The impact of DA D2 heteroreceptor complexes and their receptor-receptor interactions on Parkinson’s disease and its treatment. Focus on the A2A-D2-mGlu5 and NTS1-D2 heterocomplexes

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Heteroreceptor complexes formed between mGluR5, D2R and A2AR

Signaling

A2AR-mGluR5 synergize to reduce D2R recognition and Gi/o coupling and signaling Popoli et al.2001,Fuxe et al.2003,Agnati et al.2003, Interactions also at the level of the signaling cascades: MAPK and CREB-P. A2AR and mGluR5 agonists, NAMs synergistically increase GABA release in ventral pallidum Diaz-Cabiale et al.2002

Potential relevance PD

mGluR5 antagonists and negative allosteric modulators may significantly target the mGluR5 protomer. They increase locomotion and exert antiparkinsonian actions and antidyskinetic actions especially combined with A2A antagonists. Coccurello et al.2004,Kachroo et al. 2005 Schwarzschild et al.2006,Vallano et al.2013

Neuronal $A_{2A}$ and mGlu5 immunoreactivity

Extracellular GABA (%)

Ambulations

Schwarzschild, Agnati, Fuxe, Morelli 2006

Cabello et al. 2009
Adenosine and extrasynaptic glutamate as important VT signals
striatal local circuit of the striato-pallidal GABA neurons identified through immunoreactivity of an $A_{2A}$R interacting protein NECAB2

Ciruela et al. 2011
Conclusions

Integration of synaptic and volume transmission strongly involves receptor-receptor interactions in heteroreceptor complexes in the plasma membrane.

Heteroreceptor complexes represent a new fundamental principle in molecular medicine for integration of transmitter signals enabling diversity and bias of the receptor protomers and novel strategies for treatment of Parkinson’s disease. DA receptor agonists can target many D1 and D2 heteroreceptor complexes.

Proposal: Heterobivalent compounds with A2A and mGluR5 antagonist/NAM pharmacophors and/or small molecules with combined A2A and MGlur5 antagonist activity to be explored as novel antiparkinsonian drugs for early and chronic treatment of PD.

They can remove the brake on D2R signaling that develops with disease Progression, enhanced by chronic L-DOPA and D2 agonist treatment.

A2A and mGlu5 antagonists can target many A2A and mGlu5 heteroreceptor complexes, respectively.
Adenosine A2A receptor containing heteroreceptor complexes

1. The adenosine A1 and A2A heteroreceptor complex and the control of glutamate release in the central nervous system (Ciruela et al. 2006)


5. A2AR and cannabinoid CB1 receptor (CB1R) heteroreceptor complexes including A2A-CB1-D2 higher order heteroreceptor complexes (Carriba et al. 2007, 2008; Marcellino et al. 2008)
Neurotensin reduces the affinity of D-2 dopamine receptors in rat striatal membranes

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Evidence for a substrate of neuronal plasticity based on pre- and postsynaptic neurotensin–dopamine receptor interactions in the neostriatum

(microdialysis/D2 receptors/dopamine release/γ-aminobutyric acid release)

Kjell Fuxe†, William T. O’Connor‡, Tiziana Antonelli‡, Peter G. Osborne‡, Sergio Tanganelli*, Luigi F. Agnati§, and Urban Ungerstedt‡
Neurotensin reduces the affinity of D-2 dopamine receptors in rat striatal membranes

$K_d = 98 \text{ pM}, \text{ control}$

$K_d = 149 \text{ pM}, \text{ NT 10nM}$

Agnati, Fuxe et al. 1983, Von Euler and Fuxe 1987
Neurotensin counteracts apomorphine-induced inhibition of dopamine release as studied by microdialysis in rat neostriatum

S. Tanganelli¹, G. von Euler¹, K. Fuxe¹, L.F. Agnati² and U. Ungerstedt³
Intramembrane Interactions between Neurotensin Receptors and Dopamine D₂ Receptors as a Major Mechanism for the Neuroleptic-like Action of Neurotensin


NT receptor antagonists

SR48692

SR142948A
Effects of intrastriatal perfusion of NT alone, pergolide with or without NT, or the NT antagonist SR48692 on striatal DA release in the awake rat.
Effects of NT (10 nM) and the NT antagonist SR48692 (100 nM) and in combination on D₂ receptor binding in competitive-inhibition experiments with [¹²⁵I]-iodosulpiride vs DA in striatal sections.
BRET² studies on D2R and NTS1R heteromerization in HEK293T cells

Borroto-Escuela et al. 2013
Agonist-induced D2LR and NTS1R receptor activation in a forskolin-induced CRE-luciferase reporter gene assay.

SRE-luciferase reporter gene response after agonist-induced D2LR and NTS1R receptor activation

Borroto-Escuela et al. 2013
Three-dimensional molecular models of the D2LR and NTSR1 were built by means of the homology modeling program Accelrys Discovery Studio 2.5 (San Diego).

Schematic cross-talk signalling pathways of D2R and NTS1R. The D2R receptor is a GPCR which primarily produces cAMP inhibition via Gi/o proteins (A) but also PLC/PKC activation via beta/gamma G-protein subunits (C). In addition, G protein-coupled receptors couple to the MAPK pathway with pERK activation via both G proteins and β-arrestin recruitment (B). The latter often leads to desensitization and internalization of the GPCR. The NTS1R is a GPCR which primarily activates PLCb via Gq/11 proteins and thus also leads to pERK activation via PKC (C).
Evidence for the existence of D2R-NTS1R heteroreceptor complexes in the CPU, AcbC and AcbSh region of the rat brain

Unpublished work in collaboration with Thorsten Schäfer and Kristina Friedland
These pictures have been taken by Borroto-Escuela 2015
Schematic representation of the antagonistic pre- and postjunctional NTS1R and D2R interactions in **dorsal striatum** taking place in NTS1R/D2 heterodimers in balance with excitatory NTS1R homodimers. Neuronal plasticity: The NTS1R-D2 heteroreceptor complex switches the balance towards D1 vs D2 transmission.

Fuxe et al. 1992; Antonelli et al. 2007
Neurotensin receptor mechanisms and its modulation of glutamate transmission in the brain
Relevance for neurodegenerative diseases and their treatment

Effect of intrastriatal perfusion with NMDA alone or in combination with neurotensin (NT) or NT plus its receptor antagonist SR48692 on extracellular GABA levels from the ipsilateral globus pallidus of control (i.e. sham-operated) (A, C) and 6-OHDA lesioned (B, D) awake rats.

Neurotensin increases endogenous glutamate release in the neostriatum of the awake rat.
Compensatory activation of NT release from nigral NT terminals in response to onset of Parkinson’s disease. Increased dopamine (DA) cell firing by, e.g. activation of inhibitory NTS1/D2 autoreceptor interactions and of facilitatory NTS1/NMDA (receptor–receptor and/or cytoplasmic) interactions on glutamate (Glu) terminals and nigral and VTA DA cells.
NTS1 containing dopamine (DA) and glutamate (GLU) nerve terminals in local circuits of the striato-pallidal GABA neurons showing the DA, GLU and neurotensin (NT) signalling in the **physiological state**. One DA and one glutamate synapse are indicated. Low NT tone and baseline release of DA and glutamate are indicated. The low density of NTS1 in the striato-pallidal GABA neurons is not indicated. VT, volume transmission.

(Panel B) In **Parkinson's disease**, loss of DA terminals followed by reduced D2 activity leads to an increased synthesis and release of NT from the dendrites and dendritic spines involving increased glutamate release and increased adenosine A2A signalling due to reduced D2 inhibition of A2A signalling.

Panel C: Treatment with **D2 receptor antagonists**.

Antonelli et al. 2007
Simplified block diagram of basal ganglia–thalamocortical neuronal circuitry, showing the indirect GABA pathways and illustrating NTS1 receptor activation and its effects on striatopallidal transmission through receptor–receptor interactions with striatal D2 and NMDA receptors in intact (A) and unilaterally 6-OHDA-lesioned rats (B).

The effects of NTS1 antagonists on the signalling of the NTS1 containing heteromers and associated changes in the activity of the striatopallidal GABA neurons in 6-OHDA-lesioned rats are also shown (C).

Ferraro et al. 2012
NT peptides and neurodegeneration

- NT : amplification of glutamate-induced neurotoxicity in mesencephalic dopamine and cortical neurons.

- Mechanisms : Enhancing NTS1-NMDA interactions and antagonistic NTS1/D2 R interactions in the cortico-striatal glutamate terminals, the nigral DA cell bodies and dendrites.

- Potential increases in NT levels in the basal ganglia in Parkinson’s disease lead to the NTS1 enhancement of NTS1-NMDA receptor signaling and reduction of D2 autoreceptor signalling in the nigral DA cells may contribute to their neurodegeneration in PD.

- The use of selective NTS1 antagonists together with conventional drug treatments could provide a novel therapeutic approach for symptomatic treatment of Parkinson’s disease with potential neuroprotective actions.
Novel targets for antiparkinsonian drugs

- **A2A-D2 heteroreceptor complexes** with antagonistic A2A-D2 receptor interactions (mainly striato-pallidal GABA neurons)

- **A2A-D2-mGlu5 heteroreceptor complexes** where A2A and mGlu5 synergize to inhibit D2 receptor signaling (mainly striato-pallidal GABA neurons)

- **NTS1-D2 heteroreceptor complexes** with antagonistic NTS1-D2 receptor-receptor interactions (mainly cortico-striatal glutamate terminals, nigro-striatal DA neurons)

- **Putative NTS1-NMDA heteroreceptor complexes** with NTS1 enhancing NMDA receptor signaling (mainly cortico-striatal glutamate terminals, nigral DA nerve cells)

**Combined targeting of different pathways in motor circuits can be of special value**

Novel molecules: heterobivalent compounds; multitargeting compounds. A2A antagonists/mGlu5 antagonists/NTS1 antagonists targeting distinct D2 heteroreceptor complexes?