Pharmacology of mavoglurant, a metabotropic glutamate receptor 5 negative allosteric modulator

Kirstie A. Bennett, John A. Christopher, Alastair JH. Brown, Fiona H. Marshall

Heptares Therapeutics Ltd., BioPark, Broadwater Road, Welwyn Garden City, Hertfordshire, AL7 3AX, UK
Kirstie.Bennett@heptares.com www.heptares.com

Introduction

Metabotropic glutamate receptors (mGLuRs) are expressed throughout the central nervous system where they act to modulate neurotransmission.

Mavoglurant is a negative allosteric modulator (NAM) of the mGlur5 receptor which is in phase III trials for the treatment of Fragile X syndrome.1

Here we characterise the binding (in vitro and ex vivo) of mavoglurant.

Methods

Radioligand binding:

To measure mavoglurant affinity in vitro, HEK293 cell membranes transiently expressing human mGlur5 (2.5 µg per well) were incubated (1 h at 25 °C) in assay buffer (50 mM HEPES (pH 7.4), 150 mM NaCl) with ~1.5 nM of [3H]M-MPEP, cold mavoglurant (0.5 nM - 10 µM) and 0.1 mM MPEP to define non-specific binding. Reactions were terminated by rapid filtration through GF/B filter plates pre-soaked with 0.1% polyethyleneimine. Plates were washed 5 x 0.5 mL in water, dried and bound radioactivity measured using scintillation spectroscopy. Inhibition curves were fitted to a four-parameter logistic equation to determine IC50 values which were converted to KI values using Kd of [3H]M-MPEP (0.86 nM) determined by saturation binding and the [3H]M-MPEP concentration (~1 nM).

Pharmacokinetics (PK):

To measure exposure of mavoglurant, PK was performed in male Sprague-Dawley (SD) rats (220-250g) following a 2 mg/kg po and a 1 mg/kg iv dose (vehicle 10 % DMAC, 10 % solutol HS15 + 80 % saline) (ChemPartner, Shanghai, China).

Ex vivo receptor occupancy:

Experiments were performed at RenaSci (Nottingham, UK). Briefly, SD rats (250-300 g) were dosed with vehicle (10 % DMAC, 10 % solutol HS15 + 80 % saline) or mavoglurant (3, 10 or 30 mg/kg) po. After 1 h rats were culled and whole brains removed and rapidly frozen. Three adjacent slices were cut from the CA3 region of the hippocampus. Two slices were incubated (10 min; 25 °C) in buffer (50 mM HEPES, 150 mM NaCl, pH 7.4) containing 2 nM [3H]M-MPEP and the third slice incubated as before with 10 µM fenobam included to define non-specific binding. Slices were washed 4 x 5 min (4°C) before radioactivity bound was determined using a β-imager (16 h). Brain and plasma exposure was determined.

Results

Figure 1: Binding of mavoglurant to mGlur5 in vitro. Graph shows mean ± S.E.M from n=6 combined.

Mavoglurant was able to fully displace [3H]M-MPEP.

Affinity (pK, ± S.D) was measured as 7.97 ± 0.25 (n=93).

Table 1: Predicted receptor occupancy of mavoglurant.

<table>
<thead>
<tr>
<th>Dose (mg/kg)</th>
<th>Estimated [CSF]* (nM)</th>
<th>Predicted RO†</th>
</tr>
</thead>
<tbody>
<tr>
<td>3</td>
<td>19.59</td>
<td>65 %</td>
</tr>
<tr>
<td>10</td>
<td>65.30</td>
<td>86 %</td>
</tr>
<tr>
<td>30</td>
<td>195.90</td>
<td>95 %</td>
</tr>
</tbody>
</table>

*Estimated [CSF] (nM) = (dose mg/kg / 2)*13.06
% RO = (conc. nM/conc. nM + KI)*100

Table 2: Measured brain and plasma concentrations and receptor occupancy of mavoglurant following a 3, 10 and 30 mg/kg dose po.

<table>
<thead>
<tr>
<th>Dose (mg/kg)</th>
<th>[Brain] (µM) ± S.E.M</th>
<th>[Plasma] (µM) ± S.E.M</th>
<th>%RO (mean ± S.E.M)</th>
</tr>
</thead>
<tbody>
<tr>
<td>3</td>
<td>0.64 ± 0.11</td>
<td>0.40 ± 0.09</td>
<td>45 ± 4</td>
</tr>
<tr>
<td>10</td>
<td>2.01 ± 0.25</td>
<td>1.46 ± 0.25</td>
<td>73 ± 2</td>
</tr>
<tr>
<td>30</td>
<td>4.72 ± 1.33</td>
<td>3.14 ± 0.86</td>
<td>83 ± 3</td>
</tr>
</tbody>
</table>

Table 2: Measured brain and plasma concentrations and receptor occupancy of mavoglurant following a 3, 10 and 30 mg/kg dose po.

Exposure and receptor occupancy of mavoglurant increased linearly with dose.

Discussion

• Mavoglurant has high affinity for the mGlur5 receptor.
• In ex vivo binding experiments, following oral administration significant displacement of [3H]M-MPEP could be measured. Exposure and occupancy was linear across doses allowing an ED50 to be calculated.

References 1 Gomez-Mancilla et al., Expert Opin. Investig. Drugs 2014, 23, 125-134