**Selective Serotonergic Properties of Low-Dose Pipamperone May Enhance Antidepressant Effect: Preclinical Evidence**

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**Introduction**

- It is widely accepted that selective serotonin reuptake inhibitors (SSRIs) are a first-line therapy for major depressive disorder (MDD).1
- SSRIs generally have a slow onset of therapeutic effect and low rate of response and remission.2
- When response to SSRI monotherapy is unsatisfactory, one approach is to augment therapy with a second medication.3
- Drugs antagonizing 5-HT1 receptors can provide effective augmentation of SSRIs monotherapy4; the mechanism may involve inhibition of negative feedback on serotonergic neurons that occurs in response to elevated levels of synaptic 5-HT caused by SSRIs.5,6
- Combing a SSRI with a drug that leads to additional and selective inhibition of D2 and 5-HT6 receptor may generate additive and potentially synergistic antidepressant effects by restoring the balance between dopamine (DA) and 5-HT in the limbic system and cortical areas and by increasing both DA and 5-HT neurotransmission.7
- The neureceptor pipamperone is a high-affinity antagonist of 5-HT6 and D2 receptors and a low-affinity antagonist of D3 receptors,1,7,8

**Objective**

- To identify a dose of pipamperone that is high enough to have substantial 5-HT6 antagonism but low enough to have no relevant D2 antagonism

**Methods**

**Study Design**

- To identify appropriate pipamperone doses, a modified version of the amphetamine tryptamine-pipamperone (ATN-test) was done in rats.
- Pharmacokinetic and pharmacodynamic modeling was used to predict the binding of pipamperone at relevant plasma concentrations to 5-HT6, D2, α1, 5-HT2A, and H1 receptors in patients.
- Protocols were reviewed and approved by the Institutional Review Board and complied with the Declaration of Helsinki.

**ATN-Test**

- Male Wiga Wistar rats (Charles River, Germany; n=5 per dose) were injected with pipamperone (0.04, 0.08, 0.16, 0.31, 0.63, 1.25, 2.5, 5.0, or 10 mg/kg subcutaneous) or control (saline solution) and challenged with apomorphine (1.0 mg/kg intravenous [IV]) at 30 minutes, tryptamine (25 mg/kg IV) at 90 minutes, and norepinephrine (1.25 mg/ml IV) at 120 minutes.
- Behavioral and physiologic effects on dopamine, 5-HT, and norepinephrine neurotransmitter systems were scored.
- After apomorphine challenge: stereotypy (0 = absent, 1 = slight, 2 = moderate, 3 = pronounced), palpebral opening (1 = one quarter open, 2 = half open, 3 = three quarters open, 4 = wide open, 5 = exophthalmos).
- After tryptamine challenge: bilateral clonic seizures of the forepaws (0 = absent, 1 = slight, 2 = moderate, 3 = pronounced), palpebral opening (1–5), and hyperemia and cyanosis of the ears (0 = absent, 1 = slight, 2 = moderate, 3 = pronounced).
- After norepinephrine challenge: norepinephrine-receptor-mediated mortality at 15 minutes (yes/no).

**Pharmacokinetic/Pharmacodynamic Modeling**

- In vivo Kp values for pipamperone binding to the 5-HT6, D2, α1, 5-HT2A, and H1 receptors after twice-daily (BID) dosing were predicted from in vitro Kp values obtained from literature and corrected based on results of a plasma protein-binding study.
- Assuming sigmoidal binding (using predicted in vivo Kp and Hill coefficient of 1), occupancy of the 5-HT6, D2, α1, 5-HT2A, and H1 receptors after BID dosing was predicted for steady-state plasma concentrations of pipamperone that were determined based on literature values in healthy volunteers and assuming linear pharmacokinetics.

**Statistical Analysis**

- Median effective dose (ED50) and corresponding 95% confidence limits (CLs) were determined using the Spearman-Kaerber estimate with theoretical instead of empirical probabilities to allow tabulation as a function of the slope of the log dose-response curve.

**Results**

**ATN-Test**

- ED50 values for pipamperone in the ATN-test are shown in Table 1.

<table>
<thead>
<tr>
<th>Challenge</th>
<th>ED50 (95% CL), mg/kg</th>
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<tbody>
<tr>
<td>Apomorphine, 1.0 mg/kg IV</td>
<td>1.55 (0.96, 2.50)</td>
</tr>
<tr>
<td>Tryptamine, 25 mg/kg IV</td>
<td>4.40 (3.30, 8.70)</td>
</tr>
<tr>
<td>Norepinephrine, 1.25 mg/kg IV</td>
<td>0.13 (0.08, 0.21)</td>
</tr>
</tbody>
</table>
| Protection against lethality | ≥10%

**Pharmacokinetic/Pharmacodynamic Modeling**

- Table 2 shows the in vivo Kp values for the 5-HT6, D2, α1, 5-HT2A, and H1 receptors and the expected corresponding plasma concentrations of pipamperone required to achieve these values in vivo.
- These concentrations are above the Kp value for the 5-HT2A and D4 receptors (Table 2).

<table>
<thead>
<tr>
<th>Receptor</th>
<th>pKi, M</th>
</tr>
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<tbody>
<tr>
<td>SHT6</td>
<td>8.2</td>
</tr>
<tr>
<td>D2</td>
<td>8.0</td>
</tr>
<tr>
<td>α1</td>
<td>6.7</td>
</tr>
<tr>
<td>5-HT2A</td>
<td>6.9</td>
</tr>
<tr>
<td>H1</td>
<td>5.7</td>
</tr>
</tbody>
</table>

**Conclusions**

- At very low doses (ED50, 0.13–0.34 mg/kg), pipamperone inhibited tryptamine-induced cyanosis and bilateral convulsions in rats, indicative of blockade of peripheral and central 5-HT6 receptors, respectively.
- Inhibition of apomorphine-induced stereotypy, a D2 receptor–mediated effect, occurred only at higher doses (ED50, 1.55 mg/kg).
- Addition of low-dose pipamperone to SSRI monotherapy may improve antidepressant efficacy and accelerate symptom resolution in patients with MDD via selective 5-HT6 receptor antagonism.
- Pharmacokinetic/pharmacodynamic modeling predicted an optimal pipamperone dose between 2.5 and 7.5 mg BID in patients for effectively blocking 5-HT6 and D2 receptors without significantly affecting D3 receptors. This dose is well below clinical doses presently used in EU countries11; pipamperone is not approved in the United States.

**References**


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