Дизайн новых модуляторов ионотропных глутаматных рецепторов

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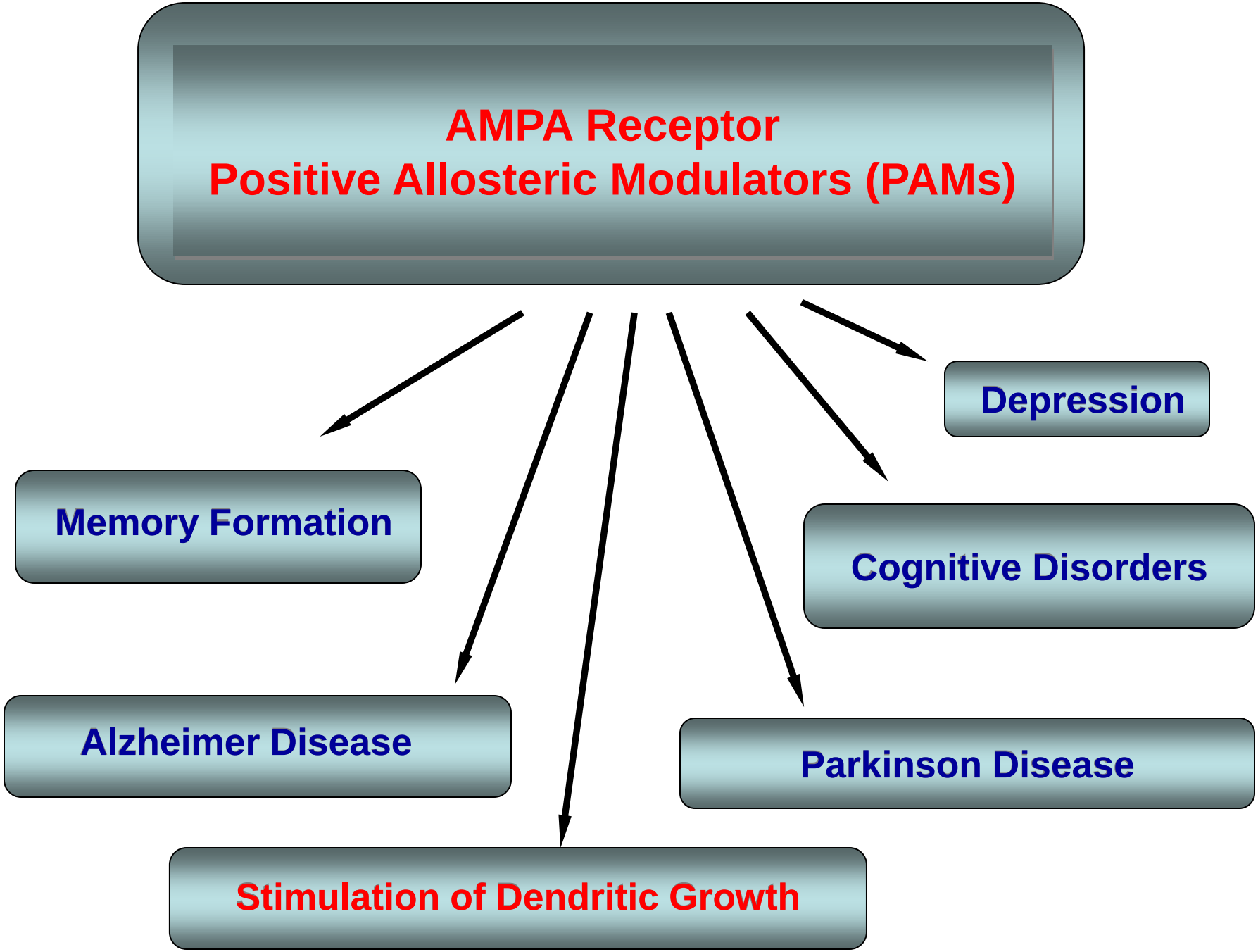
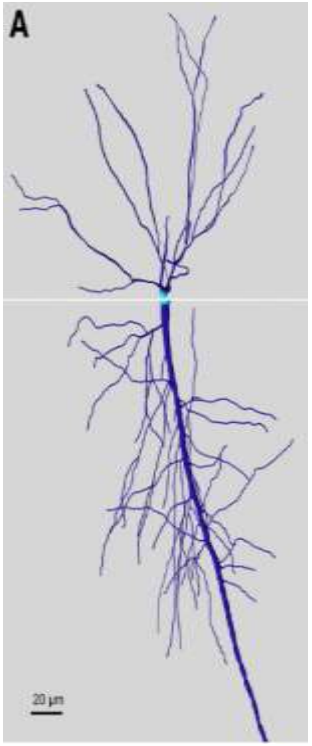
Химический факультет МГУ имени М.В.Ломоносова

Институт физиологически активных веществ РАН, Черноголовка


AMPA Receptor
Positive Allosteric Modulators (PAMs)
Depression
Memory Formation
Cognitive Disorders
Alzheimer Disease
Parkinson Disease
Stimulation of Dendritic Growth

AMPA Receptor
Positive Allosteric Modulators (PAMs)



Significance Statement

Brain aging is characterized by a progressive loss of dendritic arbors and the emergence of impairments to learning-related synaptic plasticity. The present studies show that dendritic losses are evident by middle age despite housing in an enriched environment and can be mostly reversed by long-term, oral administration of a positive allosteric modulator of AMPA-type glutamate receptors. Dendritic recovery was accompanied by improvements to both synaptic plasticity and the encoding of long-term memory of a novel, complex environment. Because the short half-life compound had no evident negative effects, the results suggest a plausible strategy for treating age-related neuronal deterioration.

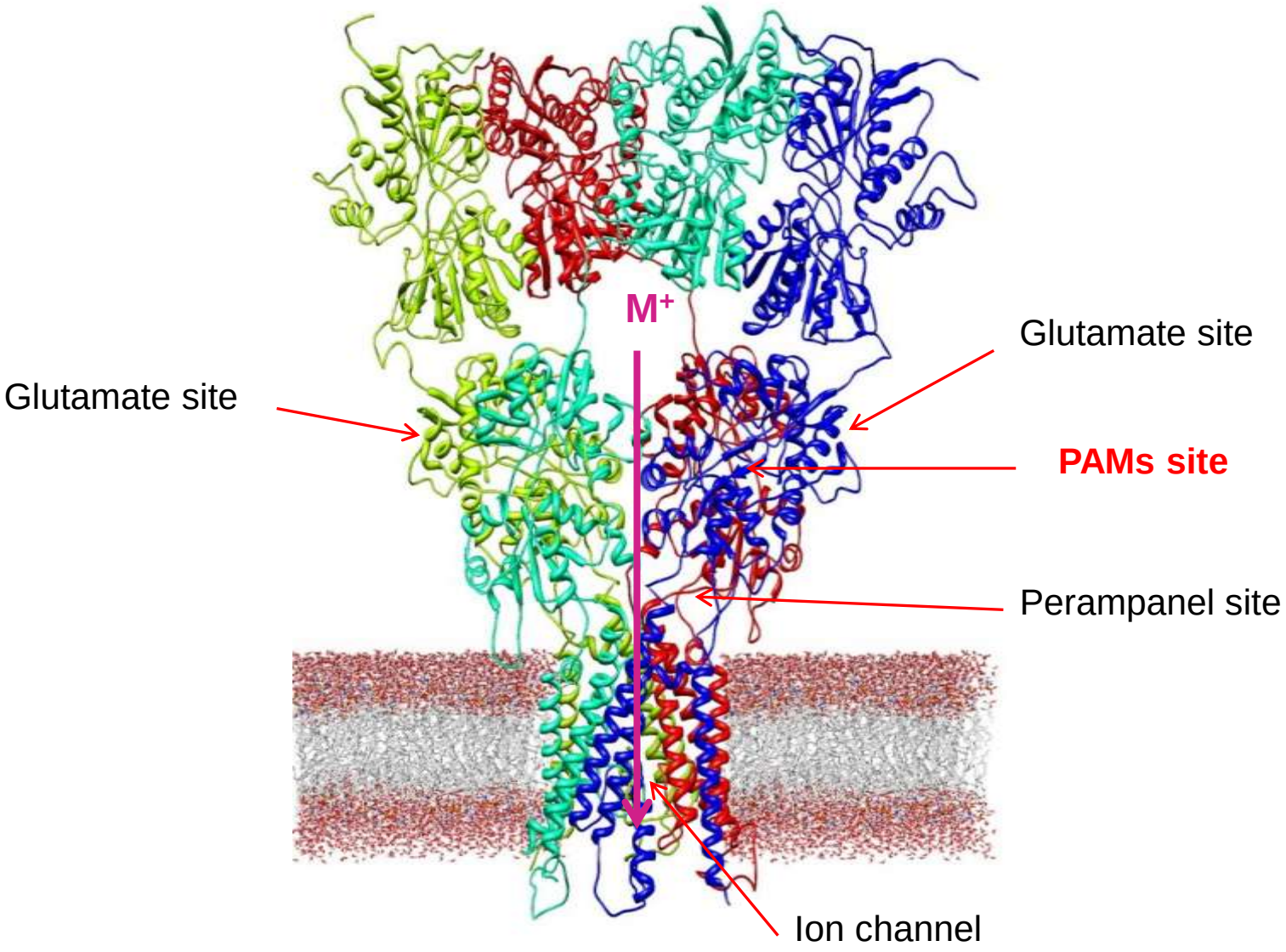
Cited from:

The Journal of Neuroscience, February 3, 2016 • 36(5):1636–1646

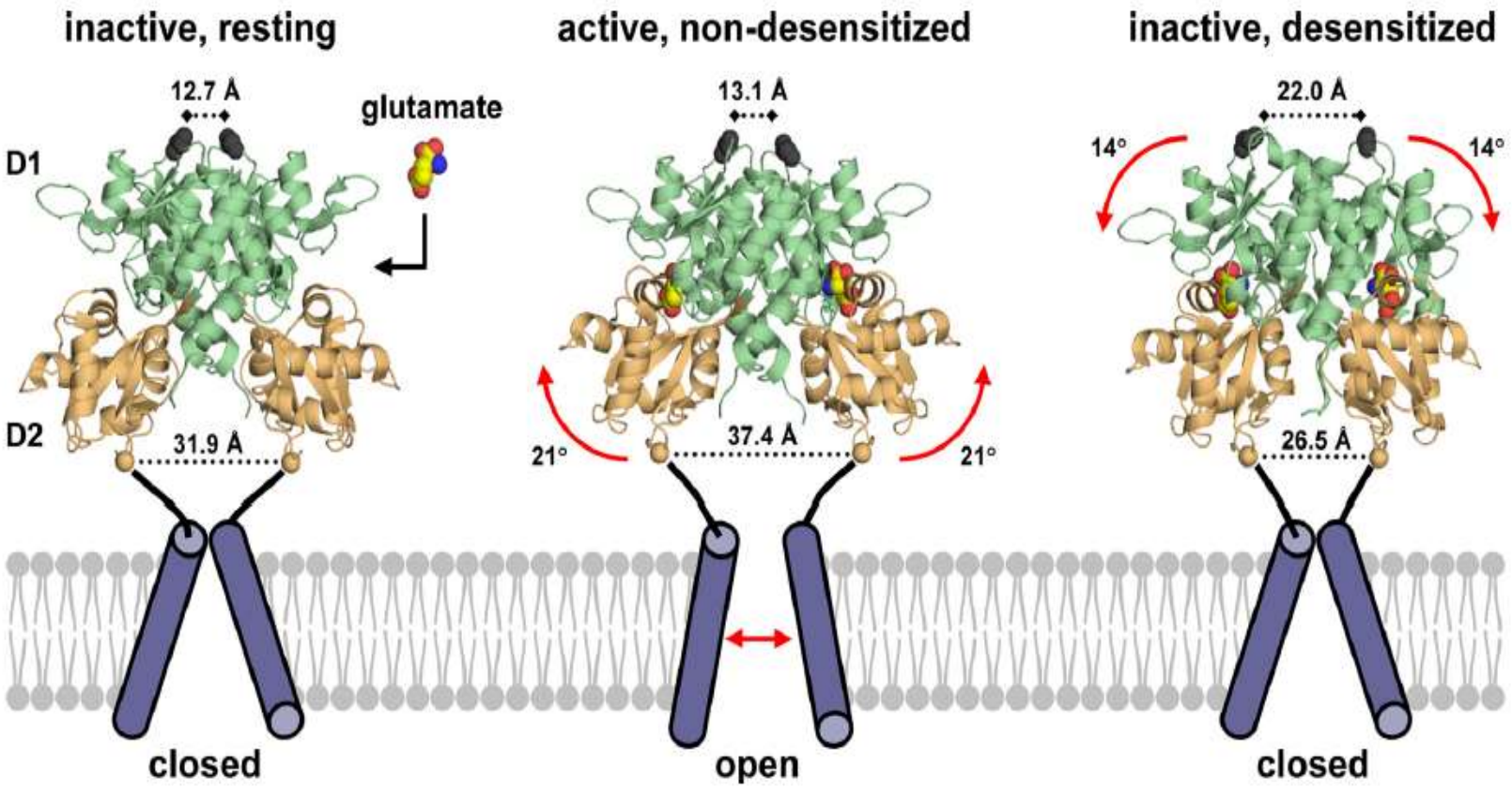
Chronic Ampakine Treatments Stimulate Dendritic Growth and Promote Learning in Middle-Aged Rats

Julie C. Lauterborn,[1*] Linda C. Palmer,[1*] Yousheng Jia,[1] Danielle T. Pham,[1] Bowen Hou,[1] Weisheng Wang,[1] Brian H. Trieu,[1] Conor D. Cox,[1] Svetlana Kantorovich,[1] Christine M. Gall,[1,2*] and Gary Lynch[1,3*]

General Structure of AMPA Receptor and Major Ligand-Binding Sites

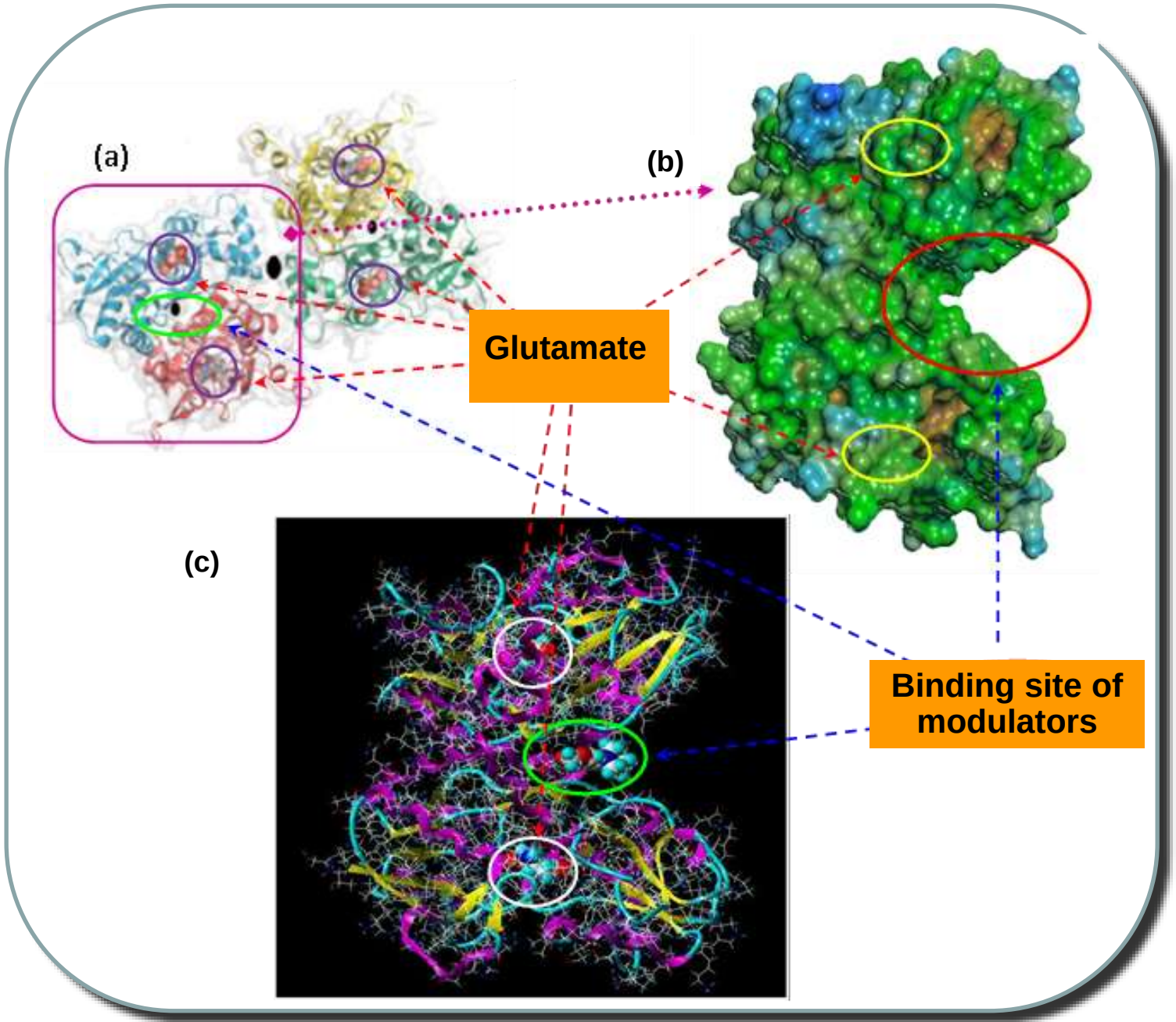


Conformational Changes in the Functioning of AMPA receptor

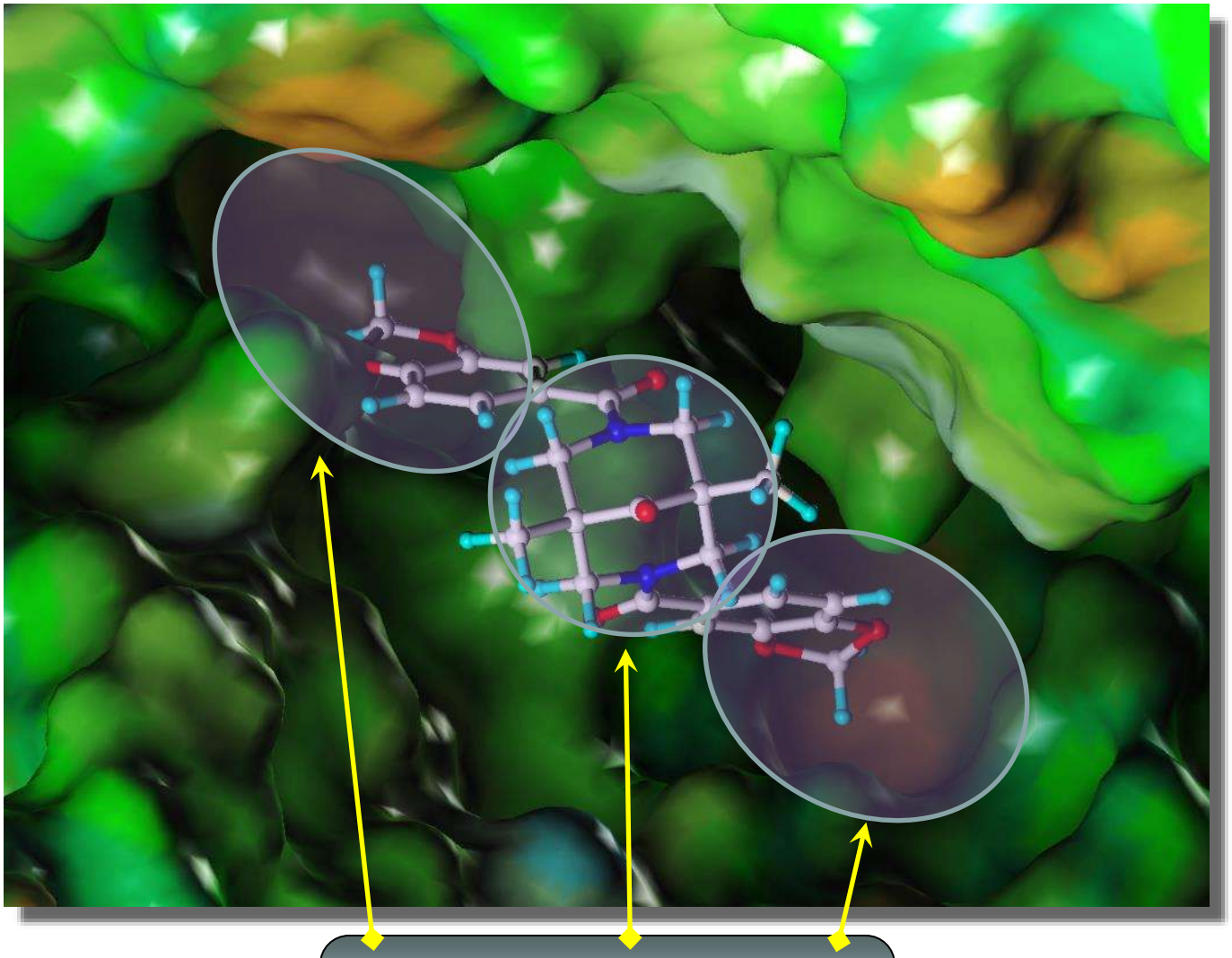


Stephen F. Traynelis et al., *Pharmacol. Rev.* 2010*, 62,* 405–496*.*

Binding Site of Positive AMPA Receptor Modulators

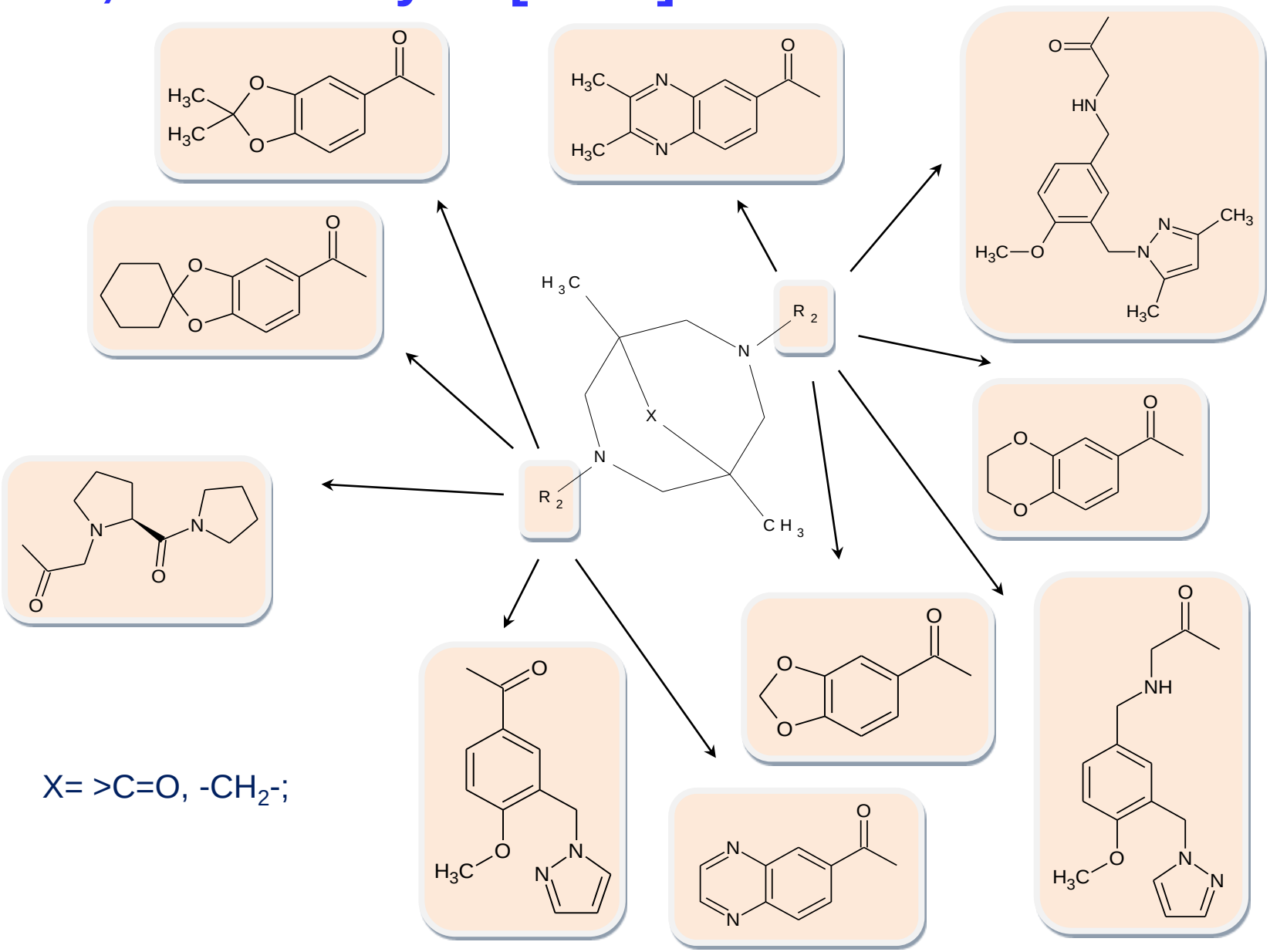


Binding of the bivalent ligand

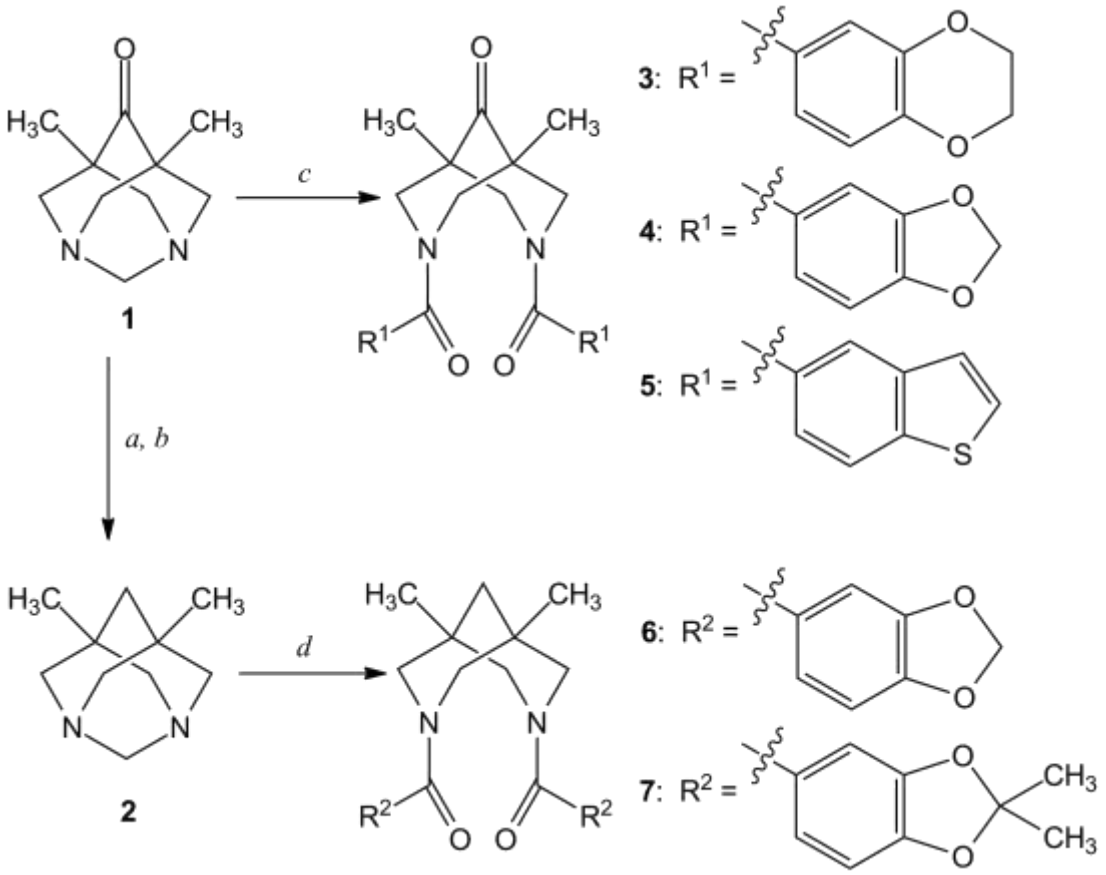


Two active fragments and the linker (spacer)

Selected for synthesis and synthesized 3,7-diazabicyclo[3.3.1]nonane derivatives



Selected for synthesis and synthesized 3,7-diazabicyclo[3.3.1]nonane derivatives



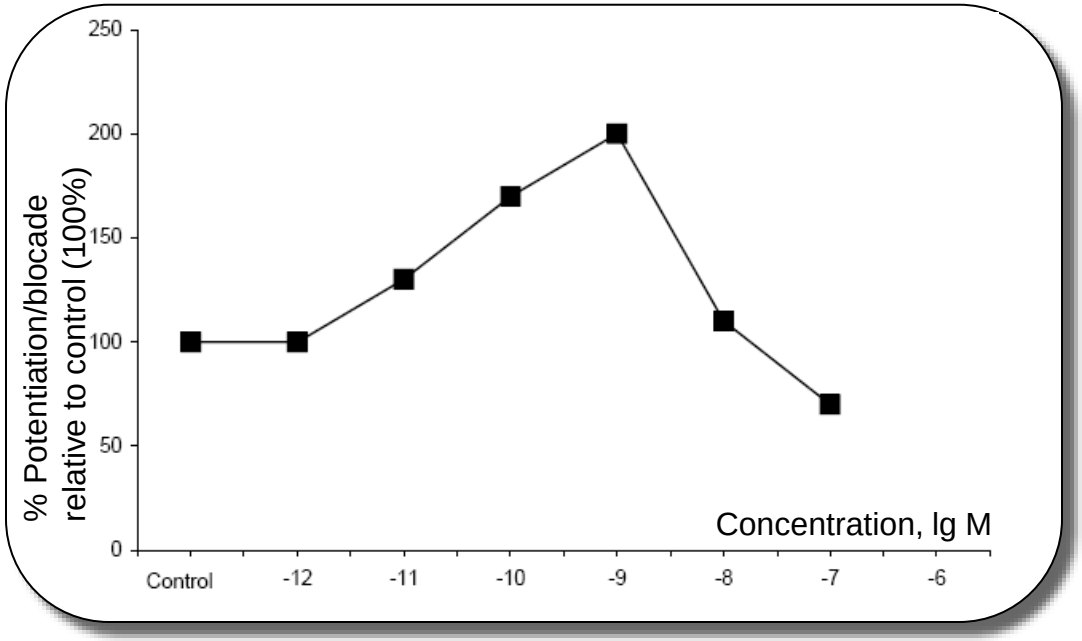
Reaction conditions: a) N_2H_4*H_2O; b) *t*-BuOK/PhMe*; c)* R^1COCl, CH_3CN/K_2CO_3, d) R_2COCl, CH_3CN/K_2CO_3.

M.I. Lavrov, D.S. Karlov, T.A. Voronina, V.V. Grigoriev, A.A. Ustyugov, S.O. Bachurin, V.A. Palyulin, *Mol. Neurobiol.*, **2020**, *57*, 191-199.

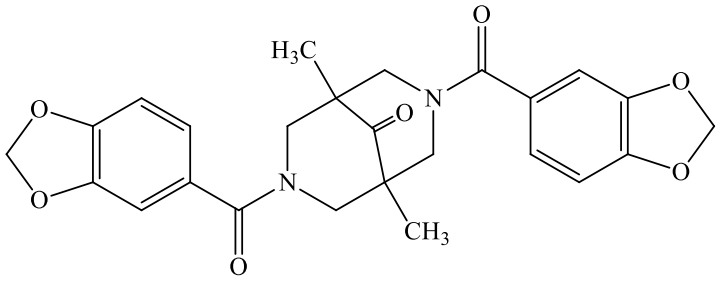
Physiological studies of new AMPA receptor modulators

Experimental biological studies have demonstrated extraordinarily high activity: 1000-10000 times higher than all known monovalent PAMs

Electrophysiological experiments



OSPL-502 – influence on kainate-induced currents in rat Purkinje neurons

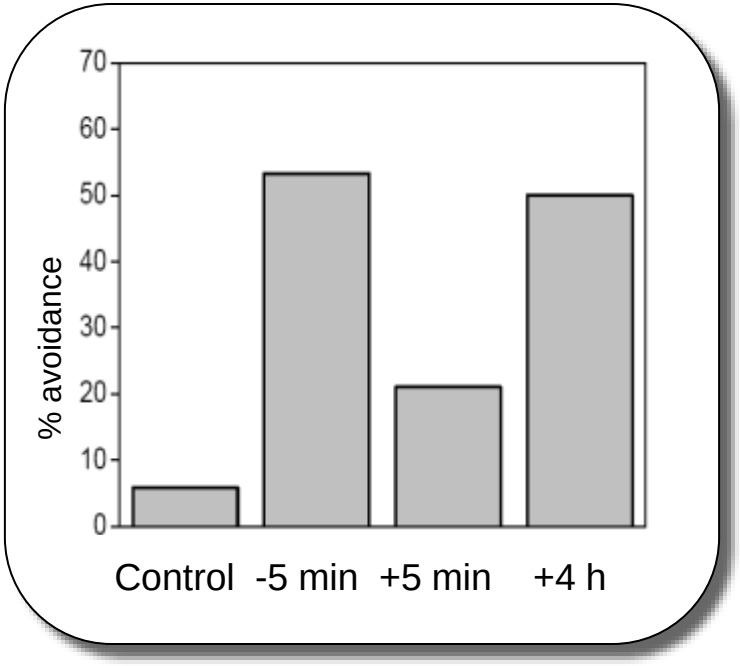


LD_{50} of the best studied bivalent ligands was close to 5000 mg/kg.

M.I.Lavrov, V.V.Grigoriev, S.O.Bachurin, V.A.Palyulin, N.S.Zefirov, *Dokl. Biochem. Biophys*. **2015**, *464*, 322-324;

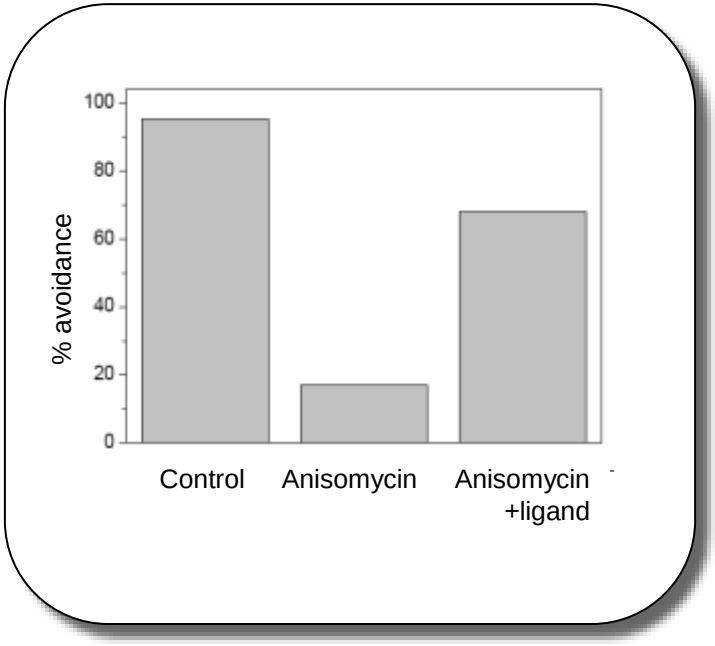
M.I.Lavrov, D.S.Karlov, T.A.Voronina, V.V.Grigoriev, A.A.Ustyugov, S.O.Bachurin, V.A.Palyulin, *Mol. Neurosci.*, **2020**, 57, 191-199.

Passive avoidance model



0.005 mg/kg of compound OSPL-502 in 5 min before training, 5 min or 4 hrs after training to reproduce the results of training after 24 hrs

Restoration of pharmacologically damaged memory



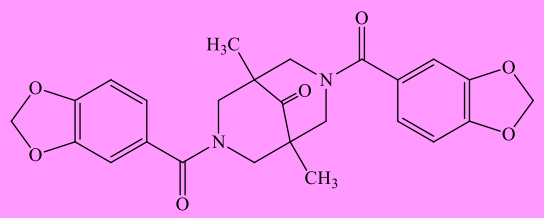
OSPL-502 prevents amnesia caused by the blockade of protein synthesis in brain (24 h, 0,005 mg/kg).

Pre-clinical studies of a novel cognition enhancer

Bio-availability: 44,4 %

Scaling up synthesis: from 50 mg up to 400 g

Sub-chronical toxicity (1 month) : 180 mg/kg



OSPL-502

Acute toxicity: >5000 mg/kg

Chronical toxicity: 675 times higher than the dose recommended for human being

Chronical toxicity (6 months) of a drug form: no any side effects

Drug form

Scopolamine test: Cure of disorders in 0.1 mg/kg dose

PAMs 2D QSAR Study: Molecular Field Topology Analysis (MFTA)

Structure-activity models based on local molecular parameters (atom properties)

Molecular supergraph

- Common frame of reference for different structures

Local descriptors

- *Q* – atomic charge
- *Re* – effective group van der Waals radius
- *Lg* – group lipophilicity
- H_d, H_a – hydrogen bond donor/acceptor ability

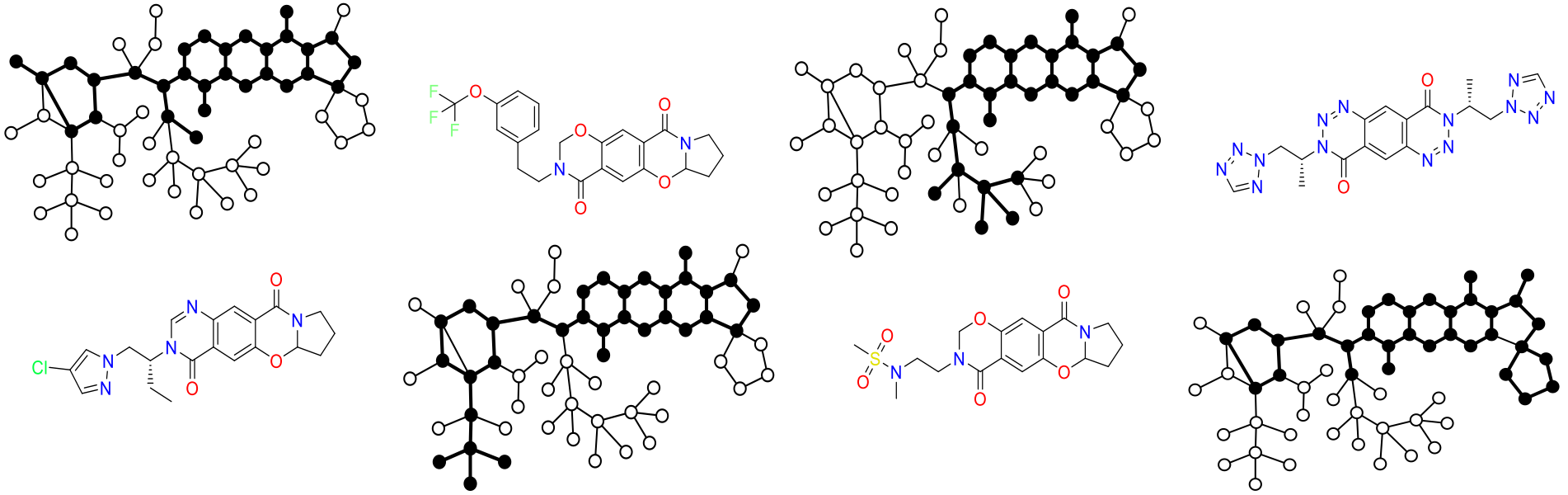
Partial least squares regression (PLSR) modeling

Radchenko E.V., Palyulin V.A., Zefirov N.S., in *Chemoinformatics Approaches to Virtual Screening*, RSC, **2008**, 150-181.
Palyulin V.A., Radchenko E.V., Zefirov N.S., *J. Chem. Inf. Comp. Sci.*, **2000**, *40(3)*, 659-667.

Positive AMPA receptor modulators. Polycyclic benzamide derivatives

Training set: 111 compounds (consistent stereochemistry, compatible scaffolds, reliable activity measurements)
Endpoint: $pEC_{2x} = \log(1 / EC_{2x})$

MFTA molecular supergraph and mapping examples



Mueller R. et al., *Bioorg. Med. Chem. Lett.*, **2011**, *21*, 3923, 3927, 6170, 7455.

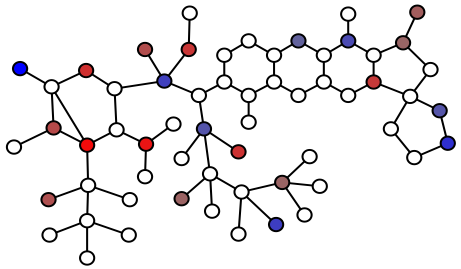
MFTA model of PAM activity

Descriptors	N_F	R^2	*RMSE*	Q^2	*RMSEcv*
Q, R_e, L_g, H_a	5	0.83	0.43	0.55	0.70

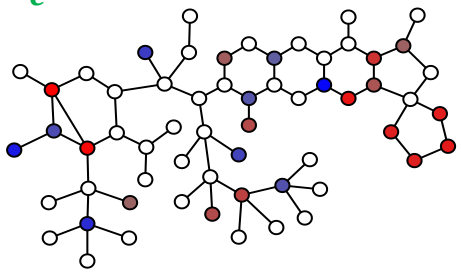
$\boldsymbol{N_F}$ = number of factors in the PLSR model, $\boldsymbol{R^2}$ = squared correlation coefficient, ***RMSE*** = root-mean-square error,
$\boldsymbol{Q^2}$ = cross-validation parameter, ***RMSEcv*** = root-mean-square error of cross validation

MFTA activity maps

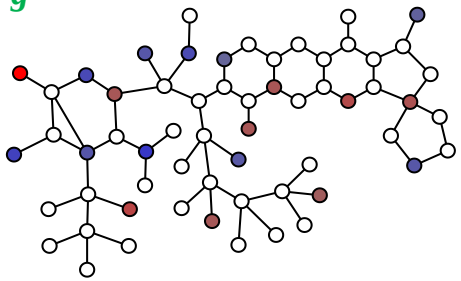
Q



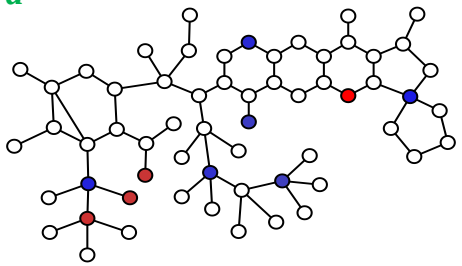
R_e



L_g



H_a

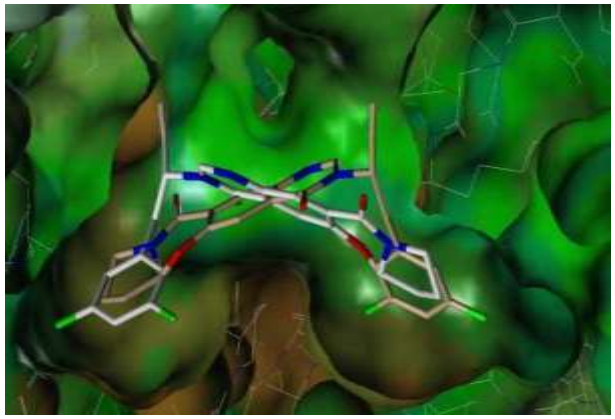


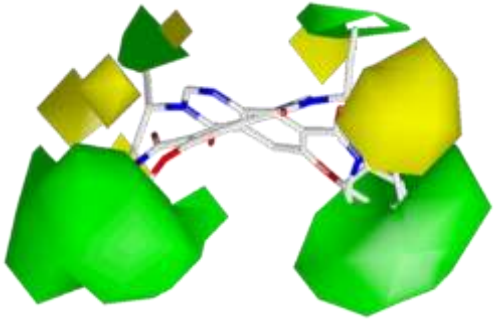
Preference for aryl or hetaryl moieties with polar hydrogen bond acceptor substituents, as well as for moderately polar and/or lipophilic open-chain substituents

E.V.Radchenko, D.S.Karlov, M.I.Lavrov, V.A.Palyulin, *Mendeleev Commun.* **2017**, *27*, 623-625.

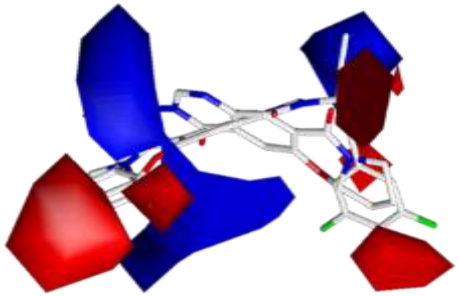
Red: activity increases **Blue: activity decreases**

PAMs Docking-Based 3D Alignment, 3D QSAR (CoMFA) and Pharmacophore



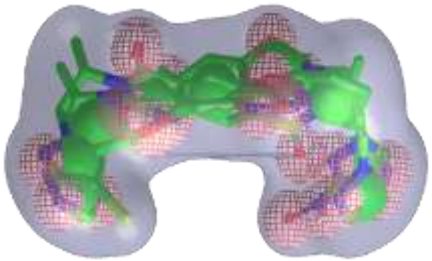


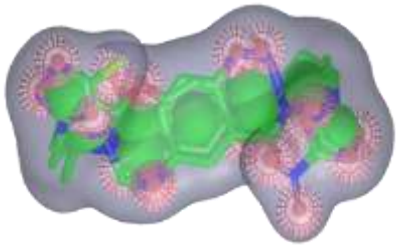
Steric Fields



Electrostatic Fields

CoMFA model:
n = 49, N_f = 4, R^2 = 0.75,
$RMSE$ = 0.47, Q^2 = 0.56

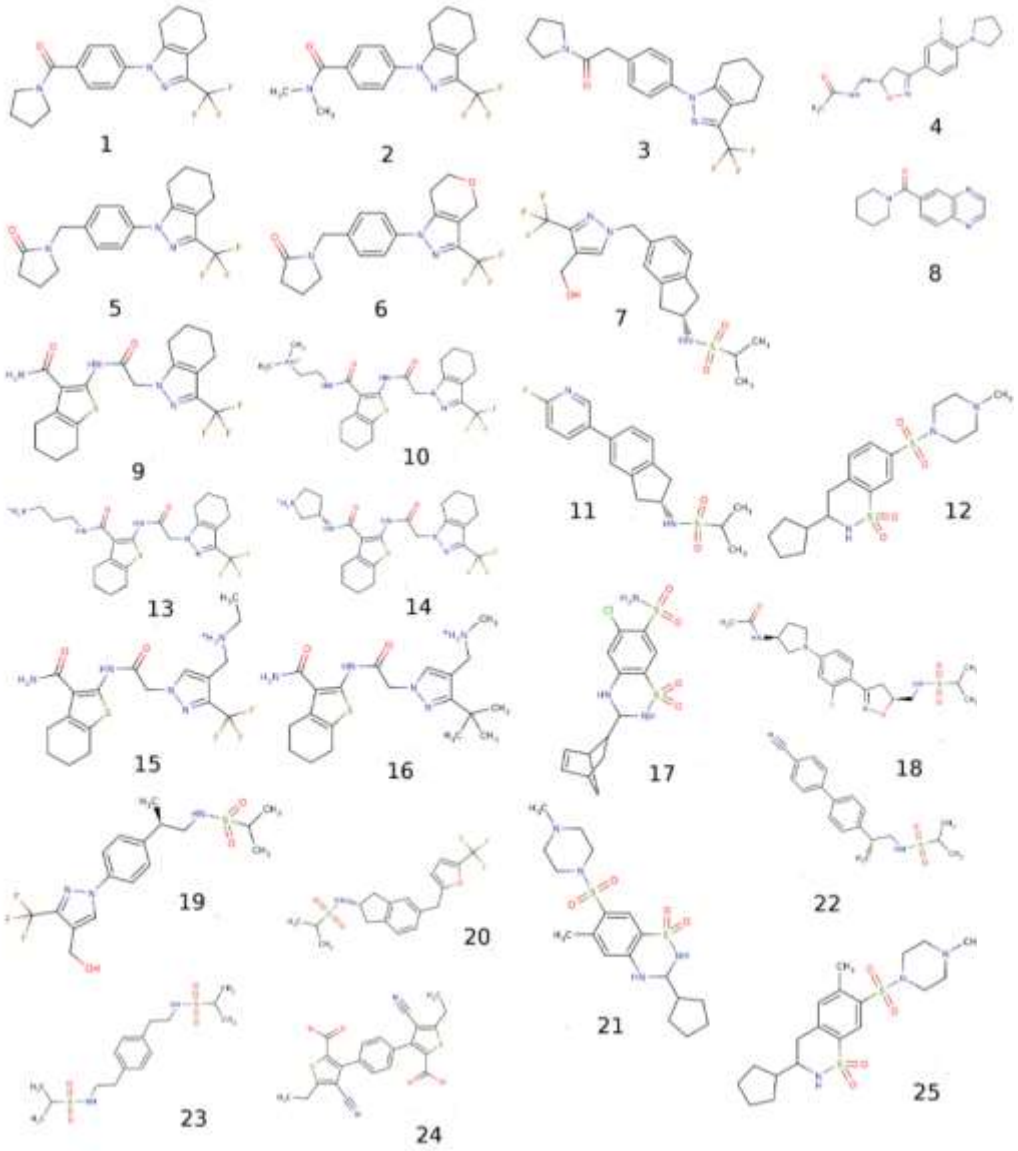


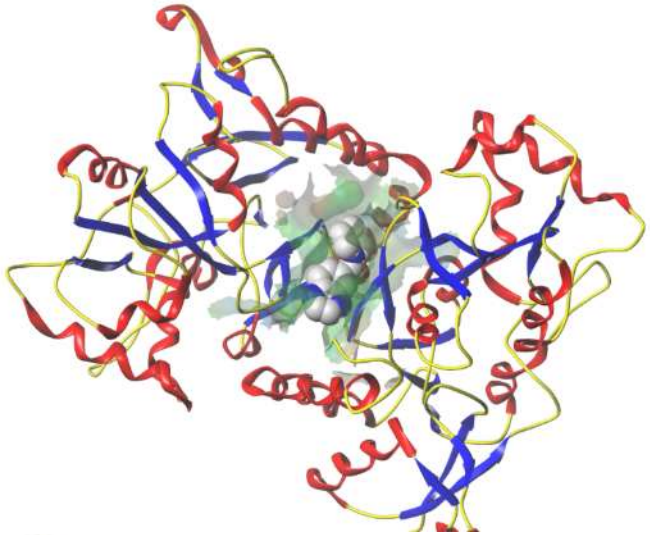


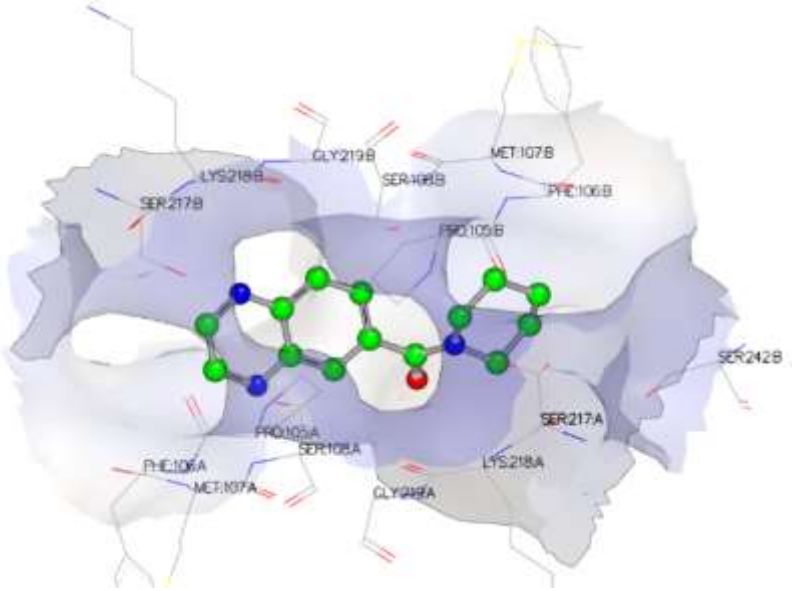
Pharmacophore: VROCS program,
OpenEye Scientific software

E.V.Radchenko, D.S.Karlov, M.I.Lavrov, V.A.Palyulin, *Mendeleev Commun.,* **2017**, *27,* 623-625.

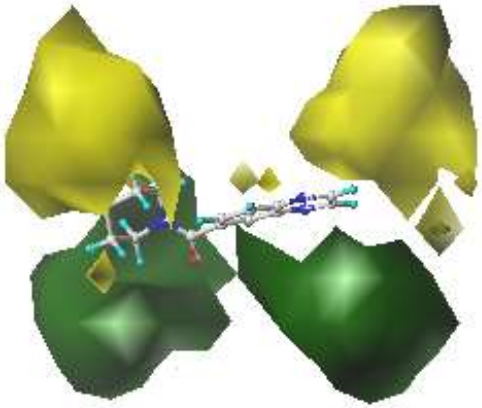
Alignment of 25 Diverse AMPA Receptor PAMs (PDB Experimental Data)

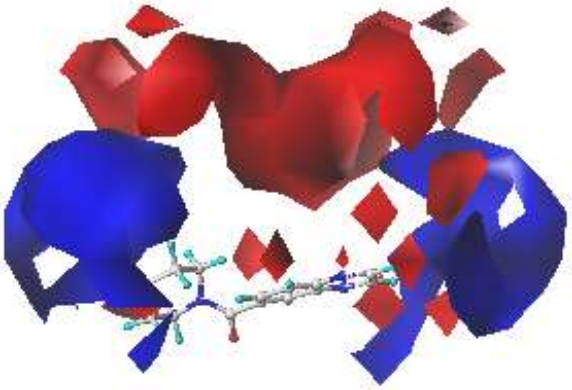




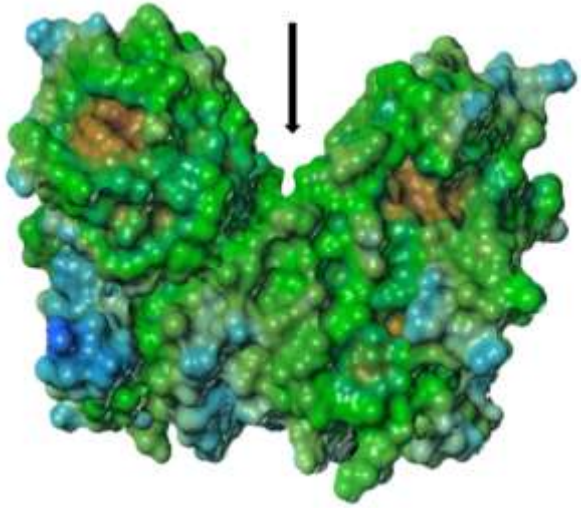


CoMFA Study of 25 Diverse AMPA Receptor PAMs

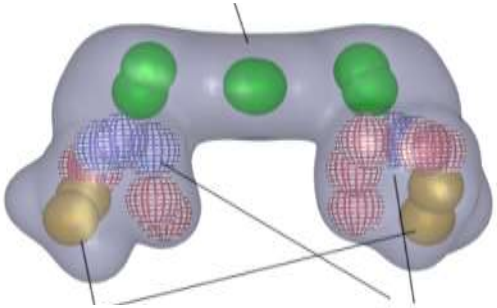




CoMFA model:
n = 25, R^2 = 0.85, Q^2 = 0.57
MMFF94 charges
Steric factors dominate

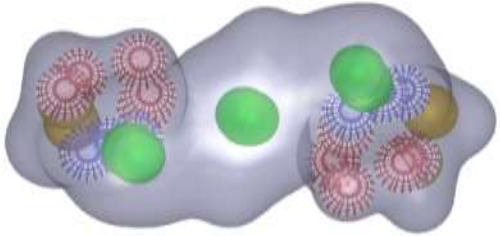


Spacer



Hydrophobic groups

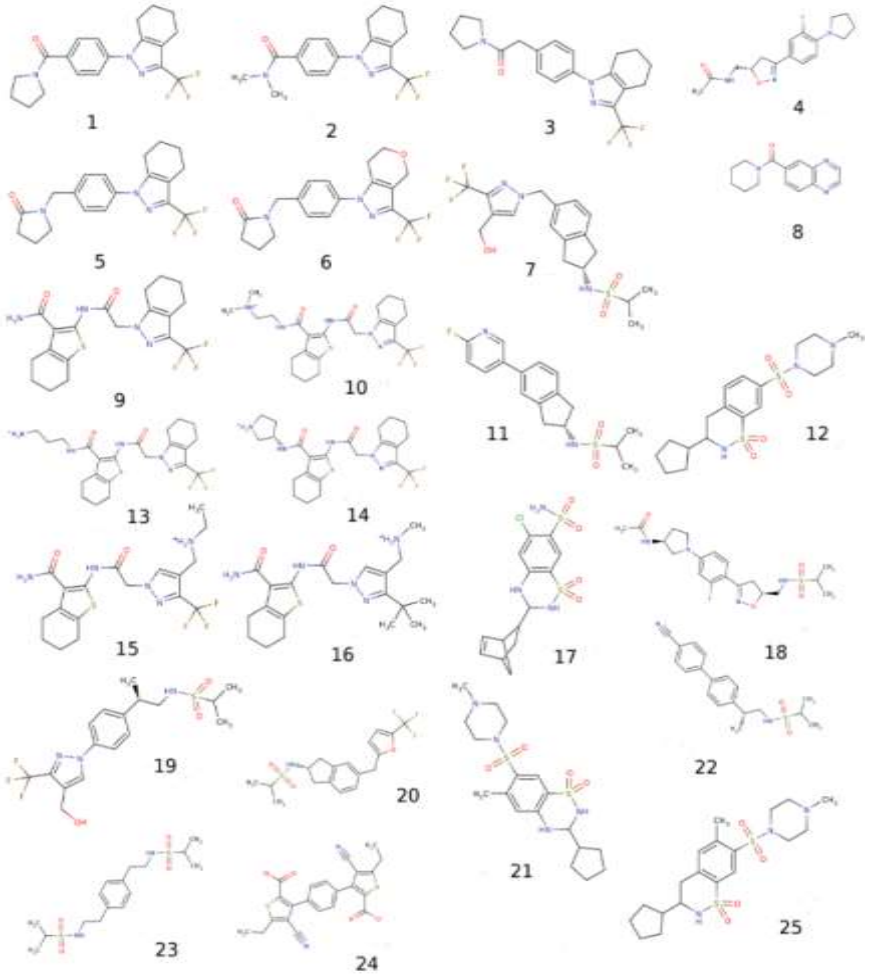
H-bond donors

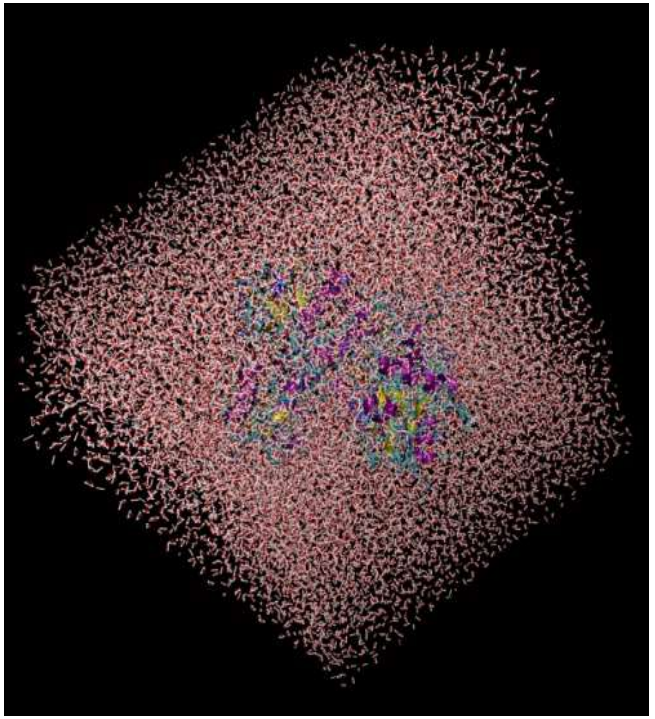


Pharmacophore:
VROCS program,
OpenEye Scientific software

D.S.Karlov, M.I.Lavrov, V.A.Palyulin, N.S.Zefirov, *Russ. Chem. Bull.*, **2016**, *65*, 581.

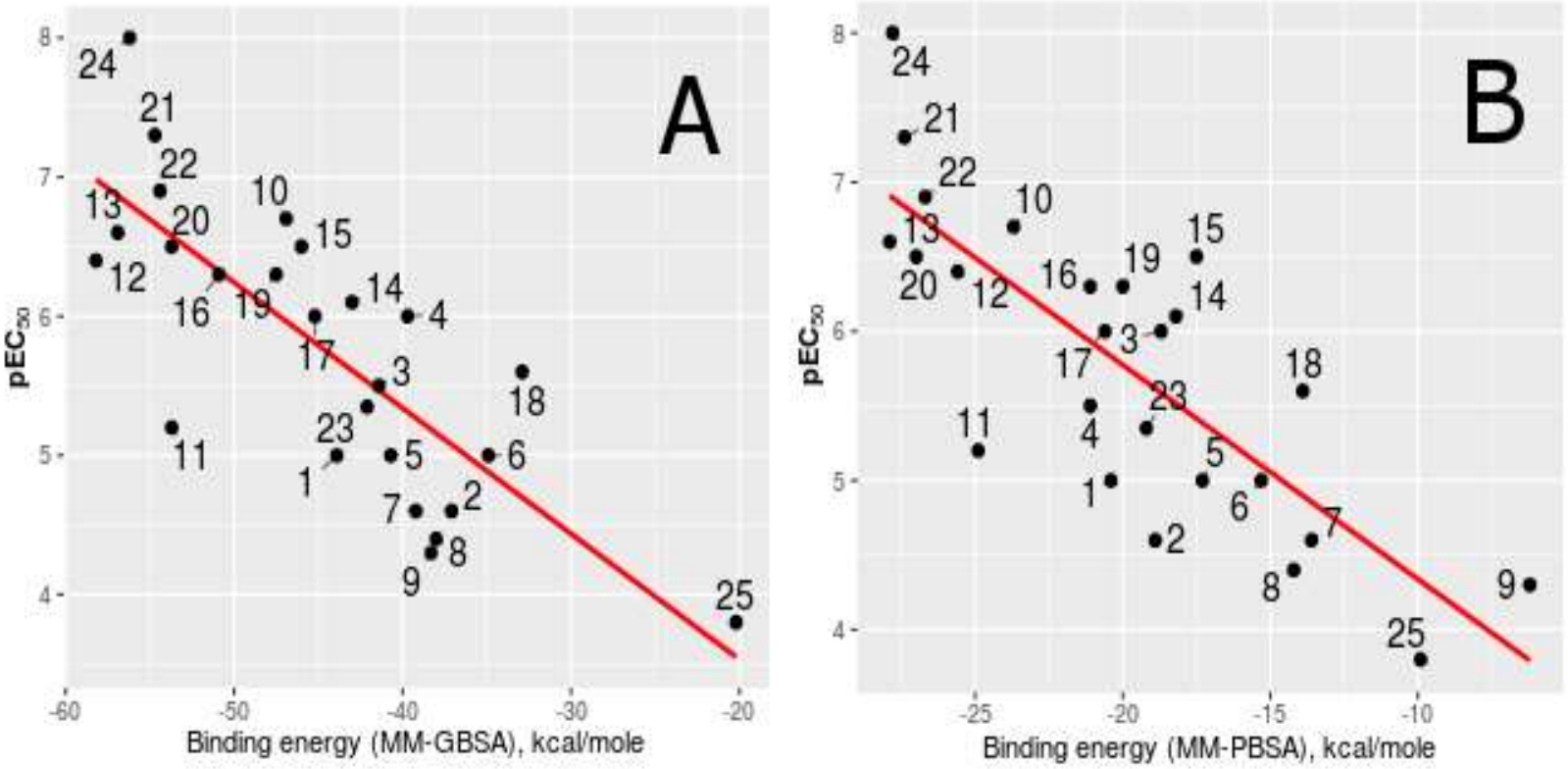
AMPA Receptor PAMs Molecular Dynamics Simulation. The Dimer of Ligand-Binding Domain + Positive Modulators (25 compounds)





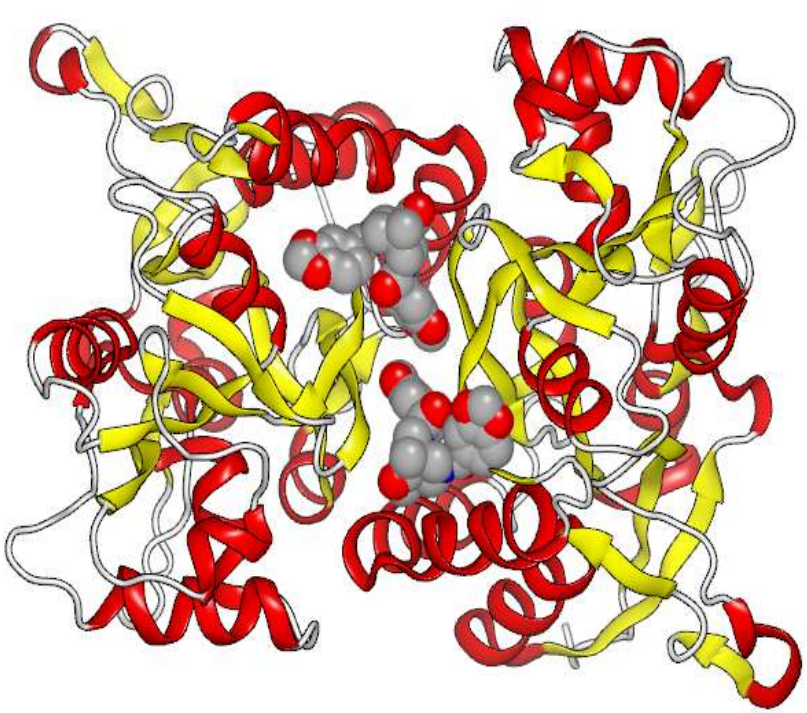
AMBER14

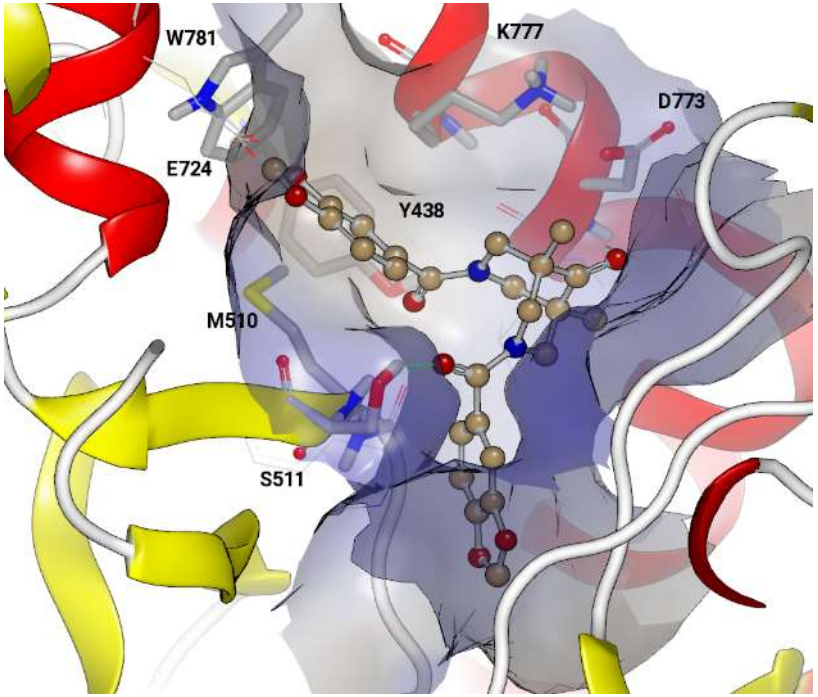
AMPA Receptor PAMs Molecular Dynamics Simulation. The Dimer of Ligand-Binding Domain + Positive Modulators (25 compounds)



D.S.Karlov, M.I.Lavrov, V.A.Palyulin, N.S.Zefirov, *J. Biomol. Struct. Dyn.*, **2018**, *36(10)*, 2508–2516.

Alternative Binding Mode of Compound OSPL-502



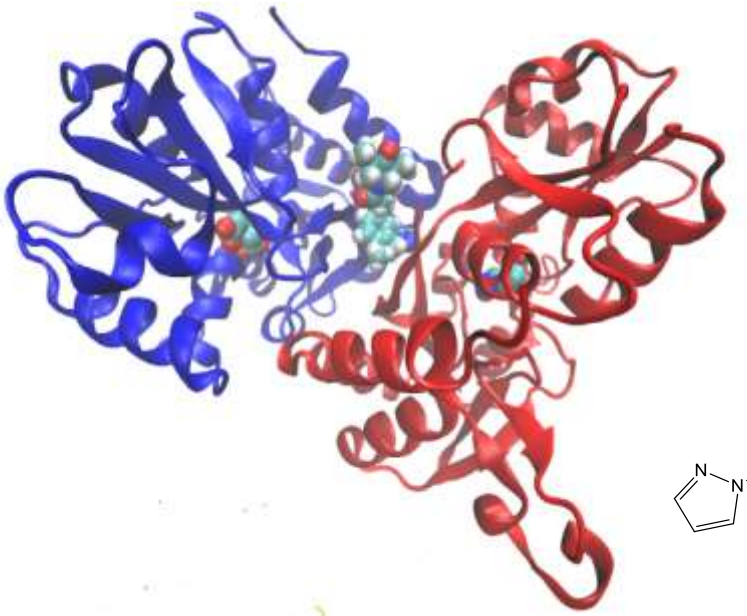


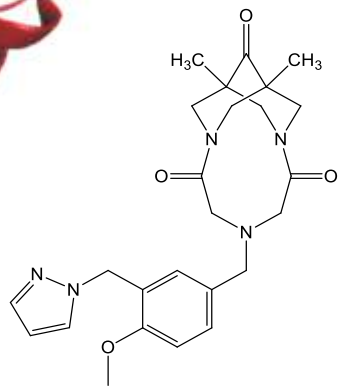
Homology models: GluA1/GluA1, GluA1/GluA2, GluA1/GluA3, GluA1/GluA4, GluA2/GluA2, GluA2/GluA3, GluA2/GluA4, GluA3/GluA3, GluA3/GluA4, GluA4/GluA4, GluK1/GluK1, GluK1/GluK2, GluK1/GluK3, GluK1/GluK4, GluK1/GluK5, GluK2/GluK2, GluK2/GluK3, GluK2/GluK4, GluK2/GluK5, GluK3/GluK3, GluK3/GluK4, GluK3/GluK5.

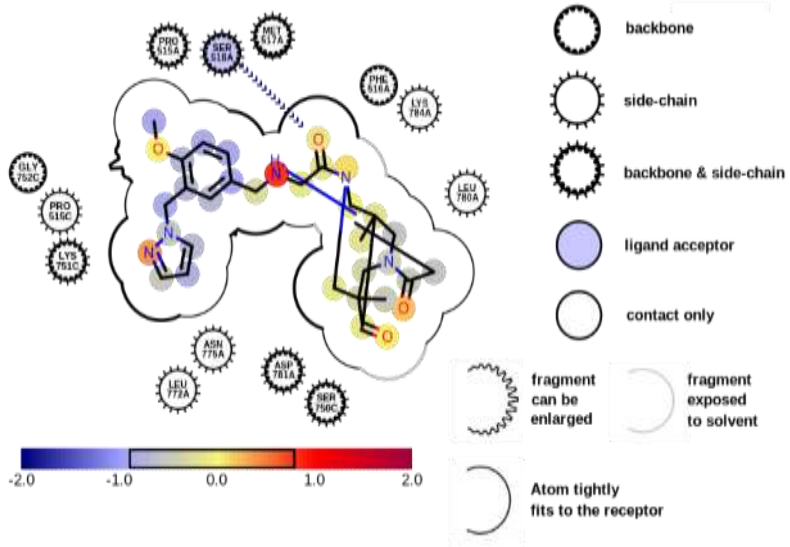
M.I.Lavrov, D.S.Karlov, T.A.Voronina, V.V.Grigoriev, A.A.Ustyugov, S.O.Bachurin, V.A.Palyulin,

Mol. Neurosci., **2020**, *57*, 191-199.

Molecular docking of tricyclic 3,7-diazabicyclo[3.3.1]nonane derivative



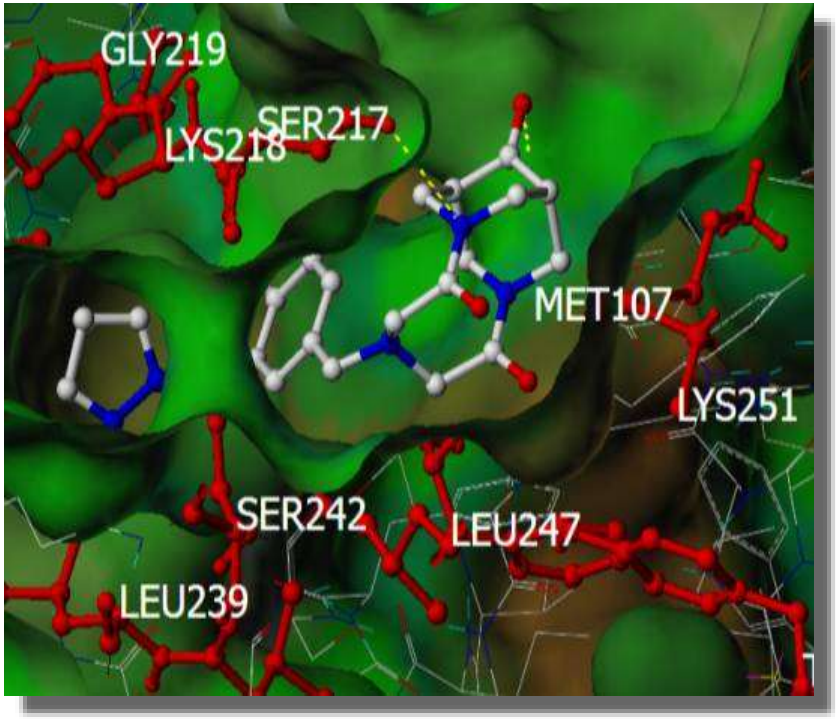


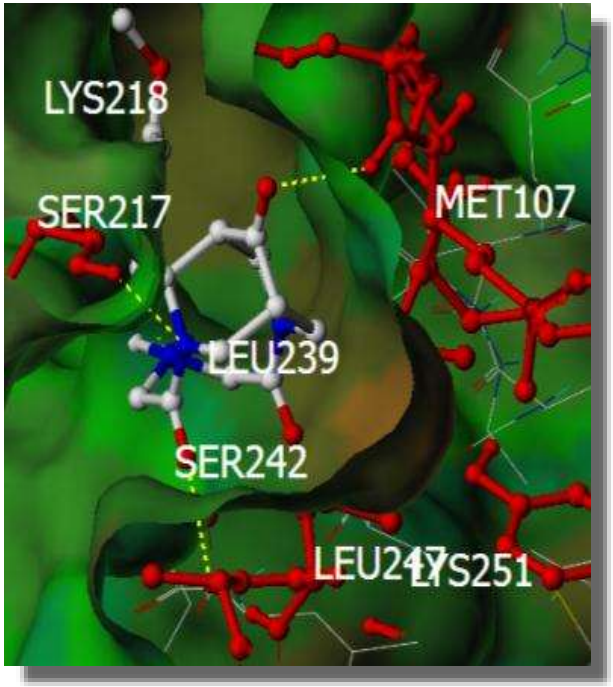


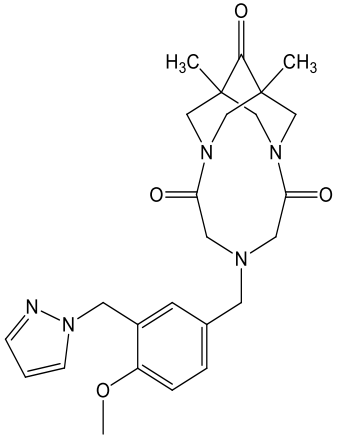
The binding pose of the tricyclic compound: a) 3D structure of the complex of the compound and GluA2 LBD homodimer; b) a schematic representation of the binding site with the colour-coded atomic contributions of *Chemgauss4* score (the colour-codes are shown on the scale under the figure, the negative contributions increase binding).

M.I. Lavrov, D.S.Karlov, V.A.Palyulin et al., *Mendeleev Commun*., **2018**, *28*, 311-313.

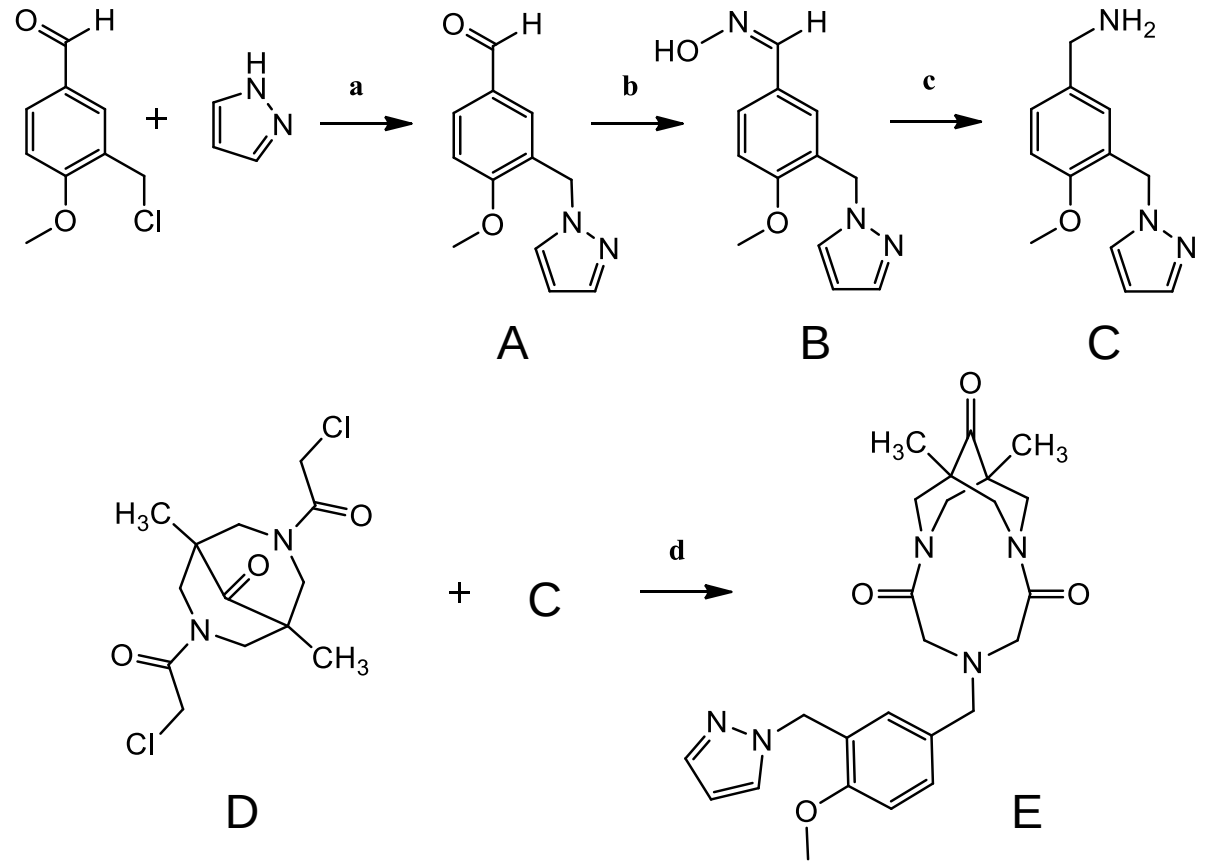
Molecular docking of tricyclic 3,7-diazabicyclo[3.3.1]nonane derivative







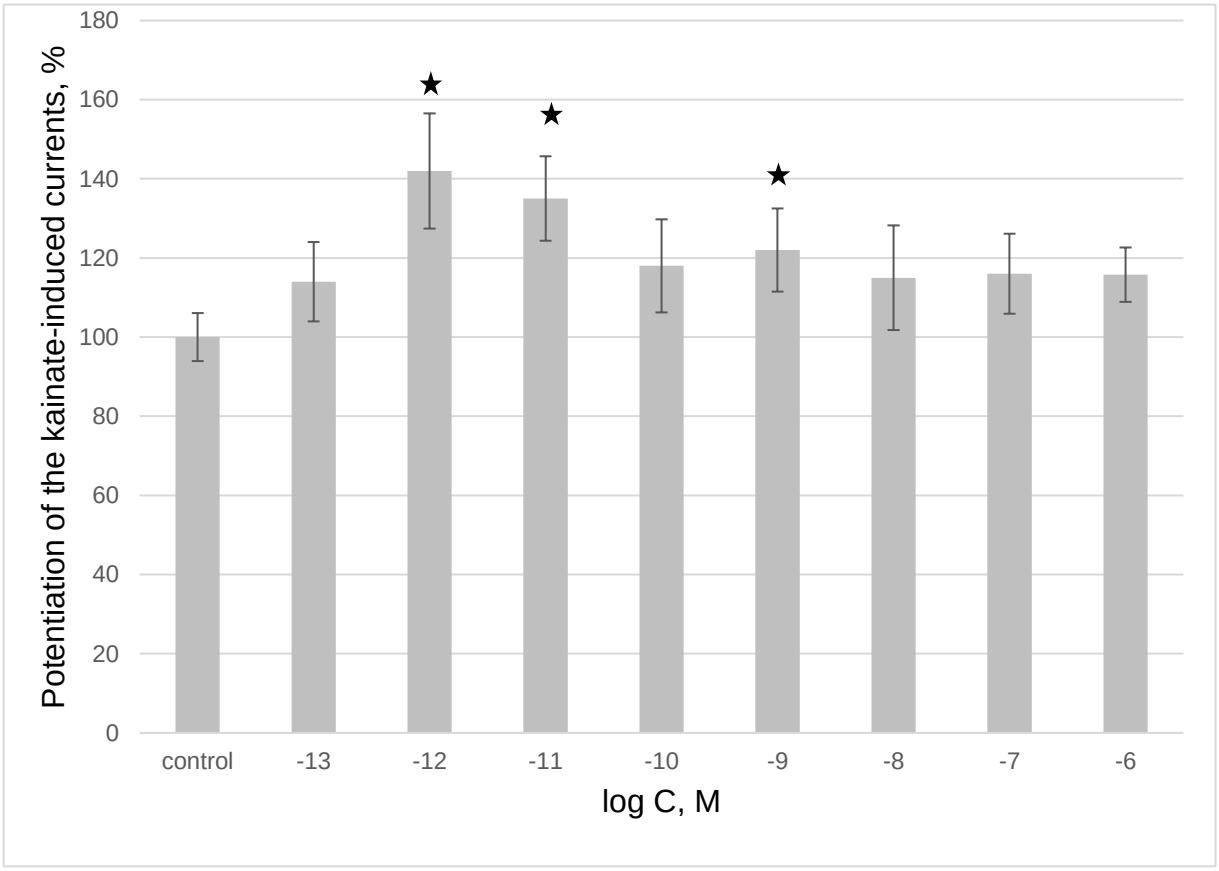
Synthesis of tricyclic 3,7-diazabicyclo[3.3.1]nonane derivative

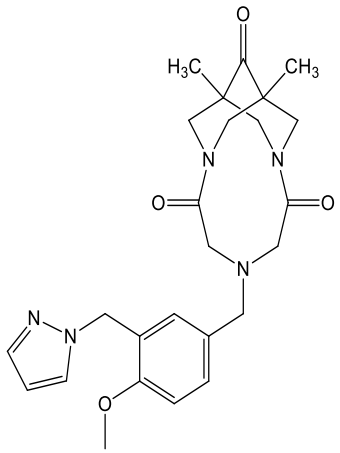


a) 18-crown-6/KOH/H_2O, 80 °C; b) $NH_2OH \cdot HCl$/pyridine/EtOH; c) $LiAlH_4$/THF; d) K_2CO_3/CH_3CN, 60 °C.

M.I. Lavrov, D.S.Karlov, V.A.Palyulin et al., *Mendeleev Commun.*, **2018**, *28*, 311-313.

In vitro study (patch-clamp) of tricyclic 3,7-diazabicyclo[3.3.1]nonane derivative





The increase in kainate-induced currents relative to the control (M±SD,%). Asterisks mark $p<0.05$.

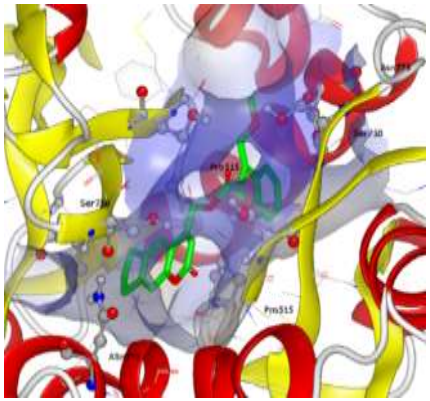
M.I. Lavrov, D.S.Karlov, V.A.Palyulin et al., *Mendeleev Commun.*, **2018**, *28*, 311-313.

Virtual Screening of New Potential AMPA PAMs

DATABASES: ZINC druglike (18 M), ZINClick (4 M), Zelinsky (150 K)

DB preparation (protonation state, conformations, etc.) –> Shape filter (10%) –> Docking (2000 structures) –> Visual analysis –> Final selection (CoMFA, Binding energy, *in silico* ADME – LogBB, HIA, hERG,…)

Shape Filter



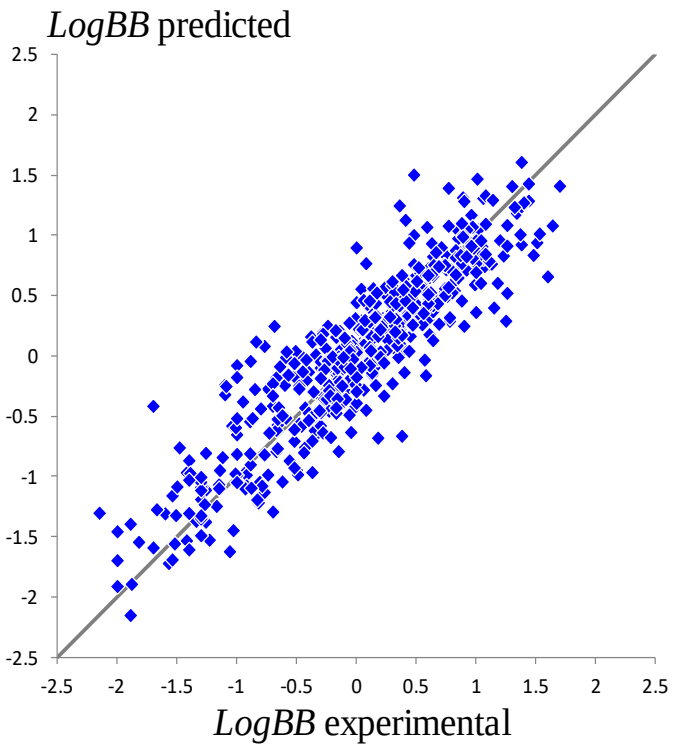
Selected Structure

30 structures were selected, then either synthesized (including close analogs) or purchased; their studies are in progress.

Evaluation of Blood-Brain Barrier Permeability

$$LogBB = \log \frac{C_{brain}}{C_{blood}}$$

http://qsar.chem.msu.ru/admet



Probably the most complete data set based on open quantitative published data – verified against original publications.
Different transport mechanisms are not considered explicitly.

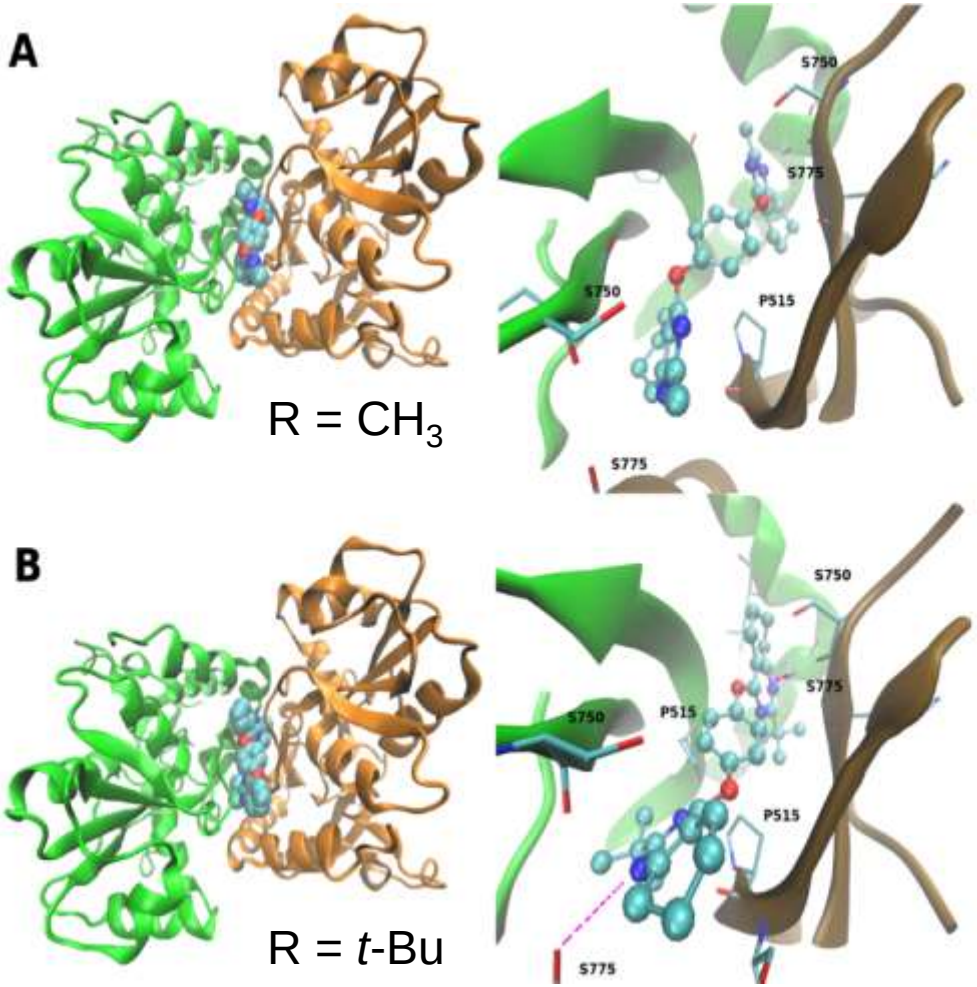
$$N = 529,\ Q^2 = 0.82,\ RMSE = 0.32$$

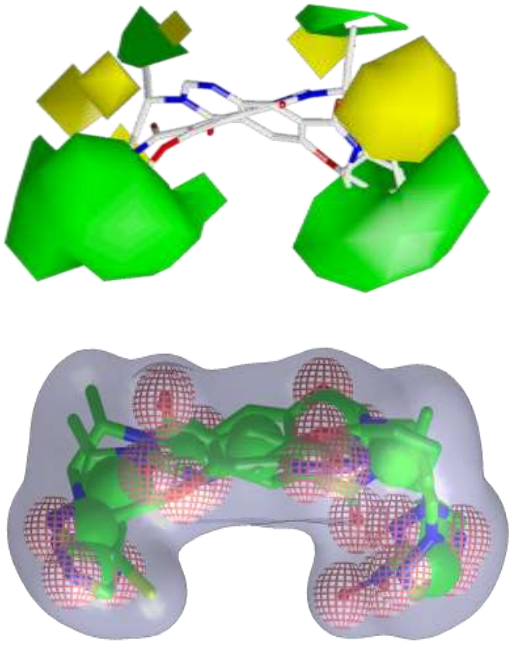
Comparable or better in accuracy and/or applicability domain compared to previously published models

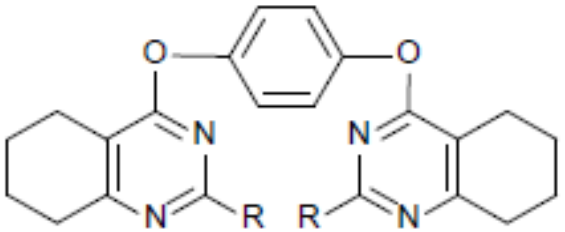
A.S. Dyabina, E. V. Radchenko, V. A. Palyulin, N. S. Zefirov, *Dokl. Biochem. Biophys.*, **2016**, *470*, 371–374.

E. V. Radchenko, A.S. Dyabina, V. A. Palyulin, *Molecules*, **2020**, *25*, 5901.

Bivalent AMPA receptor positive allosteric modulators of bis(pyrimidine) series







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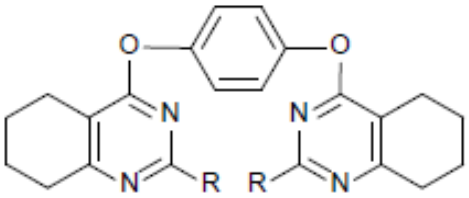
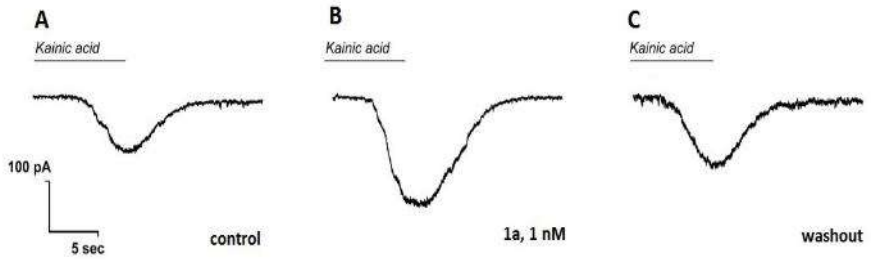
Bivalent AMPA receptor positive allosteric modulators of bis(pyrimidine) series



Patch clamp (R = Me):



R	*n*, number of neurons	Currents (%) for various concentrations of compounds (M) (control 100%)						
		10^{-12}	10^{-11}	10^{-10}	10^{-9}	10^{-8}	10^{-7}	10^{-6}
Me	7	108±5	132±5	143±9	**170±11**	123±8	85±6	78±4
Et	5	100±2	117±6	126±8	**155±5**	128±7	100±8	–
***i*-Pr**	4	100±2	84±5	72±6	82±7	92±4	98±5	–
***t*-Bu**	5	–	100±2	108±4	120±4	125±5	133±6	**145±7**
***c*-Pr**	5	–	100±2	100±2	95±4	96±3	97±2	96±5

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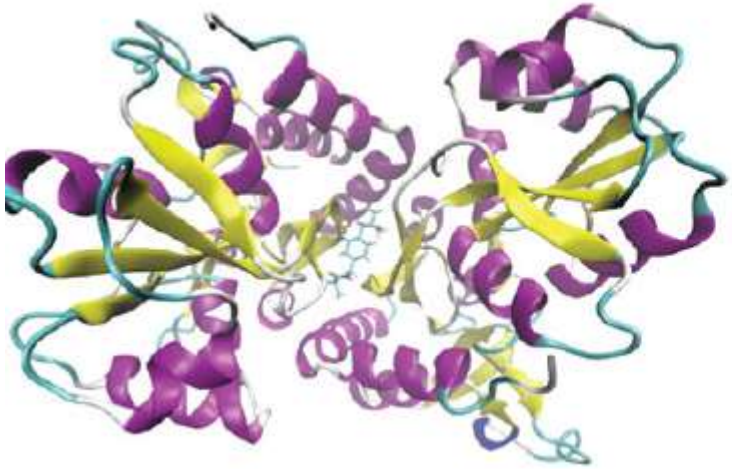
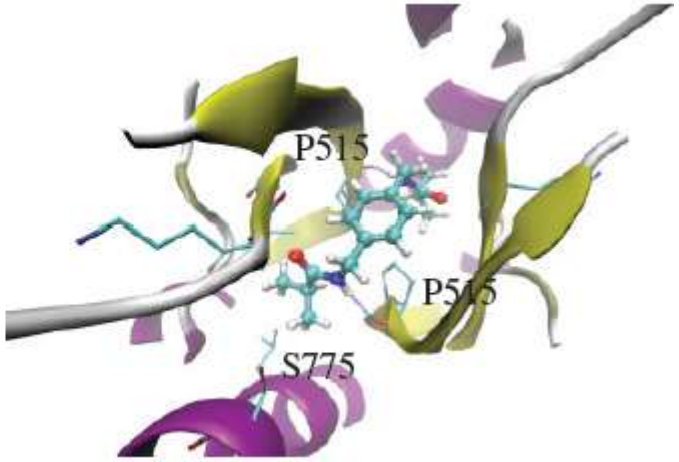
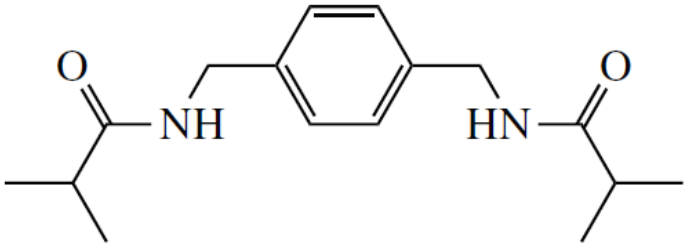
Bivalent AMPA receptor positive allosteric modulator of bis-amide series







40% potentiation at 1 nM

N.S. Temnyakova, D.A. Vasilenko, M.I. Lavrov, D.S. Karlov, Y.K. Grishin, V.L. Zamoyski, V.V. Grigoriev, E.B. Averina, V.A. Palyulin, *Mendeleev Commun.*, **2021**, *31*, 216-219.

Conclusions

A series of new positive allosteric modulators of AMPA receptor based on 3,7-diazabicyclo[3.3.1]nonane and other scaffolds was designed.

Compounds demonstrate high activity (in picomolar range), highly positive effects in in vivo tests and extremely low toxicity.

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(1935-2017)

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Biological testing:

Institute of Physiologically Active Compounds RAS,
Prof. S.O. Bachurin
Dr. V.V. Grigoriev, Dr. V.L. Zamoiski

Institute of Normal Physiology RAMS:
Laboratory of *Prof. K.V. Anokhin*

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