H₂ RECEPTOR ACTIVITY; EFFECT IN ISOLATED RABBIT HEART

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ABSTRACT... Histamine can stimulate the heart by directly interacting with cardiac histamine receptors. In the present study we have investigated the H₂ receptor activity in isolated rabbit heart. Cimetidine, a specific H₂ receptor antagonist was used. The isolated heart was mounted in Langendorff apparatus. The heart was perfused at a constant pressure with oxygenated Ringer’s Locke solution. H₂ receptor antagonist produces negative inotropic effect in the presence of histamine. This indicates that H₂ receptors are present in rabbit heart, and plays a role in mediation of positive inotropic effect produced through CAMP by histamine.

Key words: Histamine. H₂ antagonist. Cimetidine.

INTRODUCTION
The presence of histamine in large quantities in cardiac tissue is well documented⁴. The cardiovascular actions of histamine have been attributed to the activation of two different types of histamine receptor classified as histamine type H₁ receptors and type H₂ receptors²,³.

It has been observed that histamine and its analogue posses a direct stimulatory effect on heart. Such compounds also increase the activity of cardiac adenylate cyclase and cardiac phosphorylase as well as increasing the adenosine 3’5’ monophosphate⁴,⁵,⁶,⁷. Contraction or force development by smooth muscle cells depends by the elevation of Intracellular calcium in the myoplasm. This is caused by either release of I/C calcium from the storage sites like mitochondria, or entry of calcium via receptor operated channels⁸.

METHOD
In this study we used rabbits having weight of 0.75 to 1.5kg of either sex. In our vitro project Ringer Locke physiological nutrient solution was used for retrograde perfusion to the isolated rabbit heart⁹. The composition of Ringer Locke solution was NaCl, 45g; NaHCO₃, 1g; C₆H₁₂O₆, 5G; KCl 2.1G; CaCl₂, 1.6g; and H₂O, 5000ml.

Preparation and isolation of heart was based on Langendorff methods, described by kitchen¹⁰, 1984, and Burn¹¹, 1952.

For the preparation of isolated heart we first injected 0.5cc or 2500 IU of heparin intravenously and waited for 3-5 minutes. The rabbit was then sacrificed by cutting the neck with a sharp surgical knife. The chest of animal was opened and heart with at least 1cm of aorta was removed and placed in petri dish, which already contained the oxygenated Ringer Locke solution at room temperature. Heart was squeezed several times gently, to remove blood. Surrounding tissues of the heart were removed. Aorta was tied with steel cannula fixed with Langendorff apparatus⁵.

Heart was coated with liquid paraffin to prevent drying¹². Thread was attached to the tip of ventricle by heart clip and other end of thread was tied with transducer after passing the thread through two pullies. Transducer was connected with 7B Grass polygraph machine, which recorded the isolated heart activity on polygraph paper. Heart was perfused with oxygenated Ringer Locke solution and allowed to equilibrate 30-45 minutes¹⁰. Drugs were administered through the butterfly needle, which was connected with rubber tube near the
aorta. The volume of all injections were kept constant at 0.2ml; intervals of 10-29 minutes were allowed between successive injections.

RESULTS
As per protocol the tissue was prepared and EC50 was evaluated. Five observations were taken of each dilution ranging from 104 to 108. The difference of amplitude on contractility of the isolated rabbit heart was evaluated from normal in comparison with the effect produced by individual drug.

The results were tabulated in descending order and median value was taken as EC50.

The EC50 of individual dilution was used for further observations.

The observations of five responses of EC50 of histamine on amplitude of contraction were recorded. The mean value observe 1.54mm from normal as depicted in table-I. The observation of five responses of histamine EC50 with H2 blocker (Cimetidine) were recorded as shown in table-II. the mean value of five observations of histamine compared with the mean value of five observation of histamine in the presence of H2 blocker were compared as shown in table II. The difference showed a decrease from 1.54 to -4.11mm. This means that H2 blocker produces a negative inotropic effect in the presence of histamine.

<table>
<thead>
<tr>
<th>S/No</th>
<th>BD</th>
<th>AD</th>
<th>Diff</th>
<th>%Percent Diff</th>
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<td>1</td>
<td>21.16</td>
<td>23.05</td>
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<tr>
<td>2</td>
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<td>32.72</td>
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<td>56.56</td>
<td>57.69</td>
<td>1.13</td>
<td>1.9</td>
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<td>36.58</td>
<td>38.06</td>
<td>1.54</td>
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Table-I. The mean value of five observations of histamine

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<td>5</td>
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<td>Mean</td>
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Table-II. The mean value of five observations of histamine in presence of H2 blocker

and the effect was blocked by an H2, receptor blocking agent. This observation suggests that the inotropic effect of histamine was due to H2 receptor stimulation and that H2 receptor were associated with cardiac adenylate cyclase activity. The enzyme is activated by histamine and cAMP increase in the whole heart prior to the increase in force of contraction when histamine is injected. All effects are blocked by H2 blocker. Intracellular Ca+ is closely regulated by sodium-calcium exchanger (NCX) and Ca+ efflux is dependent on the I/C sodium (Na+) concentration and trans- sarcolemmal Na+ gradient13. Data from other observers also agree with our findings14,15. Histamine H2 receptors are pivotal in mediating the increase in contractility elicited by histamine in the mammalian heart5. First phase of histamines positive inotropic effect is due to an increase in cytosolic calcium resulting from enhanced calcium released from the sarcoplasmic reticulum promoted by inositol phosphate. Hence the two histamine receptors types coupled to distinct signal transduction pathways which co-exist in heart muscle produces positive inotropic effect16,17,18.

CONCLUSIONS
Our finding demonstrate that histamine produces positive inotropic effects. Whereas histamine in presence of H2 blocker (Cimetidine) produces a negative inotropic effect. This suggests that H2 receptors are present in rabbit heart.

REFERENCES
1. Feigen, G.A. and Prager, D.J. Experimental cardiac anaphylaxis physiologic pharmacologic and biochemical aspects of immune reactions in the


