulation-evoked DA release (FRS<sub>2</sub>/FRS<sub>1</sub>). (D) Bar chart shows the effect of agonists and antagonists of different type of mGluRs on the resting DA efflux (FRR<sub>2</sub>/FRR<sub>1</sub>). Data presented are mean  $\pm$  S.E.M.

The question arises that if the dopaminergic terminal does not contain the mGluRs, which other neural element is the primary target of the group II agonist 2*R*,4*R*-APDC. As the polarity of the modulation was opposite to the one we expected from the inhibitory group II mGluR receptor, we assumed that 2*R*,4*R*-APDC suppressed the activity of inhibitory neurons, presumably GABAergic cells, which would act to depress the dopaminergic terminal. To explore the possible involvement of such disinhibitory mechanism by removing the inhibitory step, we blocked GABA<sub>A</sub> receptors by bicuculline at 10  $\mu$ M. In the presence of bicuculline, 100  $\mu$ M 2*R*,4*R*-APDC failed to increase the spontaneous release of DA ( $P=0.028$ , *t*-test, Fig. 2C and D). The GABA<sub>A</sub> antagonist also decreased the DA-releasing effect of 300  $\mu$ M 2*R*,4*R*-APDC but the inhibition was not significant compared with the action of 300  $\mu$ M 2*R*,4*R*-APDC because of the larger error of the release measurement in the case of 300  $\mu$ M 2*R*,4*R*-APDC group ( $P=0.116$ , Fig. 2D).

In these experiments bicuculline was perfused throughout the experiment, which prevented to observe the acute effect

of GABA on the cochlear DA release. We wondered if the released GABA could tonically block the release of DA in the cochlea. To investigate the role of the GABAergic tone on DA release we applied bicuculline at 10  $\mu$ M from the 8th collection period and maintained it for 15 min. Bicuculline significantly increased the resting outflow of DA ( $P=0.040$ , *t*-test, Fig. 3A and B), and also tended to elevate the electrical field stimulation-evoked release of DA but the effect on the evoked release was not significant ( $P=0.365$  *t*-test, Fig. 3C and D).

In the present study, we explored that a GABAergic disinhibitory loop underlies the DA-releasing effect of the activation of group II mGluRs. Our neurochemical data suggest that these GABAergic fibers express functional mGluRs 2/3. Through these mGluRs, the IHC-released Glu is able to decrease the release of GABA, which leads to the disinhibition of DA containing LOC terminals. The facilitated DA release inhibits the activity of the afferent dendrites, which counteracts its IHC-induced activation. Our neurochemical evidence also suggests that the GABA containing LOC terminals have

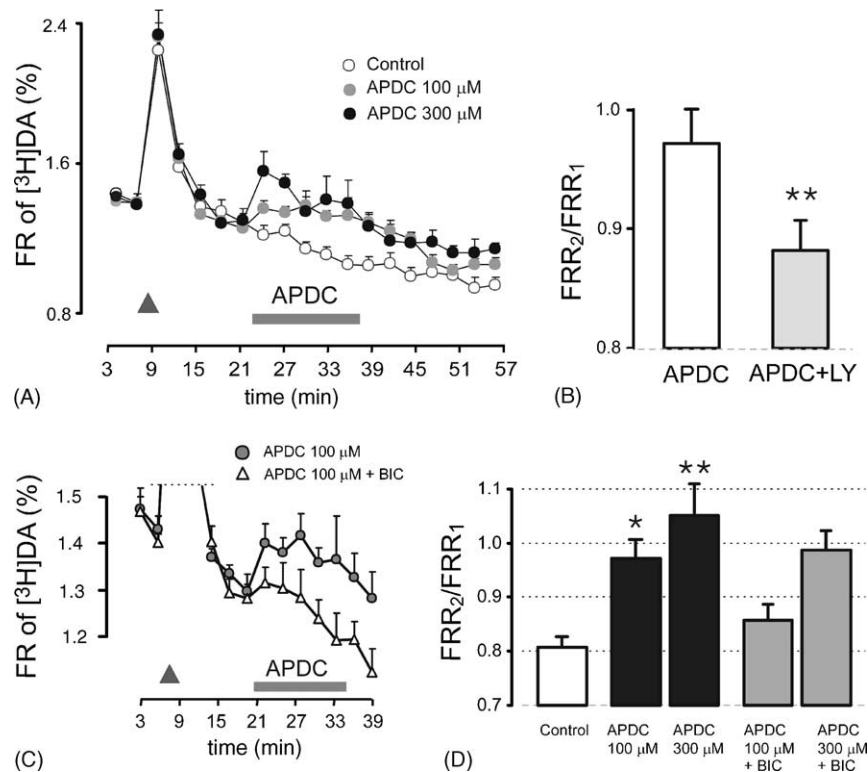


Fig. 2. The mGluR2/3 agonist *2R,4R*-APDC induced a reversible increase in the resting release of DA from the cochlea preparation. (A) *2R,4R*-APDC, applied at 100 μM (gray circle) and 300 μM (black circle) significantly increased the resting release of [<sup>3</sup>H]DA from isolated cochlea vs. control (open circle). *2R,4R*-APDC was applied from the 21st minute and perfused for 9 min as indicated by the horizontal gray line. The single field stimulation was used to control the tissue response and is indicated by the black triangle. (B) 0.2 μM LY-341495 could significantly prevent the DA-releasing effect of 100 μM *2R,4R*-APDC in isolated cochlea as revealed by the FRR<sub>2</sub>/FRR<sub>1</sub> values ( $n=9, 9$ ). Data presented are mean  $\pm$  S.E.M. (\*\* $P < 0.01$ ). (C) The [<sup>3</sup>H]DA-releasing effect of *2R,4R*-APDC applied at 100 μM (gray circle) was prevented by 10 μM bicuculline (empty triangle). *2R,4R*-APDC was applied from the 21st minute and perfused for 9 min as indicated by the horizontal gray line. Bicuculline was present throughout the experiment. The single field stimulation is indicated by the black triangle. (D) Summary bar chart of the effect of *2R,4R*-APDC given at 100 μM and 300 μM. ( $n=9, 8$ ) and in the presence of the GABA<sub>A</sub> antagonist bicuculline (10 μM,  $n=11, 9$ ) on the resting DA release as revealed by the FRR<sub>2</sub>/FRR<sub>1</sub> values. Data presented are mean  $\pm$  S.E.M.; asterisks indicate significant differences ( $n=10$ ; \* $P < 0.05$ , \*\* $P < 0.01$ ).

spontaneous GABA release that keeps the cochlear dopaminergic nerve endings under tonic inhibition. Over the physiological regulating function, the mGluR group II agonist-mediated release of DA may be an important factor during harmful excitotoxic conditions in the cochlea. Neuroprotective role of mGluRs has been described in other regions of the nervous system. The mGluR2/3 agonist, *2R,4R*-APDC could potentiate the locomotor response produced by D1 receptor activation [4]. Moreover, *2R,4R*-APDC, administered intracerebroventricularly, transiently reduced sound-induced seizure activity and inhibited clonic seizures induced by the group III mGluR antagonists in DBA/2 mice [20]. In the light of these literature data, our finding that *2R,4R*-APDC was able to induce the release of DA from the cochlear dopaminergic terminals, corroborates the theory of the protective DA release in the cochlea. The finding, that the group II mGluR agonist *2R,4R*-APDC increased the release of cochlear DA well corresponds to the neuroprotective role of this compound known in wide regions of the central nervous system, and the protective nature of the released DA within the cochlea. In our previous study we found that the release of dopamine cannot

be influenced by iGluR antagonists [14]. However, the released glutamate can interact with the cochlear dopaminergic system through the GABAergic link (Fig. 4). Since glutamate is thought to be important in excitotoxicity during ischemic insults [12], our findings indicate that neuroprotective DA release may be induced by the released glutamate to reduce the consequences of the activation of the ionotropic glutamate receptors.

The electrical field stimulation cause depolarization of all neural elements in the tissue including axon terminals. Therefore, the modulation in the ratio of two consecutive peaks by drugs given between the two stimuli would reflect the activity of presynaptic receptors because release occurs from terminals during field stimulation independently on the upstream activity. The finding that *2R,4R*-APDC failed to influence the release of DA evoked by electrical field stimulation, provides a pharmacological evidence for the lack of group II mGluRs on dopaminergic terminals indicating that the target of the mGluR agonist was elsewhere.

In addition, the polarity of effect on DA release by *2R,4R*-APDC is just the opposite of what we would expect from

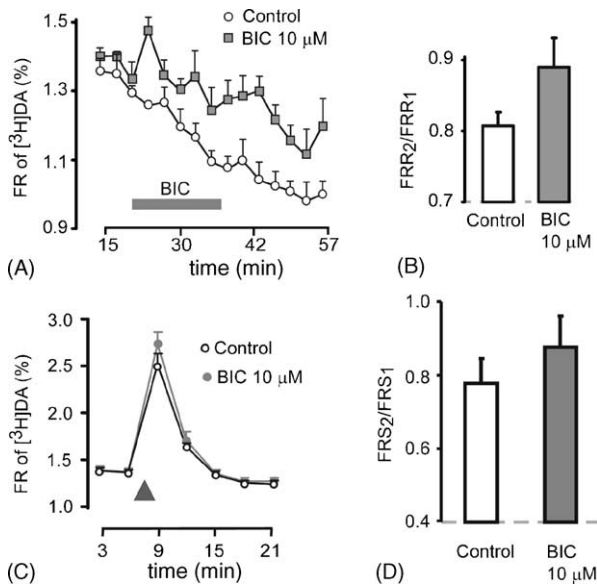


Fig. 3. The effect of the GABA<sub>A</sub> receptor antagonist bicuculline alone indicates a GABAergic tone on the cochlear DA release. (A) Bath application of bicuculline (10 μM) induced [<sup>3</sup>H]DA release from the guinea pig cochlea ( $n=8$ , gray rectangles) vs. control ( $n=9$ , open circle). Bicuculline was applied from the 21st minute and perfused for 9 min as indicated by the horizontal gray line. (B) Summary bar chart shows the effect of bicuculline (10 μM) on the resting outflow of DA ( $FRR_2/FRR_1$ ). Data presented are mean  $\pm$  SEM. (C) The apparent increase in the electrically evoked release of DA by bicuculline (10 μM,  $n=15$ , gray circle) was not significant vs. control ( $n=20$ , open circle). (D) Summary bar chart shows the effect of bicuculline (10 μM) on the field stimulation-evoked outflow of DA ( $FRS_2/FRS_1$ ). Data presented are mean  $\pm$  SEM.

an inhibitory receptor. How the group II mGluR, which is known to inhibit most cell functions, could excite release from the dopaminergic nerve terminals? Our finding that the block of GABA<sub>A</sub> receptors prevented the excitatory action of 2R,4R-APDC on DA release, suggests that most likely the GABAergic lateral olivocochlear fibers contain the mGluRs. The activation of these receptors turns off GABA release, which results in a decreased inhibition on the dopaminergic terminals (Fig. 4). Such disinhibitory mechanism is known in the central nervous system [36]. The modulation of DA release by mGluRs located on GABAergic interneurons is known in the central nervous system [5,9]. Different neurotransmitters are co-localized and released from the axons of the lateral olivocochlear efferents [34], and could diffuse out of the synapse with the inner hair cell and modulate the activity of other cells. It is still controversial which transmitters are localized together, in the same axon terminal [18]. Similarly to GABA, DA can also be released from the lateral olivocochlear fibers. The fact, that the releases of these substances are functionally connected, suggests a cross-talk between the nerve endings, classically described by Vizi [37]. Overall, this study provided functional evidence for the DA-releasing effect of nonsynaptic mGluRs in the cochlea that raises the possibility of potential therapeutic approach against local ischemic damage targeting mGluRs. Our new finding

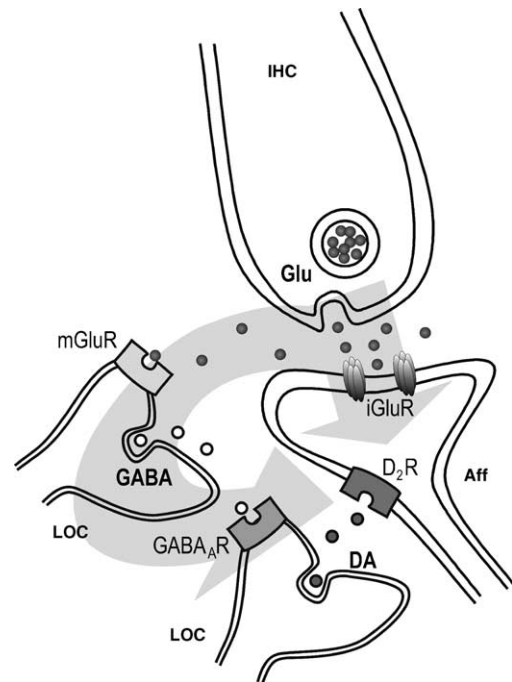


Fig. 4. Schematic drawing of the potential disinhibitory mechanism involving a GABAergic step after activation of group II mGluRs in the cochlea. (IHC: inner hair cell; LOC: lateral olivocochlear fiber; Aff: afferent fiber; Glu: glutamate; DA: dopamine; iGluR: ionotropic glutamate receptor; mGluR: type II metabotropic glutamate receptor; GABA<sub>A</sub>R: GABA<sub>A</sub> receptor; and D<sub>2</sub>R: D<sub>2</sub> dopamine receptor).

suggests a newly described ultra-short feed-back loop: the released glutamate can reduce its own excitatory effect through the lateral olivocochlear efferent system within the cochlea (Fig. 4). The pharmacological enhancement of such feed-back mechanisms could be a target of the future therapy of sensorineural hearing loss.

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## References

- [1] T. Arnold, E. Oestreicher, K. Ehrenberger, D. Felix, GABA(A) receptor modulates the activity of inner hair cell afferents in guinea pig cochlea, *Hear. Res.* 125 (1/2) (1998) 147–153.
- [2] S.R. Bilak, D.K. Morest, Differential expression of the metabotropic glutamate receptor mGluR1alpha by neurons and axons in the cochlear nucleus: in situ hybridization and immunohistochemistry, *Synapse* 28 (4) (1998) 251–270.

- [3] C.G. d'Aldin, J.L. Puel, R. Leducq, O. Crambes, M. Eybalin, R. Pujol, Effects of a dopaminergic agonist in the guinea pig cochlea, *Hear. Res.* 90 (1/2) (1995) 202–211.
- [4] H.N. David, J.H. Abiraini, Differential modulation of the D1-like- and D2-like dopamine receptor-induced locomotor responses by group II metabotropic glutamate receptors in the rat nucleus accumbens, *Neuropharmacology* 41 (4) (2001) 454–463.
- [5] Z. Diaz-Cabiale, M. Vivo, A. Del Arco, W.T. O'Connor, M.K. Harte, C.E. Muller, E. Martinez, P. Popolie, K. Fuxe, S. Ferre, Metabotropic glutamate mGlu5 receptor-mediated modulation of the ventral striopallidal GABA pathway in rats. Interactions with adenosine A2A and dopamine D2 receptors, *Neurosci. Lett.* 324 (2002) 154–158.
- [6] D.G. Drescher, G.E. Green, K.M. Khan, K. Hajela, K.W. Beisel, B.J. Morley, A.K. Gupta, Analysis of gamma-aminobutyric acidA receptor subunits in the mouse cochlea by means of the polymerase chain reaction, *J. Neurochem.* 61 (3) (1993) 1167–1170.
- [7] M. Eybalin, Neurotransmitters and neuromodulators of the mammalian cochlea, *Physiol. Rev.* 73 (2) (1993) 309–373.
- [8] D. Felix, K. Ehrenberger, The efferent modulation of mammalian inner hair cell afferents, *Hear. Res.* 64 (1) (1992) 1–5.
- [9] M.G.P. Feenstra, M.H.A. Botterbiom, J.F.M. van Uum, Local activation of metabotropic glutamate receptors inhibits the handling-induced increased release of dopamine in the nucleus accumbens but not that of dopamine or noradrenaline in the prefrontal cortex: comparison with inhibition of ionotropic receptors, *J. Neurochem.* 70 (1998) 1104–1113.
- [10] A. Gáborján, B. Lendvai, E.S. Vizi, Neurochemical evidence of dopamine release by lateral olivocochlear efferents and its presynaptic modulation in guinea-pig cochlea, *Neuroscience* 90 (1999) 131–138.
- [11] P.E. Gil-Loyzaga, Neurotransmitters of the olivocochlear lateral efferent system: with an emphasis on dopamine, *Acta Otolaryngol.* 115 (2) (1995) 222–226.
- [12] G. Halmos, Z. Doleviczényi, G. Répássy, Á. Kittel, E.S. Vizi, B. Lendvai, T. Zelles, D<sub>2</sub>-Autoreceptor inhibition reveals oxygen-glucose deprivation induced release of dopamine in guinea-pig cochlea, *Neuroscience* 132 (2005) 801–809.
- [13] G. Halmos, A. Gáborján, B. Lendvai, T. Zelles, E.S. Vizi, Interaction between dopaminergic and glutamatergic transmission in isolated guinea pig cochlea, *ARO Abstr.* (2003) 252.
- [14] G. Halmos, A. Gáborján, G. Répássy, L.Z. Szabó, E.S. Vizi, Veratridine-evoked release of dopamine in guinea pig isolated cochlea, *Hear. Res.* 144 (1–2) (2000) 89–96.
- [15] G. Halmos, B. Lendvai, A. Gáborján, M. Baranyi, L.Z. Szabó, V.L. Csokonai, Simultaneous measurement of glutamate and dopamine release from isolated guinea pig cochlea, *Neurochem. Int.* 40 (4) (2002) 243–248.
- [16] S. Kleinlogel, E. Oestreicher, T. Arnold, K. Ehrenberger, D. Felix, Metabotropic glutamate receptors group I are involved in cochlear neurotransmission, *Neuroreport* 10 (1999) 1879–1882.
- [17] X. Lin, S. Chen, P. Chen, Activation of metabotropic GABAB receptors inhibited glutamate responses in spiral ganglion neurons of mice, *Neuroreport* 11 (5) (2000) 957–961.
- [18] S.F. Maison, J.C. Adams, M.C. Liberman, Olivocochlear innervation in the mouse: immunocytochemical maps, crossed versus uncrossed contributions, and transmitter colocalization, *J. Comp. Neurol.* 455 (3) (2003) 406–416.
- [19] B. Malgrange, J.M. Rigo, P.P. Lefebvre, P. Coucke, F. Goffin, G. Xhaufflaire, S. Belachew, T.R. Van De Water, G. Moonen, Diazepam-insensitive GABAA receptors on postnatal spiral ganglion neurones in culture, *Neuroreport* 8 (1997) 591–596.
- [20] R.X. Moldrich, A. Talebi, P.M. Beart, A.G. Chapman, B.S. Meldrum, The mGlu(2/3) agonist 2R,4R-4-aminopyrrolidine-2,4-dicarboxylate, is anti- and proconvulsant in DBA/2 mice, *Neurosci. Lett.* 299 (1/2) (2001) 125–129.
- [21] F. Nicoletti, V. Bruno, A. Copani, G. Casabona, T. Knopfel, Metabotropic glutamate receptors: a new target for the therapy of neurodegenerative disorders? *Trends Neurosci.* 19 (7) (1996) 267–271.
- [22] A.S. Niedzielski, S. Safieddine, R.J. Wenthold, Molecular analysis of excitatory amino acid receptor expression in the cochlea, *Audiol. Neurootol.* 2 (1/2) (1997) 79–91.
- [23] A.S. Niedzielski, R.J. Wenthold, Expression of AMPA, kainate, and NMDA receptor subunits in cochlear and vestibular ganglia, *J. Neurosci.* 75 (3) (1995) 2338–2353.
- [24] E. Oestreicher, W. Arnold, K. Ehrenberger, D. Felix, Dopamine regulates the glutamatergic inner hair cell activity in guinea pigs, *Hear. Res.* 107 (1/2) (1997) 46–52.
- [25] J.L. Puel, Chemical synaptic transmission in the cochlea, *Prog. Neurobiol.* 47 (6) (1995) 449–476.
- [26] J.L. Puel, J. Ruel, C.G. d'Aldin, R. Pujol, Excitotoxicity and repair of cochlear synapses after noise-trauma induced hearing loss, *Neuroreport* 9 (9) (1998) 2109–2114.
- [27] R. Pujol, Lateral and medial efferents: a double neurochemical mechanism to protect and regulate inner and outer hair cell function in the cochlea, *Br. J. Audiol.* 28 (4/5) (1994) 185–191.
- [28] R. Pujol, J.L. Puel, Excitotoxicity, synaptic repair, and functional recovery in the mammalian cochlea: a review of recent findings, *Ann. N.Y. Acad. Sci.* 884 (1999) 249–254.
- [29] R. Pujol, J.L. Puel, C.G. d'Aldin, M. Eybalin, Pathophysiology of the glutamatergic synapses in the cochlea, *Acta Oto-Laryngol.* 113 (1993) 330–334.
- [30] R. Pujol, J.L. Puel, M. Eybalin, Implication of non-NMDA and NMDA receptors in cochlear ischemia, *Neuroreport* 3 (4) (1992) 299–302.
- [31] J. Ruel, R. Nouvian, G.C. d'Aldin, R. Pujol, M. Eybalin, J.L. Puel, Dopamine inhibition of auditory nerve activity in the adult mammalian cochlea, *Eur. J. Neurosci.* 14 (6) (2001) 977–986.
- [32] S. Safieddine, M. Eybalin, Co-expression of NMDA and AMPA/kainate receptors mRNAs in cochlear neurons, *Neuroreport* 3 (1992) 1145–1148.
- [33] S. Safieddine, M. Eybalin, Expression of mGluR1 alpha mRNA receptor in rat and guinea pig cochlear neurons, *Neuroreport* 7 (1) (1995) 193–196.
- [34] S. Safieddine, A.M. Prior, M. Eybalin, Choline acetyltransferase, glutamate decarboxylase tyrosine hydroxylase, calcitonin gene-related peptide and opioid peptides coexist in lateral efferent neurons of rat and guinea-pig, *Eur. J. Neurosci.* 9 (1997) 356–367.
- [35] W. Sun, R.J. Salvi, Dopamine modulates sodium currents in cochlear spiral ganglion neurons, *Neuroreport* 12 (4) (2001) 803–807.
- [36] K. Toth, T.F. Freund, R. Miles, Disinhibition of rat hippocampal pyramidal cells by GABAergic afferents from the septum, *J. Physiol.* 500 (2) (1997) 463–474.
- [37] E.S. Vizi, Role of high-affinity receptors and membrane transporters in nonsynaptic communication and drug action in the central nervous system, *Pharmacol. Rev.* 52 (1) (2000) 63–89.
- [38] E.S. Vizi, L.G. Harsing Jr., I. Zimanvi, G. Gaal, Release and turnover of noradrenaline in isolated median eminence: lack of negative feedback modulation, *Neuroscience* 16 (4) (1985) 907–916.
- [39] Y. Yamamoto, A. Matsubara, K. Ishii, K. Makinae, A. Sasaki, H. Shinkawa, Localization of gamma-aminobutyric acid A receptor subunits in the rat spiral ganglion and organ of Corti, *Acta Otolaryngol.* 122 (7) (2002) 709–714.