
Global Physicochemical Properties as Activity Discriminants for the mGluR1 Subtype of Metabotropic Glutamate Receptors

MARTA FILIZOLA, SILVINA M. TASSO,* GILDA H. LOEW,
HUGO O. VILLAR

Molecular Research Institute, 2495 Old Middlefield Way, Mountain View, California 94043

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ABSTRACT: Metabotropic glutamate receptors (mGluRs) are important as candidate therapeutic targets for many neurological disorders. In the present work, the focus has been on the mGluR1 subtype, where agonists have a proconvulsant profile while antagonists exert anticonvulsant activity. Identification of molecular determinants for the inhibition of mGluR1 provides a new avenue for the discovery and development of novel anticonvulsant drugs. Spatial configuration of key groups alone cannot explain activation selectivity at this specific receptor subtype. In fact, all known agonists and antagonists acting at mGluR1 can accommodate the same critical moieties in a similar geometric arrangement that corresponds to the extended conformation of glutamate. Therefore, other factors must account for the differences in activation. This study presents the results of an analysis of a large suite of steric, topological, electrostatic, and thermodynamic molecular properties calculated for a representative set of potent mGluR1 agonists and antagonists. Global steric parameters and the total nonpolar area provide discrimination between the mGluR1 agonists and antagonists considered in the present work. © 2001 John Wiley & Sons, Inc. J Comput Chem 22: 2018–2027, 2001

Keywords: metabotropic glutamate receptors; pharmacophore; physicochemical properties; molecular descriptors; global shape descriptors

*S. M. Tasso is a visiting scientist

Correspondence to: M. Filizola; e-mail: marta@purisima.

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Introduction

The promise of glutamate receptor ligands to aid in the pharmacotherapeutic treatment of many neurological conditions^{1–7} such as stroke, epilepsy, schizophrenia, as well as Huntington's, Alzheimer's, and Parkinson's diseases has generated a considerable body of research on them. Glutamate elicits its action via several different receptors, that are broadly classified as ionotropic (iGluRs) and metabotropic glutamate receptors (mGluRs). Ionotropic glutamate receptors are ligand-gated ion channels while metabotropic glutamate receptors belong to the G-protein coupled receptor (GPCR) superfamily.

The eight different mGluR subtypes can be divided into three groups⁸ based on sequence identity, second messengers, and subsequent downstream cascades they initiate. Group I includes mGluR1 and mGluR5, group II contains mGluR2 and mGluR3, and finally group III includes mGluR4, mGluR6, mGluR7, and mGluR8. Sequence homology is about 70% for mGluRs within the same group, while it decreases to about 40% across different groups.⁵ Furthermore, group I receptor subtypes increase phosphoinositide hydrolysis when activated, leading to intracellular calcium mobilization.^{9,10} In contrast, the mGluR groups II and III are negatively coupled to the adenylyl cyclase–cyclic adenosine activity.^{11,12}

Agonists acting at group I mGluRs enhance neural excitability and consequently have proconvulsant activity, while antagonists acting at these receptor subtypes exert anticonvulsant activity.^{13–20} A predominant anticonvulsant action is observed when receptors of group II or III are activated,^{13,19} although a mixed convulsant/anticonvulsant action can also appear at these receptors. Conversely, antagonists at mGluRs of groups II and III do not form any coherent pattern of activity as they have been found to be involved in both proconvulsant or anticonvulsant activities.^{13,21}

The structures of metabotropic glutamate receptors conform to the seven-transmembrane spanning domains common to all GPCRs, particularly those characterized by a large N-terminus. Very recently, three different crystal structures²² of the large extracellular domain of the group I mGluR1 subtype have been reported, but the corresponding crystallographic atomic coordinates have not been released yet. Computer-assisted drug design based on the knowledge of the target structure cannot be applied at this juncture.

Development of reliable computational approaches aimed at the design of novel compounds using ligand-based strategies continues to be of interest in general and in particular for mGluRs. Previous pharmacophore models for mGluR1,²³ mGluR2,²⁴ and mGluR4 subtypes,²⁵ as representatives of group I, II, and III mGluRs, respectively, provided some insight into the glutamate bound conformation and into the topology of its protein environment. Glutamate was suggested to bind in an extended conformation with maximum separation between its zwitterionic group and the distal proton-accepting center. Because all known mGluR ligands possess a zwitterionic amino acid group (α -amino and carboxylate groups) and a distal proton-accepting center, these three chemical moieties are present in all pharmacophore models developed so far. Hydrogen bond interactions were hypothesized between these three ligand moieties and the protein environment. Because of the strong directionality of these interactions, the carboxylate's oxygens were explicitly considered in the pharmacophore definitions, leading to five-point pharmacophore models for all mGluR subtypes of groups I, II, and III.

The work published to date shows that distances between pharmacophore points alone do not explain activation selectivity at mGluRs, because at least a conformation of all mGluR agonists and antagonists can be found that accommodates the same critical moieties in space, in the same geometric arrangement corresponding to the extended conformation of glutamate. This is a case in which a pure pharmacophore representation is unable to discriminate between sets of compounds with convulsant or anticonvulsant activity. Consequently, other factors are at play that are not captured by analysis of distances among putative pharmacophoric centers alone. These factors can be interpreted in terms of molecular descriptors accounting for all major aspects of activity at a given receptor, including steric, topological, electrostatic, and thermodynamic properties.

In prior studies,²³ excluded volume calculations of all agonists at group I subtypes were used to discriminate between active and inactive compounds. Global molecular descriptors that go beyond the analysis of critical distances for ligand discrimination have not been systematically explored and constitute the main purpose of the present work. A suite of molecular properties was computed for a representative set of potent agonists and antagonists at mGluR1, using a variety of techniques. These molecular descriptors were analyzed to determine putative steric, topological, electrostatic,

or thermodynamic factors that may discriminate between agonists and antagonists at mGluR1. Moreover, two new global steric properties are introduced to obtain a more detailed description of the overall molecular shape. Specifically, molecular globularity as a measurement of the spherical character of a molecule has been extended to also include other spheroids. These properties could at some times become useful topological descriptors in this and other cases of indirect drug design. The results of the present work can serve as guide for modulating the antagonistic potency of known mGluR1 ligands, ultimately aiming at the rational design of novel anticonvulsant drugs.

Methods

Structures for all the compounds selected were built in their ionic form using the MSI/Quanta package (MSI-Quanta, Biosym/MSI, San Diego, CA). The zwitterionic form was used for the amino acid groups while the distal proton-accepting centers were considered to be ionized for all selected molecules. Whenever available, geometries from X-ray crystallographic data were used as starting points for the structural analysis of the compounds included in the present work. Crystallographic structures were considered for (S)-glutamic acid,²⁶ (S)-quisqualic acid,²⁷ (1S,3R)-1-aminocyclopentane-1,3-dicarboxylic acid ((1S,3R)-ACPD),²⁸ and (S)- α -aminoadipic acid (AMADIP).²⁹ All other initial structures were built using the Quanta package.

CONFORMATIONAL ANALYSIS AND SELECTION OF A BIOACTIVE FORM

The Quanta/CHARMm force field³⁰ was used to energy minimize the initial structures of all selected mGluR1 ligands and to explore their conformational space. A dielectric constant of 80 and no cutoff were used as the simplest approximation to bulk water, appropriate for charged ligands. Energy minimization was carried out using 200 steps of steepest descent followed by conjugate gradient algorithms until the root mean square deviation (rmsd) of the changes between iterations were less than 0.01 Å.

Conformational analysis for each of the selected compounds was carried out using a systematic nested rotation procedure. Every significant rotatable bond was scanned in 30° increments from 0 to 360° and followed by energy minimization of the resulting conformations using symmetry whenever possible to reduce the number of steps in the

search procedure. The structures derived from the conformational searches were ordered according to energy and checked for uniqueness. A conformation was considered unique if one of its significant torsion angles differed by 30° or more from all other conformations identified. Accuracy of our criterion of conformational uniqueness based on differences in torsion angles higher than 30° was previously demonstrated by diverse test assessments on similar small molecules carried out in our laboratory.^{31,32}

A candidate bioactive form for all agonists and antagonists acting at mGluR1 was then identified using the in-house computer program MOLMOD. MOLMOD is a pharmacophore-generating software based on systematic pairwise comparisons of the distances between the chemical moieties common to all binders or active compounds at a given receptor. The details of this software are described elsewhere.^{31,33} Briefly, two inputs are necessary for running MOLMOD: (1) a set of candidate pharmacophoric points common to each compound, and (2) a conformational library for each compound. A comparison of the spatial arrangement of the chemical moieties common to all compounds in a class is automatically done for all the conformers using the library of unique conformers generated. Only conformations within 3 kcal/mol of the lowest energy minima found for each compound were considered. The selection of this 3 kcal/mol energy cutoff is based on recent studies published in the literature^{34,35} in which the presence of the bioactive conformation within such a threshold has been shown for a variety of ligand-protein complexes of known structure. Although the bioactive conformation of glutamate was recently suggested to correspond to the extended one,^{23,24} exhaustive conformational searches for each of the selected mGluR1 ligands were carried out to verify that this was the only possible conformation common to all of them.

The bioactive conformation selected was then used as a starting point for semiempirical calculations using the AM1 Hamiltonian³⁶ embedded in the MOPAC 7.0 software.³⁷

EVALUATION OF MOLECULAR PROPERTIES

Several physicochemical properties were evaluated for each of the selected agonists and antagonists acting at mGluR1 using empirical methods and quantum chemical techniques. All properties were expressed in terms of computable molecular descriptors accounting for the steric, topological, electrostatic, and thermodynamic in-

TABLE I.
Summary of Molecular Descriptors.

Molecular Descriptor	Symbol
Molecular weight	M_w
Total volume	V_{tot}
Polar volume	V_P
Nonpolar volume	V_{NP}
Total area	A_{tot}
Polar area	A_P
Nonpolar area	A_{NP}
Solvent-accessible surface area	SASA
Molecular globularity	G
Prolate spheroid shape index	PSSI
Oblate spheroid shape index	OSSI
Sterimol parameters	L, B1, B2, B3, B4
Wiener number	W
Shannon index	H ¹
Flexibility index	FI
Total topological index	TETS2
Hydrogen bond donors number	HBD
Hydrogen bond acceptors number	HBA
Lowest energy unoccupied molecular orbital	LUMO
Highest energy occupied molecular orbital	HOMO
Hydrophobic index	Hyd
Solvation free energy difference	ΔG_{solv}
Rotational entropy	S_{rot}
Total enthalpy	H_{tot}
Heat capacity	C
Total entropy	S_{tot}

formation contained in each selected mGluR1 agonist or antagonist. Table I summarizes the molecular descriptors used for this study and their corresponding notation.

Simple topological descriptors such as the Shannon index,³⁸ the Wiener number,³⁹ the flexibility index,⁴⁰ and the total topological index TETS2⁴¹ were calculated using empirical methods embedded in the software Molconn-X version 2.0 (Hall Associates Consulting, Quincy, MA). All these parameters provide a simple topological description of molecular shape.

Molecular descriptors accounting for the steric aspect of each molecule such as total area, total volume, total solvent-accessible area, and the sterimol parameters⁴² (L, B1, B2, B3, and B4), were also computed as part of the parameter set using the in-house software GRAPHa, which stands for GRAPHics Analysis. This computer program uses as inputs the results of the semiempirical calculations carried out using MOPAC7.³⁷ Other 3D shape descriptors were obtained by using GRAPHa.

Among them, the molecular globularity (G) of each compound was computed. Globularity has been defined in the past to describe the shape of polypeptides, but it has been extended to characterize the overall shape of molecules.⁴³ The parameter G is defined as the ratio of the area of a sphere (A^{SP}) with a volume (V) equivalent to the molecular volume (V_{mol}) over the area of the actual molecular surface (A):

$$G = \frac{[A^{SP}]_{V=V_{mol}}}{A} = \frac{(4\pi)^{1/3}(3V_{mol})^{2/3}}{A}$$

It assumes values between 0 and 1, with the upper bound corresponding to a perfect spherical molecule. Volumes and areas used the van der Waals parameters determined by Gavezzotti.⁴⁴

We have extended the concept of globularity by comparing the area of molecules of interest to other idealized bodies, including oblate and prolate spheroids, as molecular shape descriptors. To determine the area of these ideal bodies, two parameters are needed. As in the case of the globularity, one of the parameters taken is the volume for the ideal body, which is equated to the molecular volume. The second parameter is the length of the major axis for each body, which is equated to the maximum separation between two points on the molecular surface (L in the sterimol parameters). These two parameters determine the area of the ideal body, and in the case of spheroids, it also determines the length of their minor axis. The equations used to calculate a prolate spheroid shape index (PSSI) and an oblate spheroid shape index (OSSI) are reported below:

$$PSSI = \frac{1}{A} \left(2\pi B^2 \frac{2\pi LB}{\varepsilon \sin(\varepsilon)} \right)$$

where B and ε are measurements for minor axis and ellipticity, respectively, while L is the major axis and is equated to the maximum separation between two points on the molecular surface. Specifically,

$$B = \sqrt{(3V_{mol}/4\pi L)} \quad \text{and} \quad \varepsilon = \sqrt{(L^2 - B^2)/L}$$

OSSI provides a comparison to an oblate spheroid,

$$OSSI = \frac{\pi L^2}{2} + \frac{\pi B^2}{\varepsilon} \text{Log} \frac{1 + \varepsilon}{1 - \varepsilon}$$

where its minor axis (B) is given by:

$$B = \sqrt{(3V_{mol}/\pi L^2)}$$

and its ellipticity (ε) can be calculated using the equation above. Contrary to the globularity that has an upper bound of 1, these 3D shape indexes can adopt any positive value. However, a value of 1

does indicate a close similarity to the ideal shape in question.

Polar and nonpolar areas as well as polar and nonpolar volumes were computed using atomic contributions to the total area and volume. The contribution of nitrogen and oxygen atoms was considered as polar; all other atoms were considered nonpolar.

Among the global properties able to provide some insight into the electrostatic differences between agonists and antagonists, we also computed some simple parameters. The number of hydrogen-bond donors and acceptors within each molecule were considered as candidate discriminant factors. Moreover, electrostatic descriptors such as the lowest unoccupied molecular orbital (LUMO) and the highest occupied molecular orbital (HOMO), as well as thermodynamic properties such as rotational and total entropy, total enthalpy, and heat capacity were obtained by semiempirical calculations using the AM1 Hamiltonian³⁶ embedded in the MOPAC7 package.³⁷ The neutral form of all selected mGluR1 ligands was used for these calculations. Energy minimization of these initial structures was carried out until gradient changes were less than 0.1 Å. The resulting conformations were found within the domain of the original conformations generated with CHARMM. The in-house software GRAPHa was also used to calculate hydrophobic index values, using an AM1 three-parameter method developed in our laboratory.⁴⁵ Similarly, values of free energy of solvation were calculated for all the compounds included in the present work, using the AMSOL 6.5.3/AM1 package⁴⁶ and water as solvent at the geometries determined *in vacuo*. Analysis of all steric, topological, electrostatic, and thermodynamic properties was performed with the aid of the S-PLUS 2000 package (StatSci Division, MathSoft Inc., Seattle, WA).

Results and Discussion

SELECTION OF POTENT AGONISTS AND ANTAGONISTS AT mGluR1

Among the ligands for which experimental results of PI hydrolysis stimulation activity at mGluR1 have been published in the literature, only 13 compounds were found having either clear agonistic or antagonistic activity at mGluR1. Compounds with a published maximum value of 80 μ M for either EC₅₀ or IC₅₀ were considered potent mGluR1 agonists or antagonists, respectively. Figure 1 shows the chemical structures of all the compounds classified in

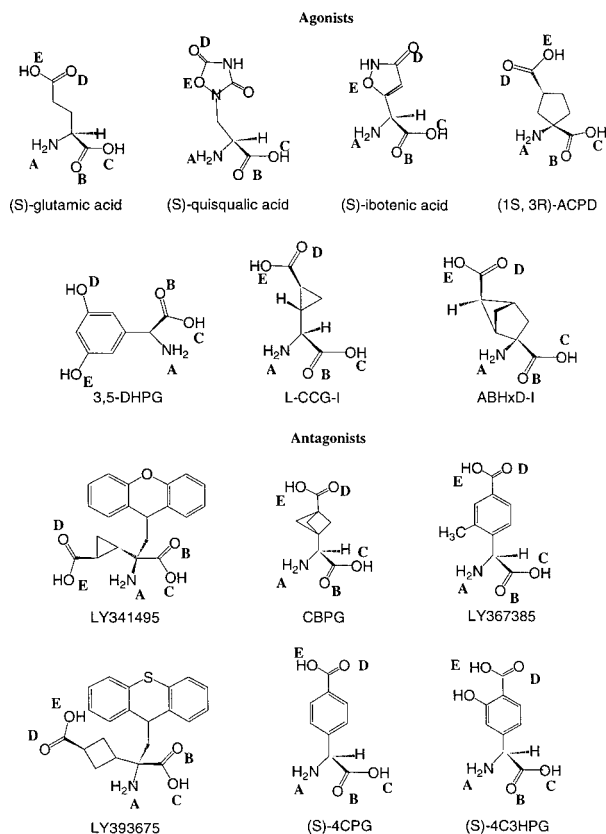


FIGURE 1. Chemical structures of the compounds considered in the present work as potent agonists and antagonists at mGluR1. Also shown are the chemical moieties common to all the compounds within the same class. Specifically, for all of them, A refers to an amino group, B and C indicate the two oxygen atoms of the proximal acidic group, while the oxygen atoms of the distal proton-accepting center are labeled by D and E.

the present work as potent agonists or antagonists at mGluR1, while Table II reports the experimental values published in the literature on each of the selected compounds. Five chemical moieties were found in common to the seven agonists and six antagonists at mGluR1. These moieties are shown as points A to E in Figure 1. A refers to an α -amino group, B and C indicate the two oxygen atoms of the proximal acidic group, while the two oxygen atoms of the distal proton-accepting centers are labeled by D and E.

Systematic pairwise comparisons of the distances between these five common chemical moieties were carried out for all the selected compounds using the low energy conformers within 3 kcal/mol from the lowest energy conformation found for each ligand, as input to an automated unbiased method embedded in the in-house computer pro-

TABLE II.
Experimental Data of Agonistic/Antagonistic Activity at mGluR1.

Compound	Class	EC ₅₀ (μ M) (PI Hydrolysis)	IC ₅₀ (μ M) (PI Hydrolysis)	Ref.
(S)-Glutamic acid	Agonist	9–13	—	48
(S)-Quisqualic acid	Agonist	0.1–3.0	—	48
(S)-Ibotenic acid	Agonist	2–60	—	48
(1S, 3R)ACPD	Agonist	5–80	—	48
3.5 DHPG	Agonist	6–30	—	48
L-CCG-I	Agonist	2–50	—	48
ABHxD-I	Agonist	1.6	—	49
LY341495	Antagonist	—	7.8	50
CBPG	Antagonist	—	25–32	48
LY367385	Antagonist	—	8	51
LY393675	Antagonist	—	0.35	52
(S)-4CPG	Antagonist	—	15–65	48
(S)-4C3HPG	Antagonist	—	10–40	48

gram MOLMOD.^{31, 33} Table III reports the results derived from these conformational searches including the number of significant rotatable bonds considered for each ligand, the total number of unique conformers and the number of unique conformers within 3 kcal/mol from the lowest energy conformation found for each compound.

MOLMOD automatically identified conformers in each of the mGluR1 ligands that displayed the selected common chemical moieties A, B, C, D, and E in the same spatial arrangement. Our results agree with previous conformational studies that suggest that the bioactive conformation of glutamate corre-

sponds to the extended one,^{23, 24} and this was the only possible conformation common to all selected mGluR1 agonists and antagonists.

Figure 2 shows the best overlay of the conformers selected for each agonist and antagonist at mGluR1. The superposition was carried out overlapping the chemical moieties A, B, C, D, and E present in the lowest energy conformer satisfying the common geometric arrangement as found with MOLMOD. Because the selected agonists and antagonists at mGluR1 can accommodate the same critical moieties in space in the same geometric arrangement, no explanation can be found for their different ef-

TABLE III.
Computational Results of the Conformational Searches Performed for all the Selected Agonists and Antagonists at mGluR1.

Compound	Class	Number of Significant Rotatable Bonds	Total Number of Unique Conformers	Number of Unique Conformers within 3 kcal/mol
(S)-Glutamic acid	Agonist	4	104	80
(S)-Quisqualic acid	Agonist	3	32	27
(S)-Ibotenic acid	Agonist	2	11	11
(1S, 3R)-ACPD	Agonist	2	24	4
3,5-DHPG	Agonist	2	29	17
L-CCG-I	Agonist	3	21	21
ABHxD-I	Agonist	2	19	19
LY341495	Antagonist	5	2027	58
CBPG	Antagonist	3	38	38
LY367385	Antagonist	3	8	8
LY393675	Antagonist	5	1697	14
(S)-4CPG	Antagonist	3	13	12
(S)-4C3HPG	Antagonist	3	10	10

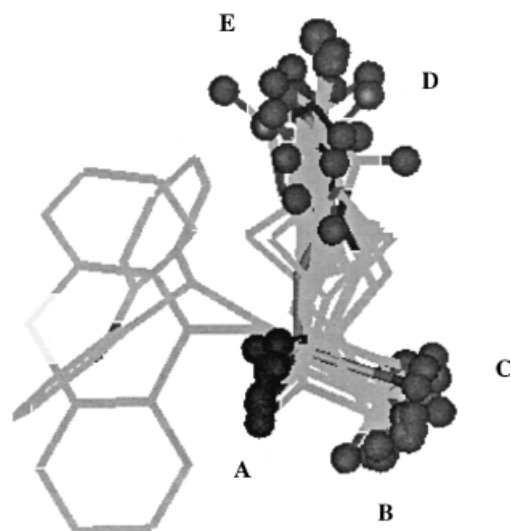


FIGURE 2. Superposition of the lowest energy conformers of all selected agonists and antagonists at mGluR1 obtained by spatial overlap of their five common chemical moieties, A, B, C, D, and E.

fect based on a distance analysis approach. The limitations of this approach were already described in previous computational studies.^{23–25} Thus, other factors that cannot be identified by simple analysis of distances among putative pharmacophoric centers alone must be systematically explored in order to identify reasons for activation selectivity at mGluR1.

Interestingly, two of the most potent antagonists acting at mGluR1, LY341495 and LY393675, have a bulky hydrophobic group in a similar spatial arrangement (left side of Fig. 2). The observation points to the potential importance of steric and hydrophobic parameters for the differentiation of agonists from antagonists at mGluR1. In the case of other anticonvulsants, such as valproic acid, different approaches for quantitative structure-activity relationship analysis (QSAR) demonstrated the importance of the lipophilicity for activity.⁴⁷

MOLECULAR DESCRIPTORS AND LINEAR DISCRIMINANT ANALYSIS

Steric, topological, electrostatic, and thermodynamic descriptors were computed for each of the mGluR1 ligands included in the present work, and considered for the purpose of discrimination between agonists and antagonists. The bioactive conformation of each compound corresponding to the extended conformation of glutamate as found by MOLMOD, was used for all the property evaluation. A list of the corresponding molecular de-

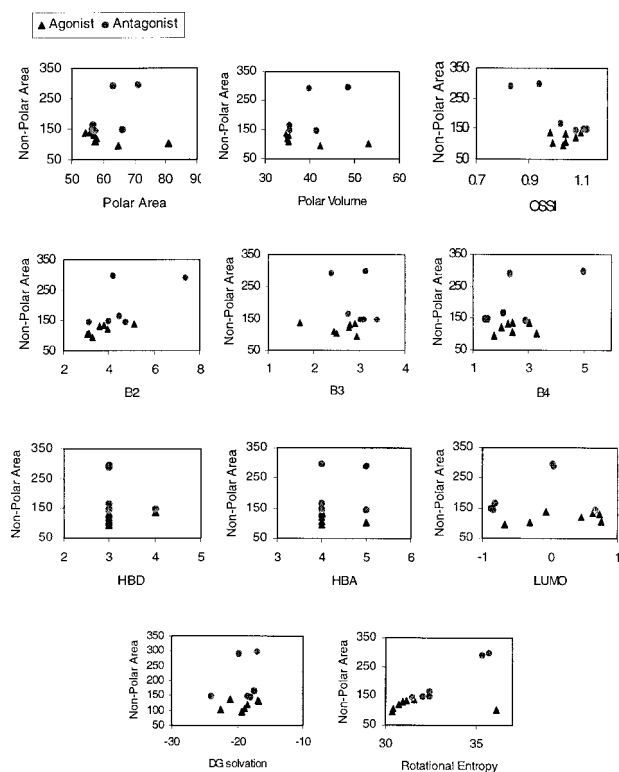


FIGURE 3. Plots of the best molecular descriptor (nonpolar area) providing discrimination between agonists and antagonists versus its noncorrelated chemical properties.

scriptors is provided in Table I. The number of electrostatic descriptors that is possible to compute is somewhat limited because of the charged character of the mGluR1 ligands. Nevertheless, as already indicated, there is some suggestion that either steric properties or hydrophobicity may play a role in the discrimination. Table IV reports the numerical values for all the steric and topological descriptors calculated for each agonist and antagonist at mGluR1. Similarly, Table V reports the numerical values for all electrostatic and thermodynamic descriptors.

Analysis of all properties suggests a crucial role of steric properties rather than thermodynamic or electrostatic factors in the discrimination between agonists and antagonists acting at mGluR1. Several properties including total molecular volume, nonpolar volume, solvent accessible, total, and nonpolar areas, globularity, and the maximum molecular span according to the L sterimol parameter provide a discrimination between agonists and antagonists. Values of total volume with a lower bound of 150 \AA^3 are requested for antagonism at mGluR1. This observation is in agreement with previous results.²³

TABLE IV.
Numerical Values of Steric and Topological Molecular Descriptors Calculated for all Selected Agonists and Antagonists at mGluR1.

Compound	M_w (g/mol)	V (\AA^3)	V_p	V_{NP}	A (\AA^2)	A_p	A_{NP}	SASA (\AA^2)	G (\AA)	PSSI	OSSI	L (\AA^3)	B_1 (\AA^3)	B_2 (\AA^3)	B_3 (\AA^3)	B_4 (\AA^3)	W	H^1	FI	TETS2
(S)-Glutamic acid	147.13	122.84	35.15	87.69	164.74	57.38	107.36	317.5	0.72	0.9	1.04	9.57	5.06	3.13	2.45	2.44	136	1	3.78	2.94
(S)-Quisqualic acid	189.13	139.67	53.15	86.52	183.63	80.85	102.78	347.06	0.71	0.87	0.99	9.7	4.77	3.07	2.5	3.27	255	1.11	2.8	8.44
(S)-Ibotenic acid	158.11	120.08	42.36	77.72	159.51	64.92	94.59	316.81	0.74	0.91	1.03	9.28	5.23	3.27	2.94	1.77	155	1.04	2.1	5.37
(1S,3R)-ACPD	173.17	146.6	35.24	111.36	188.04	57.09	130.95	342.98	0.71	0.89	1.04	10.24	4.35	3.6	2.78	2.27	190	1.08	2.27	5.32
3,5-DHPG	183.16	150.8	34.75	116.05	191.83	54.4	137.43	357.59	0.71	0.88	0.98	9.87	5.61	5.12	1.65	2.41	242	0.98	2.74	5.49
L-CCG-I	159.14	131.75	35.1	96.65	177.36	57.74	119.62	332.26	0.71	0.88	1.08	10.31	4.46	3.93	2.77	2.02	162	1.04	2.03	5.23
ABHXD-1	185.18	154.39	34.99	119.4	190.48	55.66	134.82	346.17	0.73	0.91	1.1	10.73	4.19	3.8	2.91	3.02	223	1.11	1.61	7.27
LY341495	353.37	308.42	39.71	268.71	352.81	63.09	289.72	542.91	0.63	0.76	0.83	12.09	8.77	7.36	2.39	2.31	1500	1.28	12.02	30.9
CBPG	185.18	157.41	35.4	122.01	202.28	57.56	144.72	361.97	0.7	0.87	1.08	11.01	5.05	3.12	3.02	2.88	214	1	1.47	7.83
LY367385	209.2	178.36	35.4	142.96	221.68	56.62	165.06	391.44	0.69	0.86	1.02	11.1	5.99	4.46	2.75	2.1	370	1.18	3.31	7.66
LY393675	383.46	332.09	48.51	283.58	367.27	71.12	296.15	562.7	0.63	0.79	0.94	13.6	7.02	4.17	3.14	4.97	1704	1.28	12.15	30.85
(S)-4CPG	195.17	161.94	35.34	126.6	203.71	56.33	147.38	375.06	0.7	0.89	1.11	11.23	5.11	4	3.39	1.54	320	1.06	3.08	7
(S)-4C3HPG	211.17	168.1	41.42	126.68	212.18	66.19	145.99	385.95	0.69	0.88	1.12	11.62	5.5	4.74	3.09	1.44	372	1.18	3.28	7.74

However, maximum discrimination between agonists and antagonists at mGluR1 is found in the nonpolar area, where the smaller antagonist is 7% larger than the largest agonist, or 10 \AA^2 . Figure 3 shows plots of the nonpolar area for each selected mGluR1 ligand plotted against other noncorrelated properties. The results suggest that there is an additional hydrophobic contact taking place in the case of antagonists. Antagonists may be filling at least in part a hydrophobic pocket present in the active site and the most active antagonists have significantly larger nonpolar areas than the remaining compounds. Indeed, the three most potent antagonists have nonpolar areas 30 \AA^2 larger than any of the other antagonists. The results suggest that increasing the nonpolar surface area may increase the antagonistic character of a compound.

In summary, the three requirements for antagonism at mGluR1 are: (a) the presence of an amino group, a proximal acidic group, and a distal proton-accepting center in the geometric arrangement corresponding to the extended conformation of glutamate; (b) values of total volume with a lower bound of 150 \AA^3 and (c) a larger nonpolar area accessible for interaction with the receptor. However, our analysis provides further insight because it clearly indicates that the increase of volume is due to the presence of a larger hydrophobic area for potent antagonists compared to the agonists.

Conclusions

The work carried out relies on a relatively small set of compounds, due to the lack of high affinity and activation data for compounds at the mGluR1 subtype. The results reported in the present work provide new hypotheses into the only requirements that are necessary for antagonistic activity of the mGluR1 subtype. Because inhibition of mGluR1 was recently suggested to prevent neurological conditions such as stroke, epilepsy, and schizophrenia, identification of the molecular requirements for antagonism at mGluR1 can contribute to the discovery of novel effective drugs acting at this receptor subtype. Specifically, the presence of an amino group, a proximal carboxylic group, and a distal proton-accepting center in a specific geometric arrangement and total volume values of the molecules more than 150 \AA^3 are suggested for antagonism at mGluR1. In addition, larger nonpolar areas accessible for interaction with the receptor are requested for potent antagonism at mGluR1. Finally, other aspects of the molecules, not previously analyzed, do

TABLE V.
Numerical Values of the Electrostatic and Thermodynamic Molecular Descriptors Calculated for all Selected Agonists and Antagonists at mGluR1.

Compound	HBD	HBA	HOMO	LUMO	Hyd	ΔG_{solV} (kcal/mol)	S_{rot} (cal/k/mol)	H_{tot} (cal/mol)	C (cal/k/mol)	S_{tot} (cal/k/mol)
(S)-Glutamic acid	3	4	-10.51	0.76	-2.46	-18.88	30.411	7929.56	40.3	111.17
(S)-Quisqualic acid	3	5	-10.67	-0.31	-3.57	-22.58	36.08	8632.12	45.53	119.78
(S)-Ibotenic acid	3	4	-10.27	-0.67	-2.75	-19.34	30.36	7344.84	38.52	103.25
(1S,3R)-ACPD	3	4	-10.48	0.73	-2.27	-16.65	30.93	8503.88	45.62	115.54
3,5-DHPG	4	4	-9.22	-0.06	-1.5	-21.09	31.59	8866.24	48.48	112.86
L-CCG-I	3	4	-10.58	0.46	-1.95	-18.46	30.72	8100.87	42.36	109.87
ABHxD-I	3	4	-10.33	0.63	-1.99	-16.9	31.17	8384.63	47.27	110.17
LY341495	3	5	-8.94	0.05	0.57	-19.74	35.33	15,369.36	89.74	163.01
CBPG	3	4	-10.45	0.68	-1.54	-18.08	31.48	9095.34	49.45	120.14
LY367385	3	4	-9.96	-0.82	-0.96	-17.43	32.44	10,101.68	55.34	123.02
LY393675	3	4	-8.12	0.04	1.14	-17.07	35.71	16,086.09	95.04	167.08
(S)-4CPG	3	4	-10.28	-0.87	-1.18	-18.45	32.06	9131.31	49.64	116.91
(S)-4C3HPG	4	5	-9.73	-0.84	-1.86	-23.98	32.43	10,041.57	54.1	126.51

not appear to be relevant for ligand discrimination at mGluR1. Taken together, these results can serve as a guide to the identification of compounds for screening and the indirect design of novel drugs acting at mGluR1 useful in the treatment of serious neurological disorders.

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