PROTECTIVE EFFECT OF ST-93 AGAINST OUABAIN INDUCED ARRHYTHMIAS IN GUINEA PIGS¹, ²

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Summary: ST-93, a clonidine analog, was studied for its antiarrhythmic activity in anaesthetised guinea pigs against ouabain induced arrhythmia. The amount of ouabain required (µg/kg) for the production of ventricular premature beat, ventricular fibrillation and cardiac arrest was recorded in control and drug treated group of animals. Both ST-93 and clonidine produced significant antiarrhythmic effect in guinea pigs. This protective effect was significantly blocked by yohimbine, suggesting that the antiarrhythmic effect is mediated through presynaptic α₂-adrenoceptors.

Key words: ST-93 ouabain arrhythmia guinea pig

INTRODUCTION

In the recent past a great number of clonidine analogs have been synthesised and tested for various pharmacodynamic activities. Tolonidine (ST 375), ST 93 and ST 363 have been reported to produce hypotension and bradycardia (3). Alpha-adrenoceptor effects of flutonidine, ST-93, tolonidine and alinidine (ST 567) on various tissues have been recently reported (4,5,9). Post synaptic and presynaptic α₂-adrenoceptor effects of clonidine are well known. Clonidine has been classified as selective alpha-adrenoceptor agonist (7), and due to this effect, reduced the arrhythmogenic effects of ouabain in guinea pigs (8). Furthermore, alinidine has also been shown to abolish certain experimental arrhythmias in dogs (1,2).

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In view of the above reports on newer imidazolidine analogs, the present study was aimed to observe the effect of ST 93 (2, chloro, 6-methyl, phenyliminoimidazolidine, hydrochloride) against ouabain induced-arrhythmia in guinea pigs.

**MATERIAL AND METHODS**

The method for production of arrhythmia used was as described by Thomas and Tripathi (11). Albino guinea pigs of either sex (350-450 g) were anaesthetized with pentobarbitone sodium (60 mg/kg, ip). Positive pressure artificial respiration was maintained throughout the experiment by means of a rodent ventilator at a rate of 45 min and stroke volume, (1 ml/100 g). The right jugular vein was cannulated for infusion of ouabain and injection of drugs. Limb lead II ECG was recorded on a Grass polygraph and heart rate was monitored from the ECG signals. Blood pressure was recorded from left carotid artery with Gould Statham pressure transducer. Ouabain solution (80 μg/ml) was continuously infused at the rate of 100 μg/min. Amount of ouabain required (per kg body weight) for the occurrence of ventricular premature beats (VPB), ventricular fibrillation (VF) and cardiac arrest (CA) was determined in control (normal saline) and drug treated animals.

**Drug treatment**: Group I animals were given clonidine (2.5, 5 and 10 μg/kg, iv) 10 min before the ouabain infusion. Group II animals were given ST 93 (2.5, 5 and 10 μg/kg, iv) 10 min before the ouabain infusion. Group III animals were pre-treated with yohimbine (2 mg/kg, iv) 15 min before ouabain infusion. Group IV and V animals were pretreated with yohimbine (2 mg/kg, iv) and 15 min later either clonidine (5 μg/kg) or ST 93 (5 μg/kg) was administered; ouabain infusion was started 10 min after the last treatment. All control animals received equivalent volume of normal saline and 10 min later, ouabain infusion was started.

Blood pressure and heart rate were recorded immediately before a drug treatment and 15 min after the administration of drugs.

**Drugs used**: Clonidine (Cipla, India), ST 93 (Boehringer Ingelheim, W. Germany), Yohimbine Hcl and ouabain (Sigma) were dissolved in normal saline prior to use.

**RESULTS**

**Haemodynamic effects**: The initial mean arterial blood pressure and heart rate of control animals were 51.9 ± 2.4 mm Hg and 242.5 ± 8.3 beats/min, respectively. Infusion of ouabain induced a significant increase in the heart rate and blood pressure. Clonidine and ST 93 (5 mg/kg) significantly decreased the heart rate (Table I). Yohimbine also produced similar depressant effects on heart rate. The doses of ouabain required for the occurrence of VPB, VF and CA were significantly lower in treatment compared to control animals. The protective effects of both clonidine and ST 93 were significantly greater than control animals. ST-93 was highly protective against ouabain induced arrhythmia in guinea pigs. ST-93 exhibits a selectivity toward α2-adrenoceptors in the heart, since the doses of ouabain required for the occurrence of arrhythmia were significantly increased in both clonidine and ST 93 treated animals.

Interaction between clonidine and ST 93 (2.5, 5 and 10 mg/kg) on the doses of ouabain required for the occurrence of arrhythmia were also significant. However, there was no interaction between clonidine and ST 93 in the doses of ouabain required for the occurrence of arrhythmia.

The protective effects of clonidine and ST 93 are in agreement with the selective α2-adrenoceptor blocking actions of these compounds, as previously reported. The protective effect of ST-93, which is a selective α2-adrenoceptor agonist, is also reported to be greater than that of clonidine. Results obtained in the present study provide evidence that ST-93, a selective α2-adrenoceptor agonist, is protective against ouabain-induced arrhythmia in guinea pigs.
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In the present study, the effects of ouabain-induced arrhythmias were assessed in guinea pigs. Ouabain was administered in a control solution (80 μg/ml) and in a drug solution (ST-93, clonidine). The doses of ouabain required for the production of ventricular arrhythmias (VPB, VF, and cardiac arrest) were significantly increased by pretreatment with clonidine and ST-93 (5 μg/kg, iv). However, the doses of ouabain required for VPB, VF, and cardiac arrest were not significantly different between 5 and 10 μg/kg doses of both clonidine and ST-93.

Interaction between ouabain and alpha-adrenoceptor stimulation: Both clonidine and ST-93 (5 μg/kg, iv) produced a significant increase (P<0.001) in the doses of ouabain required for the production of VPB, VF, and cardiac arrest. Yohimbine (2 mg/kg) pretreatment significantly antagonized the effects of clonidine and ST-93 on ouabain dose requirements. Yohimbine per se did not alter ouabain dose requirements for the occurrence of VPB, VF, and cardiac arrest.

**DISCUSSION**

Both ST-93 and clonidine induced significant hypotension and bradycardia in control animals. ST-93 has been reported to produce hypotension and bradycardia in cats. The selective alpha2-adrenoceptor antagonist yohimbine had no such effects. However, it significantly inhibited the hypotension and bradycardia induced by ST-93. The data suggest that ST-93-induced bradycardia in guinea pigs could be mediated through alpha2-adrenoceptors. The post synaptic and presynaptic alpha2-adrenoceptor activity of ST-93 is reported earlier.

The protective effect of ST-93 and clonidine against ouabain-induced arrhythmia was evident, but did not increase beyond dose of 5 μg/kg. The results obtained with clonidine are in agreement with the findings of Lechat and Schmitt (8). ST-567, a clonidine analog, is also reported to abolish certain experimental cardiac arrhythmias (1, 2, 6).

Results obtained with yohimbine suggest that ST-93-induced protective effect against ouabain-induced arrhythmia is mediated through stimulation of presynaptic alpha2-adrenoceptors. The activation of central alpha2-adrenoceptors by ST-93, probably decreases the sympathetic tone, thereby affording protection against ouabain arrhythmia.
TABLE I: Haemodynamic effects of clonidine, ST-93 and yohimbine interactions on anaesthetised guinea pigs. Values are presented as Mean ± SEM.

<table>
<thead>
<tr>
<th>Treatment (dose, iv)</th>
<th>n</th>
<th>Blood pressure mmHg.</th>
<th>Heart rate beat/min</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Initial</td>
<td>After ouabain</td>
</tr>
<tr>
<td>Control (Saline)</td>
<td>15</td>
<td>51.9±2.5</td>
<td>51.8±2.3</td>
</tr>
<tr>
<td>Clonidine (5 µg)</td>
<td>11</td>
<td>56.2±5.1</td>
<td>35.3±3.2*</td>
</tr>
<tr>
<td>ST-93 (5 µg)</td>
<td>9</td>
<td>62.0±3.3</td>
<td>41.8±2.6**</td>
</tr>
<tr>
<td>Yohimbine (2 mg)</td>
<td>11</td>
<td>59.4±3.6</td>
<td>54.4±3.8</td>
</tr>
<tr>
<td>Yohimbine+Clonidine (2 mg)+(5 µg)</td>
<td>9</td>
<td>59.3±4.4</td>
<td>49.1±2.6*</td>
</tr>
<tr>
<td>Yohimbine+ST-93 (2 mg)+(5 µg)</td>
<td>8</td>
<td>55.1±1.6</td>
<td>48.5±1.5*</td>
</tr>
</tbody>
</table>

* P<0.01
** P<0.001 (unpaired t' test)

TABLE II: Effect of clonidine and ST-93 and their interaction with yohimbine on ouabain induced arrhythmia in guinea pigs. Values presented are as Mean ± SEM of doses of ouabain (µg/kg) required to produce ventricular premature beat (VPB), ventricular fibrillation (VF) and cardiac arrest (CA).
<table>
<thead>
<tr>
<th>Treatment (dose, iv)</th>
<th>n</th>
<th>VPB</th>
<th>VF</th>
<th>CA</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (Saline)</td>
<td>25</td>
<td>172.80±3.80</td>
<td>186.70±4.20</td>
<td>257.10±4.65</td>
</tr>
<tr>
<td>Clonidine 2.5</td>
<td>9</td>
<td>246.16±5.17***</td>
<td>338.66±19.24***</td>
<td>415.45±19.10***</td>
</tr>
<tr>
<td></td>
<td>5.0</td>
<td>287.57±13.70***</td>
<td>361.30±21.07***</td>
<td>435.30±18.40***</td>
</tr>
<tr>
<td></td>
<td>10.0</td>
<td>307.27±18.9***</td>
<td>377.80±24.58***</td>
<td>453.80±18.04***</td>
</tr>
<tr>
<td>ST-93 2.5</td>
<td>9</td>
<td>223.65±13.96***</td>
<td>287.50±17.50**</td>
<td>372.80±19.80*</td>
</tr>
<tr>
<td></td>
<td>5.0</td>
<td>248.03±5.80**</td>
<td>307.57±12.12***</td>
<td>374.35±7.01***</td>
</tr>
<tr>
<td></td>
<td>10.0</td>
<td>253.92±7.41***</td>
<td>322.54±10.28***</td>
<td>414.46±12.84***</td>
</tr>
<tr>
<td>Yohimbine (2 mg)</td>
<td>11</td>
<td>174.38±6.71</td>
<td>208.33±6.84</td>
<td>265.60±6.10</td>
</tr>
<tr>
<td>Yohimbine + Clonidine (2 mg+5 μg)</td>
<td>10</td>
<td>204.42±8.08b</td>
<td>242.35±13.63a</td>
<td>304.04±10.50a</td>
</tr>
<tr>
<td>Yohimbine + ST-93 (2 mg+5 μg)</td>
<td>8</td>
<td>197.43±12.79b</td>
<td>229.96±13.52b</td>
<td>274.93±14.80b</td>
</tr>
</tbody>
</table>

*P<0.05
bP<0.001 vs ST-93 (5 μg)

**P<0.01

***P<0.001 vs Clonidine (5 μg)

(unpaired 't' test)
ACKNOWLEDGEMENTS

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